



Positron Emission Tomography of the Brain

Edited by W.-D. Heiss and M. E. Phelps

With 108 Figures

Springer-Verlag
Berlin Heidelberg New York 1983

Professor Dr. med. Wolf-Dieter Heiss
Max-Planck-Institut für
Neurologische Forschung und Neurologische
Klinik des Krankenhauses Merheim
Ostmerheimer Straße 200
D-5000 Köln 91

Professor Dr. Michael E. Phelps
University of California
Division of Biophysics
Department of Radiological
Sciences and Laboratory
of Nuclear Medicine
Los Angeles, California 90024
USA

ISBN-13: 978-3-540-12130-5 e-ISBN-13: 978-3-642-95428-3
DOI: 10.1007/978-3-642-95428-3

Library of Congress Cataloging in Publication Data

Main entry under title:

Positron emission tomography of the brain.

Bibliography: p. Includes index. 1. Brain-Diseases-Diagnosis. 2. Brain – Radiography. 3. Tomography, Emission. 4. Positrons – Emission. I. Heiss, W.-D. (Wolf-Dieter), 1939 –. II. Phelps, M. E. (Michael E.)
RC386.6.T64P67 1983 616.8'047572 82-23600

ISBN-13: 978-3-540-12130-5

This work is subject to copyright. All rights are reserved, whether the whole or part of the material is concerned, specifically those of translation, reprinting, re-use of illustrations, broadcasting, reproduction by photocopying machine or similar means, and storage in data banks. Under § 54 of the German Copyright Law where copies are made for other than private use, a fee is payable to "Verwertungsgesellschaft Wort", Munich.

© by Springer-Verlag Berlin Heidelberg 1983

The use of general descriptive names, trade marks, etc. in this publication, even if the former are not especially identified, is not to be taken as a sign that such names, as understood by the Trade Marks and Merchandise Marks Act, may accordingly be used freely by anyone.

Product Liability: The publisher can give no guarantee for information about drug dosage and application thereof contained in this book. In every individual case the respective user must check its accuracy by consulting other pharmaceutical literature.

2125/3140-543 210

Preface

The International Symposium on Positron Emission Tomography of the Brain was held at Schloss Auel in Lohmar, near Cologne, FRG, on May 3rd to 8th, 1982. This isolated place was chosen to bring together and keep together people active in research employing PET. The participants were asked to report their ongoing work, to discuss upcoming problems, and to indicate future developments. In contrast to the large international meetings which suffer from lack of time for discussion and are affected by the distractions of major cities, our symposium provided ample opportunity for discussion during the scientific sessions and guaranteed a free exchange of ideas during the leisure time spent together. It also gave us the chance to celebrate the founding of a new Max Planck Institute for Neurological Research, whose two departments are headed by Dr. K.-A. Hossmann and myself, and to mark the start of positron emission tomography in our laboratory.

The book contains the invited papers presented at the meeting and the formal minipapers given at the final general discussion, moderated superbly by Dr. L. Sokoloff. In order to accelerate publication, the discussions of the individual papers and the final discussion have been omitted. However, every participant will remember the atmosphere and the highlights of the discussions, which gave a special character to the whole event.

The style of the symposium was made possible by a generous sponsor, the UCB Company, Kerpen, FRG, whom I would like to thank for their financial support. I also express my gratitude to Dr. W. Thiede, Mr. P. Hansen, and the staff of UCB for helping to organize and realize the scientific and social aspects of the gathering.

Köln, January 1983

W.-D. Heiss

Table of Contents

Preface (W.-D. Heiss)	V
---------------------------------	---

Principles

Biochemical Principles for the Measurement of Metabolic Rates in Vivo (L. Sokoloff and C. Smith)	2
---	---

Experimental Application of Triple-Labeled Quantitative Autoradiography for Measurement of Cerebral Blood Flow, Glucose Metabolism and Protein Biosynthesis (G. Mies, W. Bodsch, W. Paschen, and K.-A. Hossmann)	19
---	----

Methodology

New Design Concepts for Quantitive Positron Emission Computed Tomography of the Brain (E. J. Hoffman, J. B. Barton, M. E. Phelps, and S.-C. Huang)	30
--	----

Design Characteristics of a Multiring Positron Camera System for Emission Tomography of the Brain (L. Eriksson, C. Bohm, M. Bergström, K. Ericson, T. Greitz, G. Blomqvist, J. Litton, L. Widén, P. Hansen, S. Holte, and H. Stjernberg)	40
---	----

Design Concepts and Performances of HEADTOME, a Multiring Hybrid Emission Tomograph for the Brain (I. Kanno, K. Uemura, Y. Miura, and S. Miura)	46
---	----

Radiochemicals

Labeled Compounds for Positron Emission Tomography (A. P. Wolf and J. S. Fowler)	52
---	----

Biochemical Parameters in Radiopharmaceutical Design (J. R. Barrio)	65
--	----

Cerebral Blood Flow

Regional Cerebral Blood Flow Measurement and Dynamic Positron Emission Tomography (Y. L. Yamamoto, E. Meyer, D. Menon, P. Roland, and M. Diksic)	78
--	----

The Measurement of Regional Cerebral Blood Flow in Man Using an in Vivo Autoradiographic Technique and Positron Emission Tomography (A. H. Lockwood, M. D. Ginsberg, B. E. Howard, P. Freed, and W. Hassel)	85
--	----

Regional Cerebral Blood Flow Measurement with Fluoromethane and Positron Tomography (J. E. Holden, S. J. Gatley, R. J. Nickles, R. A. Koeppe, G. G. Celesia, and R. E. Polcyn)	90
---	----

A Method for Local Evaluation of the Volume of Rapidly Exchangeable Water in the Human Brain (J. C. Depresseux) . . .	95
--	----

Oxygen Consumption

Measurement of Regional Cerebral Oxygen Consumption and Blood Flow in Man (M. E. Raichle, P. Herscovitch, W. Martin, and J. Markham) . . .	104
--	-----

Clinical Application of Measurement of Regional Cerebral Blood Flow and Oxygen Utilization by Positron Emission Tomography (T. Jones, R. Frackowiak, R. Wise, and G. L. Lenzi) .	107
--	-----

Positron Emission Tomography of Stroke Patients (R. H. Ackerman, N. M. Alpert, S. M. Davis, R. E. Kelley, J. A. Correia, G. L. Brownell, and J. M. Taveras)	113
---	-----

Local Cerebral Blood Flow and Oxygen Consumption in Evolving Irreversible Ischemic Infarction (J. C. Baron, D. Rougemont, P. Lebrun-Grandié, M. G. Bousser, E. Cabanis, J. Bories, D. Comar, and P. Castaigne)	120
---	-----

Glucose Metabolism

Mapping Local Cerebral Glucose Utilization in Normal Aging and in Cerebrovascular, Degenerative, and Epileptic Disorders (D. E. Kuhl)	128
---	-----

Human Sensory Stimulation and Deprivation as Demonstrated by Positron Computed Tomography (M. E. Phelps and J. C. Mazziotta)	139
--	-----

Measurement of Local Cerebral Glucose Metabolism: Application to the Study of Stroke (M. Reivich, J. H. Greenberg, M. J. Kushner, and A. Alavi)	153
---	-----

Remote Functional Depression of Glucose Metabolism in Stroke and its Alteration by Activating Drugs (W.-D. Heiss, H. W. Ilsen, R. Wagner, G. Pawlik, and K. Wienhard)	162
--	-----

Regional Perfusion and Glucose Uptake Determination with ^{11}C -Methyl-Glucose and Dynamic Positron Emission Tomography (K. Vyska, G. Kloster, L. E. Feinendegen, W.-D. Heiss, G. Stöcklin, A. Höck, C. Freundlieb, A. Aulich, F. Schuier, H. U. Thal, V. Becker, and A. Schmid)	169
Glycolytic Rate and Histologic Grade of Human Cerebral Gliomas: A Study with [^{18}F] Fluorodeoxyglucose and Positron Emission Tomography (G. DiChiro, R. A. Brooks, L. Sokoloff, N. J. Patronas, R. L. DeLaPaz, B. H. Smith, and P. L. Kornblith)	181
Positron Emission Tomography Studies of Brain Energy Metabolism in Schizophrenia (L. Widén, M. Bergström, G. Blomqvist, C. Bohm, E. Ehrin, S. Elander, K. Ericson, L. Eriksson, T. Greitz, D. H. Ingvar, J.-E. Litton, L. Nilsson, G. Sedvall, and F. Wiesel)	192
Regional Correlations in Patterns of Glucose Use in Patients with Schizophrenia and Normal Subjects During Mild Pain Stimulation (R. M. Kessler, C. M. Clark, M. S. Buchsbaum, H. Holcomb, R. A. Margolin, J. Cappeletti, M. Channing, D. P. vanKammen, A. C. King, J. Johnson, and R. G. Manning)	196
Evaluation of Regional Glucose Metabolism with Positron Emission Tomography in Normal and Psychiatric Populations (J. D. Brodie, A. P. Wolf, N. Volkow, D. R. Christman, P. DeFina, M. DeLeon, T. Farkas, S. H. Ferris, J. S. Fowler, F. Gomez-Mont, J. Jaeger, J. A. G. Russell, R. Stamm, and Y. Yonekura)	201
Protein Synthesis and Receptor Studies	
Local Brain Protein Metabolism in Dementia and Schizophrenia: In Vivo Studies with ^{11}C -L-Methionine and Positron Emission Tomography (P. Bustany, J. F. Henry, T. Sargent, E. Zarifian, E. Cabanis, P. Collard, and D. Comar)	208
Brain Regional Pharmacokinetics of ^{11}C -Labeled Diphenylhydantoin and Pimozide in Man (J. C. Baron, D. Comar, C. Crouzel, D. Roeda, G. Mestelan, E. Zarifian, C. Munari, C. Stoffels, J. Bancaud, J. P. Chodkiewicz, H. Loo, and Y. Agid)	212
Mini-Papers	
Potential Bromine-75-Labeled Brain Radiopharmaceuticals (G. Kloster, H. Scholl, P. Laufer, W. Wutz, and G. Stöcklin)	226
Biochemical Prerequisites for Measurement of Protein Synthesis (W. Bodsch)	229

Discrepancy Between Glucose Availability and Calculated
Glucose Consumption During Acute Brain Ischemia
(G. Mies, W. Paschen, and K.-A. Hossmann) 232

The Relationship of Regional Cerebral Blood Flow, Oxygen
Metabolism and Glucose Metabolism Following Acute
Stroke (J.M. Gibbs, C.G. Rhodes, R.J.S. Wise,
R.S.J. Frackowiak, and T. Jones) 234

Subject Index 237

List of Authors

Ackerman, R. H.

Carotid Evaluation and Cerebral Blood Flow
Laboratories, Massachusetts General Hospital,
Boston, Massachusetts 02114 – USA

Agid, Y.

Hôpital la Salpêtrière,
75013 Paris – France

Alavi, A.

Cerebrovascular Research Center of the
Departments of Neurology and Radiology,
University of Pennsylvania,
36th and Hamilton Walk,
Philadelphia, Pennsylvania 19104 – USA

Alpert, N. M.

Massachusetts General Hospital,
Boston, Massachusetts 02114 – USA

Aulich, A.

Neurologische Klinik der Universität,
Moorenstr. 5,
4000 Düsseldorf – FRG

Bancaud, J.

Hôpital Sainte-Anne, Rue Cabanis,
75014 Paris – France

Baron, J. C.

Service Hospitalier Frédéric Joliot, CEA
Département de Biologie,
91406 Orsay – France

Barrio, J. R.

UCLA School of Medicine, Department of
Radiological Sciences, Division of Biophysics,
Los Angeles, California 90024 – USA

Barton, J. B.

UCLA School of Medicine, Department of
Radiological Sciences, Division of Biophysics,
Los Angeles, California 90024 – USA

Becker, V.

Kernforschungsanlage Jülich GmbH., Institut
für Medizin,
5170 Jülich – FRG

Bergström, M.

Department of Neuroradiology,
Karolinska Hospital,
10401 Stockholm – Sweden

Blomqvist, G.

Department of Neurophysiology,
Karolinska Hospital,
10401 Stockholm – Sweden

Bodsch, W.

Max-Planck-Institut für neurologische
Forschung, Abteilung für experimentelle
Neurologie, Ostmerheimer Str. 200,
5000 Köln 91 – FRG

Bohm, C.

Institute of Physics, University of Stockholm,
Vanadisvas 9,
11346 Stockholm – Sweden

Bories, J.

Hôpital la Salpêtrière, 47 Blvd. de l'Hôpital,
75013 Paris – France

Bousser, M. G.

Hôpital la Salpêtrière, 47 Blvd. de l'Hôpital,
75013 Paris – France

Brodie, J. D.

Department of Psychiatry, New York University
Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

Brooks, R. A.

Neuroradiology and Computed Tomography
Section, Clinical Center – Building 10,
National Institutes of Health,
Bethesda, Maryland 20205 – USA

Brownell, G. L.
Massachusetts General Hospital,
Boston Massachusetts 02114 – USA

Buchsbaum, M. S.
Department of Psychiatry,
University of California,
Irvine, California 92717 – USA

Bustany, P.
Service Hospitalier Frédéric Joliot,
CEA Département de Biologie,
91406 Orsay – France

Cabanis, E.
Centre National d'Ophthalmologie
des Quinze-Vingt,
75012 Paris – France

Cappeletti, J.
Biological Psychiatry Branch, National Institute
of Mental Health, 9000 Rockville Pike,
Bethesda, Maryland 20205 – USA

Castaigne, P.
Hôpital la Salpêtrière, 47 Blvd. de l'Hôpital,
75013 Paris – France

Celesia, G. G.
Departments of Medical Physics, Radiology
and Neurology, Clinical Sciences Center, University
of Wisconsin, 600 Highland Avenue,
Madison, Wisconsin 53792 – USA

Channing, M.
National Institutes of Health,
Depart. of Nuclear Medicine,
Clinical Center, 9000 Rockville Pike
Bethesda, Maryland 20205 – USA

Chodkiewicz, J. P.
Hôpital Sainte-Anne, Rue Cabanis,
75014 Paris – France

Christman, D. R.
Department of Chemistry,
Brookhaven National Laboratory
Upton, New York 11973 – USA

Clark, C. M.
National Institutes of Health,
Depart. of Nuclear Medicine,
Clinical Center, 9000 Rockville Pike,
Bethesda, Maryland 20205 – USA

Collard, P.
Service Hospitalier Frédéric Joliot,
CEA Département de Biologie
91406 Orsay – France

Comar, D.
Service Hospitalier Frédéric Joliot,
CEA Département de Biologie,
91406 Orsay – France

Correia, J. A.
Massachusetts General Hospital
Boston, Massachusetts 02114 – USA

Crouzel, C.
Service Hospitalier Frédéric Joliot,
CEA Département de Biologie,
91406 Orsay – France

Davis, S. M.
Royal Melbourne Hospital, Department of
Neurology, Grattan Street, Parkville
Victoria 3050 – Australia

DeFina, P.
Department of Psychiatry, New York University
Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

DeLaPaz, R. L.
Neuroradiology and Computed Tomography
Section, Clinical Center – Building 10,
National Institutes of Health,
Bethesda, Maryland 20205 – USA

DeLeon, M.
Department of Psychiatry, New York University
Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

Depresseux, J. C.
Unité Médicale du Cyclotron,
Université de Liège, Sart Tilman B 30,
4000 Liège 1 – Belgium

DiChiro, G.
Neuroradiology and Computed Tomography
Section, Clinical Center – Building 10, Na-
tional Institutes of Health,
Bethesda, Maryland 20205 – USA

Diksic, M.
Neuroisotope Laboratory,
Montreal Neurological Institute,
McGill University, 3801 University Street,
Montreal,
Quebec, Canada H3A 2B4

Ehrin, E.
Karolinska Hospital,
10401 Stockholm – Sweden

Elander, S.
Karolinska Hospital,
10401 Stockholm – Sweden

Ericson, K.
Department of Neuroradiology,
Karolinska Hospital,
10401 Stockholm – Sweden

Eriksson, L.
Institute of Physics, University of Stockholm,
Vanadisvas 9
11346 Stockholm – Sweden

Farkas, T.
Department of Psychiatry, New York Universi-
ty Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

Feinendegen, L. E.
Kernforschungsanlage Jülich GmbH.,
Institut für Medizin,
5170 Jülich – FRG

Ferris, S. H.
Department of Psychiatry, New York Universi-
ty Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

Fowler, J. S.
Department of Chemistry,
Brookhaven National Laboratory,
Upton, N. Y. 11973 – USA

Frackowiak, R. S. J.
MRC Cyclotron Unit,
Hammersmith Hospital, Ducane Road,
London W12 OHS – United Kingdom

Freed, P.
Department of Mathematics,
University of Miami,
Coral Gables, Florida 33124 – USA

Freundlieb, C.
Kernforschungsanlage Jülich GmbH.,
Institut für Medizin,
5170 Jülich – FRG

Gatley, S. J.
Departments of Medical Physics, Radiology
and Neurology, Clinical Sciences Center, Uni-
versity of Wisconsin, 600 Highland Avenue,
Madison, Wisconsin 53792 – USA

Gibbs, J. M.
MRC Cyclotron Unit, Hammersmith Hospital,
Ducane Road,
London W12 OHS – United Kingdom

Ginsberg, M. D.
Department of Neurology (D4–5),
University of Miami, School of Medicine,
Miami, Florida 33101 – USA

Gomez-Mont, F.
Department of Psychiatry, New York Universi-
ty Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

Greenberg, J. H.
University of Pennsylvania, Cerebrovascular
Research Center of the Department
of Neurology, 36th and Hamilton Walk,
Philadelphia, Pennsylvania 19104 – USA

Greitz, T.
Department of Neuroradiology,
Karolinska Hospital,
10401 Stockholm – Sweden

Hansen, P.
Instrument AB Scanditronics,
18303 Täbi/Stockholm – Sweden

Hassel, W.
Division of Nuclear Medicine, Mt. Sinai Medical Center, 4300 Alton Road, Miami Beach, Florida 33140 – USA

Heiss, W.-D.
Max-Planck-Institut für neurologische Forschung, Abteilung für allgemeine Neurologie, Ostmerheimer Str. 200, 5000 Köln 91 – FRG

Henry, J. F.
Hôpital Charles Foy, USN-F, 94200 Ivry – France

Herscovitch, P.
Division of Radiation Sciences, MIR Washington University School of Medicine St. Louis, Missouri 63110 – USA

Höck, A.
Kernforschungsanlage Jülich GmbH, Institut für Medizin, 5170 Jülich – FRG

Hoffman, E. J.
Division of Biophysics, Department of Radiological Sciences, UCLA School of Medicine, Los Angeles, California 90024 – USA

Holcomb, H.
Biological Psychiatry Branch, National Institute of Mental Health, 9000 Rockville Pike Bethesda, Maryland 20205 – USA

Holden, J. E.
Departments of Medical Physics, Radiology, and Neurology, Clinical Sciences Center, University of Wisconsin, 600 Highland Avenue, Madison, Wisconsin 53792 – USA

Holte, S.
Instrument AB Scanditronics, 18303 Täbi/Stockholm – Sweden

Hossmann, K.-A.
Max-Planck-Institut für neurologische Forschung, Abteilung für experimentelle Neurologie, Ostmerheimer Str. 200 5000 Köln 91 – FRG

Howard, B. E.
Department of Mathematics, University of Miami, Coral Gables, Florida 33124 – USA

Huang, S.-C.
Division of Biophysics, Department of Radiological Sciences, UCLA School of Medicine, Los Angeles, California 90024 – USA

Ilsen, H. W.
Neurologische Klinik des Krankenhauses Merheim, Ostmerheimer Str. 200, 5000 Köln 91 – FRG

Ingvar, D. H.
University of Lund, Department of Clinical Neurophysiology, University Hospital, 22185 Lund 5 – Sweden

Jaeger, J.
Department of Psychiatry, New York University Medical Center, 550 First Avenue New York, N. Y. 10016 – USA

Johnson, J.
Biological Psychiatry Branch, National Institute of Health, 9000 Rockville Pike, Bethesda, Maryland 20205 – USA

Jones, T.
MRC Cyclotron Unit and Department of Medicine, Hammersmith Hospital, Ducane Road, London W12 OHS – United Kingdom

Kanno, I.
Division of Radiology and Nuclear Medicine, Research Institute of Brain and Blood Vessels, 6-10 Senshu kubota machi, Akita 010 – Japan

Kelley, R. E.
Department of Neurology, Temple University Hospital, 3401 N. Broad Street, Philadelphia, Pennsylvania 19140 – USA

Kessler, R. M.
National Institutes of Health, Department of Nuclear Medicine, Clinical Center, 9000 Rockville Pike Bethesda, Maryland 20205 – USA

King, A. C.
Biological Psychiatry Branch National Institute of Mental Health, 9000 Rockville Pike
Bethesda, Maryland 20205 – USA

Kloster, G.
Kernforschungsanlage Jülich GmbH,
Institut für Chemie 1 (Nuklearchemie),
5170 Jülich – FRG

Koeppel, R. A.
Departments of Medical Physics, Radiology,
and Neurology, Clinical Sciences Center, University of Wisconsin, 600 Highland Avenue,
Madison, Wisconsin 53792 – USA

Kornblith, P. L.
Surgical Neurology Branch, Clinical Center –
Building 10A, National Institutes of Health,
Bethesda, Maryland 20205 – USA

Kuhl, D. E.
Laboratory of Nuclear Medicine and Department of Radiological Sciences, UCLA School of Medicine,
Los Angeles, California 90024 – USA

Kushner, M. J.
University of Pennsylvania, Cerebrovascular Research Center of the Department of Neurology, 36th and Hamilton Walk,
Philadelphia, Pennsylvania 19104 – USA

Laufer, P.
Kernforschungsanlage Jülich GmbH,
Institut für Chemie 1 (Nuklearchemie),
5170 Jülich – FRG

Lebrun-Grandié, P.
Service Hospitalier Frédéric Joliot,
CEA Departement de Biologie,
91406 Orsay – France

Lenzi, G. L.
MRC Cyclotron Unit and Department of Medicine, Hammersmith Hospital,
Ducane Road,
London W12 0HS – United Kingdom

Litton, J.-E.
Department of Neurophysiology,
Karolinska Hospital,
10401 Stockholm – Sweden

Lockwood, A. H.
Department of Neurology (D4–5),
University of Miami School of Medicine,
Miami, Florida 33101 – USA

Loo, H.
Hôpital Sainte-Anne, Rue Cabanis,
75014 Paris – France

Manning, R. G.
National Institutes of Health, Department of Nuclear Medicine, Clinical Center,
9000 Rockville Pike
Bethesda, Maryland 20205 – USA

Margolin, R. A.
National Institutes of Health, Department of Nuclear Medicine, Clinical Center,
9000 Rockville Pike
Bethesda, Maryland 20205 – USA

Markham, J.
Division of Radiation Sciences,
MIR Washington University
School of Medicine
St. Louis, Missouri 63110 – USA

Martin, W.
Division of Radiation Sciences,
MIR Washington University
School of Medicine
St. Louis, Missouri 63110 – USA

Mazziotta, J. C.
UCLA School of Medicine, Department of Neurology, Laboratory of Biomedical and Environmental Sciences,
Los Angeles, California 90024 – USA

Menon, D.
Neuroisotope Laboratory, Montreal Neurological Institute, McGill University,
3801 University Street,
Montreal, Quebec, Canada H3A 2B4

Mestelan, G.
Service Hospitalier Frédéric Joliot,
CEA Departement de Biologie
91406 Orsay – France

Meyer, E.
Neuroisotope Laboratory, Montreal Neurological Institute, McGill University,
3801 University Street,
Montreal, Quebec, Canada H3A 2B4

Mies, G.
Max-Planck-Institut für neurologische
Forschung, Abteilung für experimentelle
Neurologie, Ostmerheimer Str. 200,
5000 Köln 91 – FRG

Miura, S.
Division of Radiology and Nuclear Medicine,
Research Institute of Brain and Blood Vessels,
6–10 Senshu kubota machi,
Akita 010 – Japan

Miura, Y.
Division of Radiology and Nuclear Medicine,
Research Institute of Brain and Blood Vessels,
6–10 Senshu kubota machi,
Akita 010 – Japan

Munari, C.
Hôpital Sainte-Anne, Rue Cabanis,
75014 Paris – France

Nickles, R.J.
Departments of Medical Physics, Radiology,
and Neurology, Clinical Sciences Center,
University of Wisconsin, 600 Highland Avenue,
Madison, Wisconsin 53792 – USA

Nilsson, L.
Karolinska Hospital,
10401 Stockholm – Sweden

Paschen, W.
Max-Planck-Institut für neurologische
Forschung, Abteilung für experimentelle
Neurologie, Ostmerheimer Str. 200,
5000 Köln 91 – FRG

Patronas, N.J.
Neuroradiology and Computed Tomography
Section, Clinical Center – Building 10,
National Institutes of Health,
Bethesda, Maryland 20205 – USA

Pawlik, G.
Neurologische Klinik des Krankenhauses
Merheim, Ostmerheimer Str. 200,
5000 Köln 91 – FRG

Phelps, M.E.
UCLA School of Medicine, Department of
Radiological Sciences, Division of Biophysics,
Los Angeles, California 90024 – USA

Polcyn, R.E.
Departments of Medical Physics, Radiology,
and Neurology, Clinical Sciences Center,
University of Wisconsin, 600 Highland
Avenue,
Madison, Wisconsin 53792 – USA

Raichle, M.E.
Division of Radiation Sciences, The Edward
Mallinckrodt Institute of Radiology,
Washington University School of Medicine,
St. Louis, Missouri 63110 – USA

Reivich, M.
University of Pennsylvania, Room 429, John-
son Pavilion/G2, 36th and Hamilton Walk,
Philadelphia, Pennsylvania 19104 – USA

Rhodes, C.G.
MRC Cyclotron Unit, Hammersmith Hospital,
Ducane Road,
London W12 OHS – United Kingdom

Roeda, D.
Service Hospitalier Frédéric Joliot,
CEA Departement de Biologie,
91406 Orsay – France

Roland, P.
Neuroisotope Laboratory, Montreal Neurological Institute, McGill University,
3801 University Street,
Montreal, Quebec, Canada H3A 2B4

Rougemont, D.
Service Hospitalier Frédéric Joliot,
CEA Departement de Biologie
91406 Orsay – France

Russell, J. A. G.
Department of Chemistry,
Brookhaven National Laboratory,
Upton, N. Y. 11973 – USA

Sargent, T.
Donner Laboratory, University of California,
Berkeley, California 94720 – USA

Schmid, A.
Kernforschungsanlage Jülich GmbH.,
Institut für Medizin,
5170 Jülich – FRG

Scholl, H.
Kernforschungsanlage Jülich GmbH.,
Institut für Chemie 1 (Nuklearchemie),
5170 Jülich – FRG

Schuijer, F.
Neurologische Klinik der Universität,
Moorenstr. 5,
4000 Düsseldorf – FRG

Sedvall, G.
Karolinska Hospital,
10401 Stockholm – Sweden

Smith, B. H.
Surgical Neurology Branch, Clinical Center –
Building 10A, National Institutes of Health,
Bethesda, Maryland 20205 – USA

Smith, C. B.
Laboratory of Cerebral Metabolism,
National Institute of Mental Health,
Bethesda, Maryland 20205 – USA

Sokoloff, L.
Laboratory of Cerebral Metabolism, Building
36, National Institute of Mental Health,
Bethesda, Maryland 20205 – USA

Stamm, R.
Department of Chemistry,
Brookhaven National Laboratory,
Upton, N. Y. 11973 – USA

Stjernberg, H.
Instrument AB Scanditronics, 18303 Täbi/
Stockholm – Sweden

Stöcklin, G.
Kernforschungsanlage Jülich GmbH.,
Institut für Chemie 1 (Nuklearchemie),
5170 Jülich – FRG

Stoffels, C.
Hôpital Sainte-Anne, Rue Cabanis,
75014 Paris – France

Taveras, J. M.
Massachusetts General Hospital,
Boston, Massachusetts 02114 – USA

Thal, H. U.
Neurochirurgische Klinik der Universität,
Moorenstr. 5,
4000 Düsseldorf – FRG

Uemura, K.
Division of Radiology and Nuclear Medicine,
Research Institute of Brain and Blood Vessels,
6–10 Senshu kubota machi,
Akita 010 – Japan

Van Kammen, D. P.
Biological Psychiatry Branch, National Insti-
tute of Health, 9000 Rockville Pike
Bethesda, Maryland 20205 – USA

Volkow, N.
Department of Psychiatry, New York Universi-
ty Medical Center, 550 First Avenue,
New York, N. Y. 10016 – USA

Vyska, K.
Kernforschungsanlage Jülich GmbH.,
Institut für Medizin,
5170 Jülich – FRG

Wagner, R.
Max-Planck-Institut für neurologische
Forschung, Abteilung für allgemeine
Neurologie, Ostmerheimer Str. 200,
5000 Köln 91 – FRG

Widén, L.
Department of Neurophysiology,
Karolinska Hospital,
10401 Stockholm – Sweden

Wienhard, K.
Max-Planck-Institut für neurologische
Forschung, Abteilung für allgemeine
Neurologie, Ostmerheimer Str. 200,
5000 Köln 91 – FRG

Wiesel, F.
Karolinska Hospital,
10401 Stockholm – Sweden

Wise, R.J.S.
MRC Cyclotron Unit, Hammersmith Hospital,
Ducane Road,
London W12 OHS – United Kingdom

Wolf, A.P.
Department of Chemistry,
Brookhaven National Laboratory,
Upton, N. Y. 11973 – USA

Wutz, W.
Kernforschungsanlage Jülich GmbH.,
Institut für Chemie 1 (Nuklearchemie),
5170 Jülich – FRG

Yamamoto, Y.L.
Neuroisotope Laboratory
Montreal Neurological Institute
McGill University
3801 University Street
Montreal, Quebec, Canada H3A 2B4

Yonekura, Y.
Department of Medicine,
Brookhaven National Laboratory
Upton, N. Y. 11973 – USA

Zarifian, E.
Hôpital Sainte-Anne, Rue Cabanis,
75014 Paris – France

Principles

(Chairman: D. H. Ingvar)

Biochemical Principles for the Measurement of Metabolic Rates in Vivo

By L. Sokoloff and C. Smith

Positron emission tomography provides the opportunity to assay the rates of biochemical processes in tissues in man. It does not, however, obviate the need to adhere to established principles of chemical and enzyme kinetics and tracer theory. A chemical reaction is the conversion of one species of molecule to another. The rate of this reaction can be measured by determining the rate of disappearance of one or more of the reactants or the formation of one or more of the products. The addition of a radioactive label to one of the reactants in molecular concentrations too negligible to alter the kinetics of the reaction facilitates the measurement of either the reactants or the products, but it introduces new complications. The rate of chemical transformation of the labeled species is measured, but this is not the rate of the total reaction including the nonradioactive species, which is the reaction to be measured. To derive the rate of the total reaction from measurement of the reaction rate of the labeled species, it is necessary to know the integrated specific activity (i.e., the ratio of labeled to total molecules) of the precursor pool. Occasionally the labeled species exhibits a kinetic difference from the natural compound, the so-called isotope effect; this effect can be calibrated, and appropriate correction made for it. In assays of biochemical reactions in vivo it is generally impossible to measure the integrated specific activity of the precursor pool directly. This would require the measurement of the complete time courses of the concentrations of the labeled and unlabeled precursor molecules in the tissue at the enzyme site. It is, therefore, usually necessary to determine the precursor pool specific activity indirectly from measurements in the blood supplied to the tissue. The specific activity in the arterial blood or plasma can be readily measured directly, and the precursor-specific activity calculated from it by correcting for the lag of the precursor pool in the tissue behind that of the plasma. To apply this correction it is

necessary to know the kinetics of the equilibration of the precursor pool in the tissue with that of the plasma.

The rate of a chemical reaction can be determined by measurement of precursor disappearance or product accumulation; generally the errors are smaller with the latter measurement because the percent change in the amount of product is much greater than the percent change in the amount of precursor. Emission tomography, which measures only the total concentration of the radioactive molecules, cannot distinguish among the various chemical species which may be labeled, neither the precursor nor any of the possible labeled products. Strategies must, therefore, be developed that ensure that the radioactivity is contained exclusively in the precursor and/or in one or more of the products specific to the chemical reaction to be assayed. The labeled precursor should be so selected that its chemical transformations are limited only to the pathway under study.

These general principles have been more or less successfully applied in two currently operational methods for the measurement of energy metabolism in the nervous system of animals and man. One method is the steady-state O_2 consumption technique which is based on the measurement of substrate disappearance (Fraczowski et al. 1980). Cerebral blood flow is measured by positron emission tomography with $C^{15}O_2$ and, when multiplied by arterial O_2 content, provides the steady-state delivery of O_2 to the tissues. Local cerebral O_2 extraction from the blood is measured by positron emission tomography with $^{15}O_2$. The product of local oxygen extraction, blood flow, and arterial O_2 content provides the values for local O_2 consumption. The other method is the radioactive deoxyglucose technique for the measurement of local cerebral glucose utilization. It has been used extensively in animals with autoradiography (Sokoloff et al. 1977; Sokoloff 1981), but it has been adapted to man by the use of

[^{18}F]fluorodeoxyglucose and positron emission tomography. The deoxyglucose technique is based on the measurement of product accumulation. Because it encompasses almost all the principles to be considered in the measurement of biochemical processes in vivo, it will serve as an informative example of their application.

The Deoxyglucose Method

Theoretical Basis

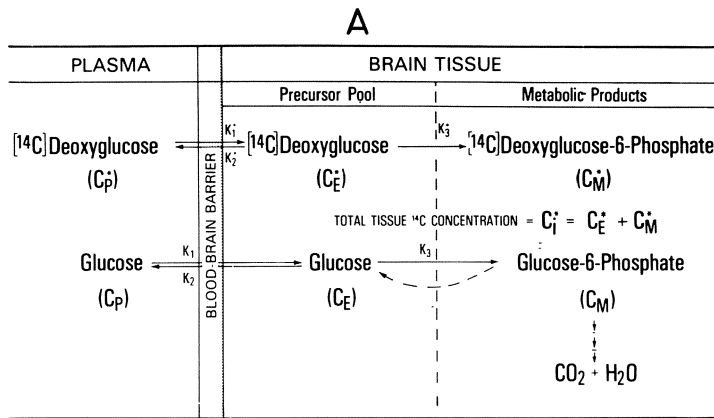
The deoxyglucose method was developed to measure the rates of glucose utilization simultaneously in all structural and/or functional components of the central nervous system in conscious animals (Sokoloff et al. 1977). It was developed first for use with [^{14}C]deoxyglucose and was designed specifically to take advantage of the localization made possible by quantitative autoradiography although its principles are equally applicable with any type of emission tomography as well. Although the purpose was to measure the local rates of glucose utilization, the analogue of glucose, 2-deoxy-D-glucose, rather than glucose itself, was selected as the labeled precursor because its biochemical properties make it easier to adhere to the biochemical principles for the measurement of glucose utilization in vivo by autoradiography or other emission tomographic techniques. If radioactive glucose is used as the precursor, some of the labeled products of glucose metabolism, particularly CO_2 or water, are lost too rapidly from the tissue, and the retention of many other labeled products is dependent on additional chemical reactions other than mere glucose metabolism. With radioactive deoxyglucose as precursor, the label is retained in the tissues either in the unmetabolized precursor molecules or in the immediate product of its metabolism. As will become clear subsequently, the use of deoxyglucose serves to isolate the chemical reaction under study to a well-defined reaction, the hexokinase-catalyzed phosphorylation of the hexose, the first step in the biochemical pathway of glucose metabolism.

The method was derived by analysis of a model based on the biochemical properties of 2-de-

oxyglucose and glucose in brain (Fig. 1 A) (Sokoloff et al. 1977). 2-Deoxyglucose (DG) is transported bidirectionally between blood and brain by the same carrier that transports glucose across the blood-brain barrier. In the cerebral tissues it is phosphorylated like glucose by hexokinase to produce 2-deoxyglucose-6-phosphate (DG-6-P). Deoxyglucose and glucose are, therefore, competitive substrates for both blood-brain transport and hexokinase-catalyzed phosphorylation. Unlike glucose-6-phosphate, however, which is metabolized further eventually to CO_2 and water, DG-6-P cannot be converted to fructose-6-phosphate, and it is also a poor substrate for glucose-6-phosphate dehydrogenase. There is relatively little glucose-6-phosphatase activity in the brain and even less deoxyglucose-6-phosphatase activity. Deoxyglucose-6-phosphate, once formed, remains, therefore, essentially trapped in the cerebral tissues, at least for 45 min (Sokoloff 1982).

If the interval of time is kept short enough, for example, less than 1 h, to allow the assumption of negligible loss of [^{14}C]DG-6-P from the tissues, then the quantity of [^{14}C]DG-6-P accumulated in any cerebral tissue at any given time following the introduction of [^{14}C]DG into the circulation is equal to the integral of the rate of [^{14}C]DG phosphorylation by hexokinase in that tissue during that interval of time. This integral is in turn related to the amount of glucose that has been phosphorylated over the same interval, depending on the time courses of the relative concentrations of [^{14}C]DG and glucose in the precursor pools and the Michaelis-Menten kinetic constants for hexokinase with respect to both [^{14}C]DG and glucose. With cerebral glucose consumption in a steady state, the amount of glucose phosphorylated during the interval of time equals the steady-state flux of glucose through the hexokinase-catalyzed step times the duration of the interval, and the net rate of flux of glucose through this step equals the rate of glucose utilization.

These relationships can be rigorously combined into a model (Fig. 1 A) which can be mathematically analyzed to derive an operational equation (Fig. 1 B), provided that the following assumptions are made: (1) steady state for glucose (i.e., constant plasma glucose concentration and constant rate of glucose con-

**B**

Functional Anatomy of the Operational Equation of the [^{14}C] Deoxyglucose Method

General Equation for Measurement of Reaction Rates with Tracers:

$$\text{Rate of Reaction} = \frac{\text{Labeled Product Formed in Interval of Time, 0 to T}}{\left[\begin{array}{c} \text{Isotope Effect} \\ \text{Correction Factor} \end{array} \right] \left[\begin{array}{c} \text{Integrated Specific Activity} \\ \text{of Precursor} \end{array} \right]}$$

Operational Equation of [^{14}C] Deoxyglucose Method:

$$R_i = \frac{\overbrace{\text{Labeled Product Formed in Interval of Time, 0 to T}}^{\text{Total } ^{14}\text{C in Tissue at Time, T}}}{\underbrace{\left[\begin{array}{c} \lambda \cdot V_m^* \cdot K_m \\ \Phi \cdot V_m \cdot K_m^* \end{array} \right]}_{\text{Isotope Effect Correction Factor}} \left[\underbrace{\int_0^T \left(\frac{C_p^*}{C_p} \right) dt}_{\text{Integrated Plasma Specific Activity}} - \underbrace{e^{-(k_2^* + k_3^*)T} \int_0^T \left(\frac{C_p^*}{C_p} \right) e^{(k_2^* + k_3^*)t} dt}_{\text{Correction for Lag in Tissue Equilibration with Plasma}} \right]}$$

Integrated Precursor Specific Activity in Tissue

sumption) throughout the experimental period; (2) homogeneous tissue compartment within which the concentrations of [^{14}C]DG and glucose are uniform and exchange directly with the plasma; and (3) concentrations of [^{14}C]DG (i.e., molecular concentrations of free [^{14}C]DG essentially equal to zero). The operational equation which defines R_i , the rate of glucose utilization per unit mass of tissue, i , in terms of measurable variables is presented in Fig. 1 B.

The rate constants, k_1^* , k_2^* and k_3^* , are determined in a separate group of animals by a nonlinear, iterative process which provides the least squares best-fit of an equation which defines the time course of tissue ^{14}C concentration in terms of the time, the history of the plasma [^{14}C]DG concentration, and the rate constants to the experimentally determined time courses of tissue and plasma concentrations of ^{14}C (Sokoloff et al. 1977). The λ , Φ , and the enzyme kinetic constants are grouped together to constitute a single, lumped constant (see equation in Fig. 1 B). It can be shown mathematically that this lumped constant is equal to the asymptotic value of the product of the ratio of the cerebral extraction ratios of [^{14}C]DG and glucose and the ratio of the arterial blood to plasma specific activities when the arterial plasma [^{14}C]DG concentration is maintained constant. The lumped constant is also determined in a separate group of animals from arterial and cerebral venous blood samples drawn during a programmed intravenous infusion which produces and maintains a constant

arterial plasma [^{14}C]DG concentration (Fig. 2) (Sokoloff et al. 1977; Kennedy et al. 1978).

Despite its complex appearance, the operational equation is really nothing more than a general statement of the standard relationship by which rates of enzyme-catalyzed reactions are determined from measurements made with radioactive tracers (Fig. 1 B). The numerator of the equation represents the amount of radioactive product formed in a given interval of time; it is equal to C_T^* , the combined concentrations of [^{14}C]DG and [^{14}C]DG-6-P in the tissue at time T , measured by the quantitative autoradiographic technique or emission tomography, less a term that represents the free unmetabolized [^{14}C]DG still remaining in the tissue. The denominator represents the integrated specific activity of the precursor pool times a factor, the lumped constant, which is equivalent to a correction factor for an isotope effect. The term with the exponential factor in the denominator takes into account the lag in the equilibration of the tissue precursor pool with the plasma.

By the use of labeled 2-deoxyglucose as a probe, a single biochemical reaction, the first step in the pathway of glucose metabolism has been isolated (Fig. 3). This is the hexokinase-catalyzed phosphorylation of glucose. The total amount of radioactive product formed and the integrated specific activity of the precursor at the enzyme site can be determined. From these data and the use of a correction factor, the "lumped constant", for the difference in the kinetic behavior of deoxyglucose and glucose,

Fig. 1 A, B. Theoretical basis of radioactive deoxyglucose method for measurement of local cerebral glucose utilization (Sokoloff et al. 1977). **A** Diagrammatic representation of the theoretical model. C_T^* represents the total ^{14}C concentration in a single homogeneous tissue of the brain. C_P^* and C_P represent the concentration of [^{14}C]deoxyglucose and glucose in the arterial plasma, respectively; C_E^* and C_E represent their respective concentrations in the tissue pools that serve as substrates for hexokinase. C_M^* represents the concentration of [^{14}C]deoxyglucose-6-phosphate in the tissue. The constants k_1^* , k_2^* , and k_3^* represent the rate constants for carrier-mediated transport of [^{14}C]deoxyglucose from plasma to tissue, for carrier-mediated transport back from tissue to plasma, and for phosphorylation by hexokinase, respectively; the constants k_1 , k_2 , and k_3 are the equivalent rate constants for glucose. [^{14}C]Deoxyglucose and glucose

share and compete for the carrier that transports both between plasma and tissue and for hexokinase which phosphorylates them to their respective hexose-6-phosphates. The *dashed arrow* represents the possibility of glucose-6-phosphate hydrolysis by glucose-6-phosphatase activity, if any. **B** Operational equation of the radioactive deoxyglucose method and its functional anatomy. T represents the time of termination of the experimental period; λ equals the ratio of the distribution space of deoxyglucose in the tissue to that of glucose; Φ equals the fraction of glucose which once phosphorylated continues down the glycolytic pathway; and K_m^* and V_m^* and K_m and V_m represent the familiar Michaelis-Menten kinetic constants of hexokinase for deoxyglucose and glucose, respectively. The other symbols are the same as those defined in A

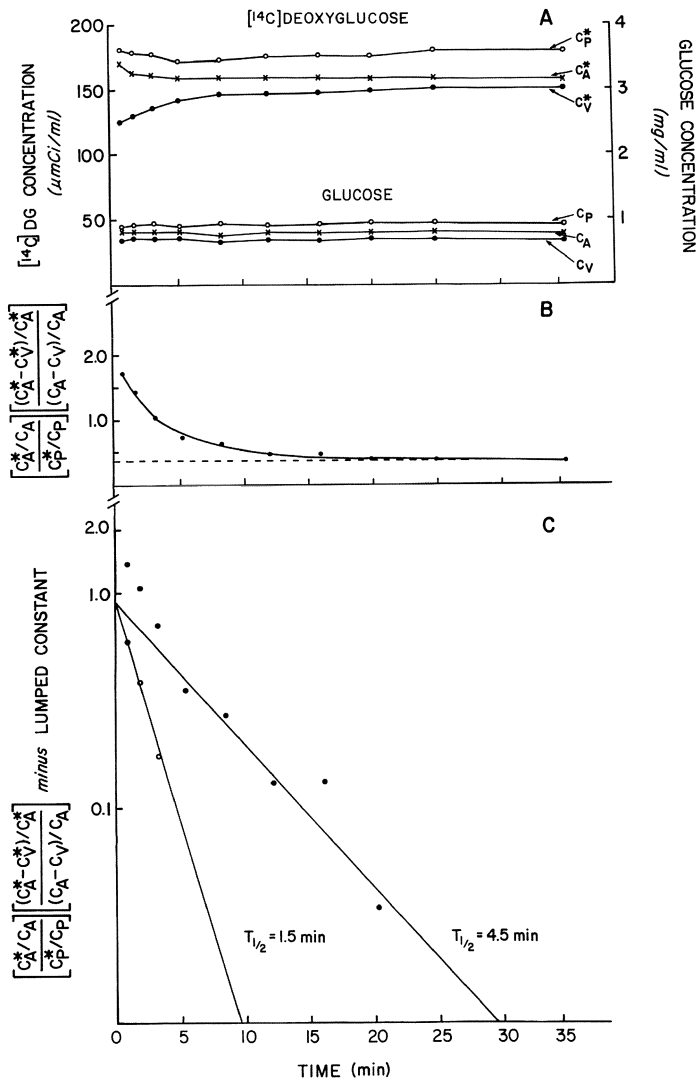


Fig. 2 A–C. Data obtained and their use in determination of the lumped constant and the combination of rate constants ($k_2^* + k_3^*$) in a representative experiment. **A** Time courses of arterial blood and plasma concentrations of $[^{14}\text{C}]\text{DG}$ and glucose and cerebral venous blood concentrations of $[^{14}\text{C}]\text{DG}$ and glucose during programmed intravenous infusion of $[^{14}\text{C}]\text{DG}$. **B** Arithmetic plot of the function derived from the variables in **A** and combined as indicated in the formula on the ordinate against time. This function declines exponentially, with a rate constant equal to $(k_2^* + k_3^*)$, until it reaches an asymptotic value equal to the lumped constant, 0.35 in this experiment (dashed line). **C** Semilogarithmic plot of the curve in **B** less the lumped constant, i. e., its asymptot-

ic value. *Solid circles* represent actual values. This curve is analyzed into two components by a standard curve-peeling technique to yield the two *straight lines* representing the separate components. *Open circles* are points for the fast component, obtained by subtracting the values for the slow component from the *solid circles*. The rate constants for these two components represent the values of $(k_2^* + k_3^*)$ for two compartments; the fast and slow compartments are assumed to represent gray and white matter, respectively. In this experiment the values for $(k_2^* + k_3^*)$ were found to equal 0.462 (half-time = 1.5 min) and 0.154 (half-time = 4.5 min) in gray and white matter, respectively (Kennedy et al. 1978)

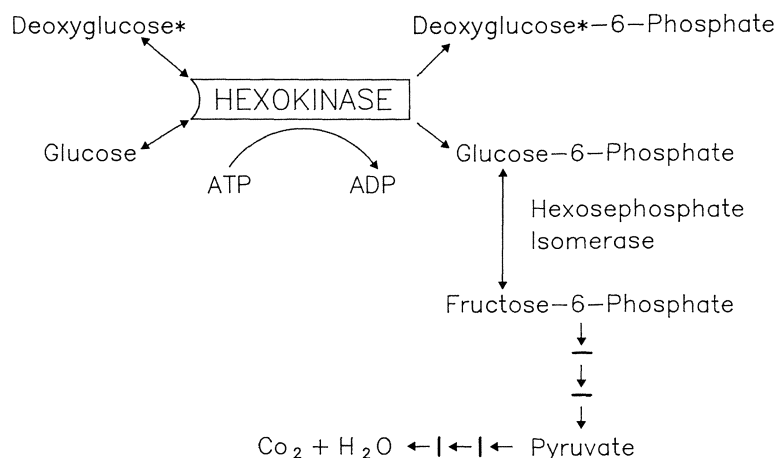


Fig. 3. Schema illustrating the fundamental principle of the radioactive deoxyglucose method for the measurement of local cerebral glucose utilization. Glucose utilization commences with the hexokinase-catalyzed phosphorylation of glucose by ATP, but the product of this reaction, glucose-6-phosphate, is not retained in the tissues. Instead, it is metabolized further to products like CO_2 and H_2O that leave the tissue. Deoxyglucose, an analogue and competitive substrate with glucose in the hexokinase reaction, leads to a product, deoxyglucose-6-phosphate, that does accumulate in the tissue quantitatively for a reasonable length of time. By putting a label on the deoxyglucose, it is possible to measure the rate of labeled deoxyglucose-6-phosphate formation. From

a knowledge of the time course of the relative concentrations of labeled deoxyglucose and glucose in the tissue at the enzyme site and the relative Michaelis-Menten constants of hexokinase for the two substrates, it is possible to calculate how much glucose must have also been phosphorylated during the production of the measured amount of deoxyglucose-6-phosphate. The integrated relative concentrations of labeled deoxyglucose and glucose in the tissue are calculated from the measured time courses of the two compounds in the arterial plasma by subtracting from the integrated plasma-specific activity a term that corrects for the lag of the tissue behind the plasma

the net rate of glucose phosphorylation can be calculated by the operational equation. In a steady state the net rate through any step in a pathway equals the net rate through the overall pathway. The deoxyglucose method, therefore, measures in vivo the net rate of glucose phosphorylation and in a steady state the net rate of the entire glycolytic pathway.

Applications of Biochemical Principles to Design of Procedure

The operational equation dictates the variables to be measured to determine the local rates of cerebral glucose utilization. The specific procedure employed is designed to evaluate these variables and to minimize potential errors that might occur in the actual application of the method. If the rate constants, k_1^* , k_2^* , and k_3^* , are precisely known, then the equation is generally applicable with any mode of administration of

$[^{14}\text{C}]\text{DG}$ and for a wide range of time intervals. At the present time the rate constants have been fully determined for $[^{14}\text{C}]\text{DG}$ only in the conscious rat (Sokoloff et al. 1977) (Table 1). Partial determination of the rate constants indicates that they are similar in the monkey (Kennedy et al. 1978). These rate constants can be expected to vary with the condition of the animal, however, and for most accurate results should be redetermined for each condition studied. The structure of the operational equation suggests a more practical alternative. All the terms in the equation that contain the rate constants approach zero with increasing time if the $[^{14}\text{C}]\text{DG}$ is so administered that the plasma $[^{14}\text{C}]\text{DG}$ concentration also approaches zero. From the values of the rate constants determined in normal animals and the usual time course of the clearance of $[^{14}\text{C}]\text{DG}$ from the arterial plasma following a single intravenous pulse at zero time, it has been determined that an interval of 30–45 min after a pulse is adequate for these terms to become sufficiently

Table 1. Values of rate constants in the normal conscious albino rat (Sokoloff et al. 1977)

Structure	Rate constants			Distribution volume	Half-life of precursor pool
	(min ⁻¹)			(ml/g)	pool (min)
	k_1^*	k_2^*	k_3^*	$k_1^*/(k_2^* + k_3^*)$	$\text{Log}_e 2 / (k_2^* + k_3^*)$
Gray matter					
Visual cortex	0.189 ± 0.048	0.279 ± 0.176	0.063 ± 0.040	0.553	2.03
Auditory cortex	0.226 ± 0.068	0.241 ± 0.198	0.067 ± 0.057	0.734	2.25
Parietal cortex	0.194 ± 0.051	0.257 ± 0.175	0.062 ± 0.045	0.608	2.17
Sensory motor cortex	0.193 ± 0.037	0.208 ± 0.112	0.049 ± 0.035	0.751	2.70
Thalamus	0.188 ± 0.045	0.218 ± 0.144	0.053 ± 0.043	0.694	2.56
Medial geniculate body	0.219 ± 0.055	0.259 ± 0.164	0.055 ± 0.040	0.697	2.21
Lateral geniculate body	0.172 ± 0.038	0.220 ± 0.134	0.055 ± 0.040	0.625	2.52
Hypothalamus	0.158 ± 0.032	0.226 ± 0.119	0.043 ± 0.032	0.587	2.58
Hippocampus	0.169 ± 0.043	0.260 ± 0.166	0.056 ± 0.040	0.535	2.19
Amygdala	0.149 ± 0.028	0.235 ± 0.109	0.032 ± 0.026	0.558	2.60
Caudate putamen	0.176 ± 0.041	0.200 ± 0.140	0.061 ± 0.050	0.674	2.66
Superior colliculus	0.198 ± 0.054	0.240 ± 0.166	0.046 ± 0.042	0.692	2.42
Pontine gray matter	0.170 ± 0.040	0.246 ± 0.142	0.037 ± 0.033	0.601	2.45
Cerebellar cortex	0.225 ± 0.066	0.392 ± 0.229	0.059 ± 0.031	0.499	1.54
Cerebellar nucleus	0.207 ± 0.042	0.194 ± 0.111	0.038 ± 0.035	0.892	2.99
Mean	0.189	0.245	0.052	0.647	2.39
± SEM	± 0.012	± 0.040	± 0.010	± 0.073	± 0.40
White matter					
Corpus callosum	0.085 ± 0.015	0.135 ± 0.075	0.019 ± 0.033	0.552	4.50
Genu of corpus callosum	0.076 ± 0.013	0.131 ± 0.075	0.019 ± 0.034	0.507	4.62
Internal capsule	0.077 ± 0.015	0.134 ± 0.085	0.023 ± 0.039	0.490	4.41
Mean	0.079	0.133	0.020	0.516	4.51
± SEM	± 0.008	± 0.046	± 0.020	± 0.171	± 0.90

small that considerable latitude in inaccuracies of the rate constants is permissible without appreciable error in the estimates of local glucose consumption (Sokoloff et al. 1977). An additional advantage derived from the use of a single pulse of [¹⁴C]DG followed by a relatively long interval before killing the animal for measurement of local tissue ¹⁴C concentration is that by then most of the free [¹⁴C]DG in the tissues has been either converted to [¹⁴C]DG-6-P or transported back to the plasma; the radioactivity in the tissues and the optical densities in the autoradiographs then represent mainly the concentrations of the product, [¹⁴C]DG-6-P, and, therefore, reflect directly the relative rates of glucose utilization in the various cerebral tissues.

The experimental procedure (Sokoloff et al. 1977) is to inject a pulse of [¹⁴C]DG intravenously at zero time and to decapitate the animal and freeze the brain at a measured time, *T*,

30–45 min later; in the interval timed arterial samples are taken for the measurement of plasma [¹⁴C]DG and glucose concentrations. To ensure that tracer conditions are maintained, the dose of [¹⁴C]DG should not exceed 2.5 μmol deoxyglucose/kg body wt. Tissue ¹⁴C concentrations, *C_t*^{*}, are measured at time *T* by quantitative autoradiography of 20-μm frozen-dried sections prepared serially from the entire brain and calibrated radioactive standards (Fig. 4) (Sokoloff et al. 1977). Local cerebral glucose utilization is then calculated by the operational equation.

The design of the deoxyglucose method is based on an operational equation, derived by the mathematical analysis of a model of the biochemical behavior of [¹⁴C]deoxyglucose and glucose in brain (Fig. 1). Although the model and its mathematical analysis are as rigorous and comprehensive as reasonably possible, it must be recognized that models almost always

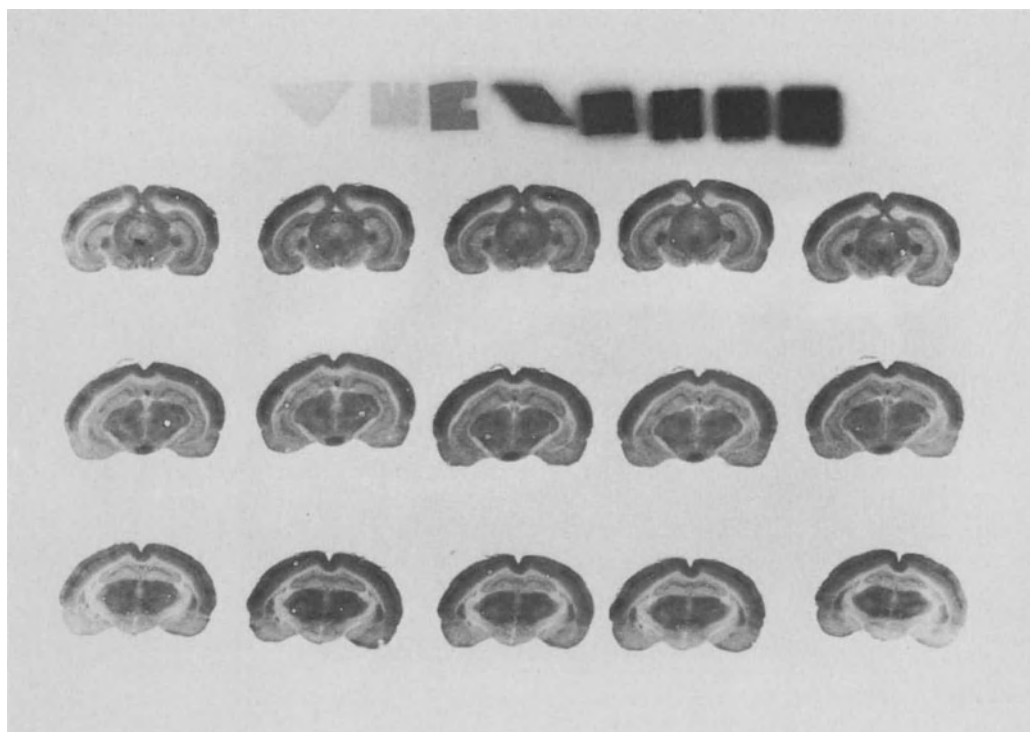


Fig. 4. [^{14}C]Deoxyglucose autoradiographs of sections of conscious rat brain and of calibrated [^{14}C]methylmethacrylate standards used to quantify ^{14}C

concentration in tissues by quantitative densitometry. (Sokoloff et al. 1977)

represent idealized situations and cannot possibly take into account every single known, let alone unknown, property of a complex biological system. There remained, therefore, the possibility that extensive experience with the [^{14}C]deoxyglucose method might uncover weaknesses, limitations, or flaws serious enough to limit its usefulness or even to invalidate it. Several years have now passed since its introduction, and numerous applications of it have been made. The results of this experience generally establish the validity and worth of the method. There still remain, however, some potential problems in specialized situations, and several theoretical and practical issues need further clarification.

The main potential sources of error are the rate constants and the lumped constant. The problem with them is that they are not determined in the same animals and at the same time when local cerebral glucose utilization is being measured. They are measured in separate groups of comparable animals and then used subse-

quently in other animals in which glucose utilization is being measured. The part played by these constants in the method is defined by their role in the operational equation of the method (Fig. 1B).

Rate Constants

The rate constants, k_1^* , k_2^* , and k_3^* , for deoxyglucose have thus far been fully determined for various cerebral tissues only in the normal conscious albino rat (Sokoloff et al. 1977) (Table 1). Partial determination of the rate constants in the normal conscious rhesus monkey indicates that they are quite similar to those in the rat. All the rate constants vary from tissue to tissue, but the variation among gray structures and among white structures is considerably less than differences between the two types of tissues (Table 1). The rate constants k_2^* and k_3^* appear in the equation only as their sum, and $(k_2^* + k_3^*)$ is equal to the rate constant for the turnover of the free [^{14}C]deoxyglucose pool in the

tissue. The half-life of the free [^{14}C]deoxyglucose pool can then be calculated by dividing $(k_2^* + k_3^*)$ into the natural logarithm of 2 and has been found to average 2.4 min in gray matter and 4.5 min in white matter in the normal conscious rat (Table 1).

The rate constants vary not only from structure to structure but can be expected to vary with the condition. For example, k_1^* and k_2^* are influenced by both blood flow and transport of [^{14}C]deoxyglucose across the blood-brain barrier, and because of the competition for the transport carrier, the glucose concentrations in the plasma and tissue affect the transport of [^{14}C]deoxyglucose and, therefore, also k_1^* and k_2^* . The constant k_3^* is related to phosphorylation of [^{14}C]deoxyglucose and will certainly change when glucose utilization is altered. To minimize potential errors due to inaccuracies in the values of the rate constants used, it was decided to sacrifice time resolution for accuracy.

If the [^{14}C]deoxyglucose is given as an intravenous pulse and sufficient time is allowed for the plasma to be cleared of the tracer, then the influence of the rate constants, and the functions that they represent, on the final result diminishes with increasing time until ultimately it becomes zero. This relationship is implicit in the structure of the operational equation (Fig. 1B); as C_p^* approaches zero, then the terms containing the rate constants also approach zero with increasing time. The significance of this relationship is graphically illustrated in Fig. 5. From typical arterial plasma [^{14}C]deoxyglucose and glucose concentration curves obtained in a normal conscious rat, the portion of the denominator of the operational equation underlined by the heavy bar was computed with a wide range of values for $(k_2^* + k_3^*)$ as a function of time. The values for $(k_2^* + k_3^*)$ are presented as their equivalent half-lives calculated as described above. The values of $(k_2^* +$

INFLUENCE OF RATE CONSTANTS ON INTEGRATED POOL SPECIFIC ACTIVITY

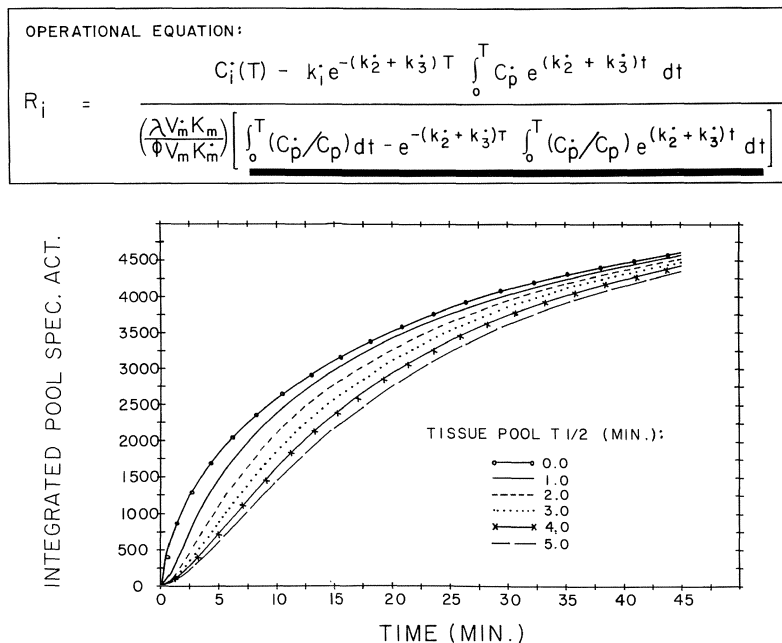


Fig. 5. Influence of time and rate constants $(k_2^* + k_3^*)$ on integrated precursor pool specific activity in a normal conscious rat given an intravenous pulse of $50 \mu\text{Ci}$ [^{14}C]deoxyglucose at zero time. The time courses of the arterial plasma [^{14}C]deoxyglucose and glucose concentrations were measured following the

pulse. The portion of the equation underlined, corresponding to integrated pool specific activity, was computed as a function of time with different values of $(k_2^* + k_3^*)$, as indicated by their equivalent half-lives, calculated according to $T_{1/2} = 0.693/(k_2^* + k_3^*)$ (Sokoloff 1979)

k_3^*) vary from infinite (i.e., $T_{1/2} = 0$ min) to 0.14/min (i.e., $T_{1/2} = 5$ min) and more than cover the range of values to be expected under physiological conditions. The portion of the equation underlined and computed represents the integral of the precursor pool specific activity in the tissue. The curves represent the time course of this function, one each for every value of $(k_2^* + k_3^*)$ examined. It can be seen that these curves are widely different at early times but converge with increasing time until at 45 min the differences over the entire range of $(k_2^* + k_3^*)$ equal only a small fraction of the value of the integral. These curves demonstrate that at short times enormous errors can occur if the values of the rate constants are not precisely known, but only negligible errors occur at 45 min, even over a wide range of rate constants of severalfold. In fact, it was precisely for this reason that [^{14}C]deoxyglucose rather than [^{14}C]glucose was selected as the tracer for glucose metabolism. The relationships are similar for glucose. Because the products of [^{14}C]glucose metabolism are so rapidly lost from the tissues, it is necessary to limit the experimental period to short times, when enormous errors can occur if the rate constants are not precisely known. [^{14}C]Deoxyglucose permits the prolongation of the experimental period to times when inaccuracies in rate constants have little effect on the final result.

It should be noted, however, that in pathological conditions, such as severe ischemia or hyperglycemia, the rate constants may fall far beyond the range examined in Fig. 5. There is evidence, for example, that this occurs with hyperglycemia and ischemia (Hawkins et al. 1981). In such abnormal conditions it may be necessary to redetermine the rate constants for the particular condition under study.

Lumped Constant

The lumped constant is composed of six separate constants. One of these, Φ , is a measure of the study-state hydrolysis of glucose-6-phosphate to free glucose and phosphate. Because in normal brain tissue there is little such phosphohydrolase activity (Hers 1957), Φ is normally approximately equal to unity. The other components are arranged in three ratios: λ , which is the ratio of distribution spaces in the

tissues for deoxyglucose and glucose; V_m^*/V_m ; and K_m^*/K_m . Although each individual constant may vary from structure to structure and condition to condition, it is likely that the ratios tend to remain the same under normal conditions. For reasons described in detail previously (Sokoloff et al. 1977), it is reasonable to believe that the lumped constant is the same throughout the brain and is characteristic of the species of animal, but only in normal tissue. Although reasonable, it is not certain, and there are theoretical possibilities that it may not be so. Empirical experience thus far indicates that it is. The greatest experience has been accumulated in the albino rat. In this species the lumped constant for the brain as a whole has been determined under a variety of conditions (Sokoloff et al. 1977). In the normal conscious rat local cerebral glucose utilization, determined by the [^{14}C]deoxyglucose method with the single value of the lumped constant for the brain as a whole, correlates almost perfectly ($r = 0.96$) with local cerebral blood flow, measured by the [^{14}C]iodoantipyrine method, an entirely independent method (Sokoloff 1978). It is generally recognized that local blood flow is adjusted to local metabolic rate, but if the single value of the lumped constant did not apply to the individual structures studied, then errors in local glucose utilization would occur that might be expected to obscure the correlation. Also, the lumped constant has been directly determined in the albino rat in the normal conscious state, under barbiturate anesthesia, and during the inhalation of 5% CO_2 ; no significant differences were observed (Table 2). The lumped constant does vary with the species of animal. It has now also been determined in the rhesus monkey (Kennedy et al. 1978), cat (M. Miyaoka, J. Magnes, C. Kennedy, M. Shinohara, and L. Sokoloff, unpublished data), and beagle puppy (Duffy et al. 1979), and each species has a different value (Table 2). The values for local rates of glucose utilization determined with these lumped constants in these species are very close to what might be expected from measurement of energy metabolism in the brain as a whole by other methods (Table 3).

Although there is no experimental evidence yet to indicate that the lumped constants change under physiological conditions, they may change in pathological states. In severe hypo-

Table 2. Values of the lumped constant in the albino rat, rhesus monkey, cat, and dog

Animal	No. of animals	Mean $\pm$ SD	SEM
Albino rat			
Conscious	15	0.464 $\pm$ 0.099 ^a	$\pm$ 0.026
Anesthetized	9	0.512 $\pm$ 0.118 ^a	$\pm$ 0.039
Conscious (5% CO ₂)	2	0.463 $\pm$ 0.122 ^a	$\pm$ 0.086
Combined	26	0.481 $\pm$ 0.119	$\pm$ 0.023
Rhesus monkey			
Conscious	7	0.344 $\pm$ 0.095	$\pm$ 0.036
Cat			
Anesthetized	6	0.411 $\pm$ 0.013	$\pm$ 0.005
Dog (beagle puppy)			
Conscious	7	0.558 $\pm$ 0.082	$\pm$ 0.031

The values were obtained as follows: rat (Sokoloff et al. 1977); monkey (Kennedy et al. 1978); cat (M. Miyaoka, J. Magnes, C. Kennedy, M. Shinohara, and L. Sokoloff, unpublished data); dog (Duffy et al. 1979).

^a No statistically significant difference between normal conscious and anesthetized rats ($0.3 < p < 0.4$) and conscious rats breathing 5% CO₂ ($p > 0.9$)

glycemia there is a marked increase in the lumped constant (Suda et al. 1981), and in severe hyperglycemia there is a moderate decrease (Schuier et al. 1981). Tissue damage may also disrupt the normal cellular compartmentation, and there is no assurance that λ , the ratio of the distribution spaces for [¹⁴C]deoxyglucose and glucose, is the same in damaged tissue as in normal tissue. In pathological states there may be release of lysosomal acid hydrolases that may hydrolyze glucose-6-phosphate and thus alter the value of Φ . It is advisable, therefore, to reevaluate the lumped constant in pathological states.

Glucose-6-Phosphatase

Although relatively low the activity of glucose-6-phosphatase in brain is not zero (Sokoloff 1982). This enzyme is capable of hydrolyzing the product [¹⁴C]deoxyglucose-6-phosphate back to free [¹⁴C]deoxyglucose, thus causing loss of product. Fortunately, the product and enzyme are in separate cellular compartments,

Table 3. Representative values for local cerebral glucose utilization in the normal conscious albino rat and monkey ($\mu\text{mol}/100 \text{ g}/\text{min}$)

Structure	Albino rat ^a (10)	Monkey ^b (7)
Gray matter		
Visual cortex	107 $\pm$ 6	59 $\pm$ 2
Auditory cortex	162 $\pm$ 5	79 $\pm$ 4
Parietal cortex	112 $\pm$ 5	47 $\pm$ 4
Sensory motor cortex	120 $\pm$ 5	44 $\pm$ 3
Thalamus: lateral nucleus	116 $\pm$ 5	54 $\pm$ 2
Thalamus: ventral nucleus	109 $\pm$ 5	43 $\pm$ 2
Medial geniculate body	131 $\pm$ 5	65 $\pm$ 3
Lateral geniculate body	96 $\pm$ 5	39 $\pm$ 1
Hypothalamus	54 $\pm$ 2	25 $\pm$ 1
Mamillary body	121 $\pm$ 5	57 $\pm$ 3
Hippocampus	79 $\pm$ 3	39 $\pm$ 2
Amygdala	52 $\pm$ 2	25 $\pm$ 2
Caudate putamen	110 $\pm$ 4	52 $\pm$ 3
Nucleus accumbens	82 $\pm$ 3	36 $\pm$ 2
Globus pallidus	58 $\pm$ 2	26 $\pm$ 2
Substantia nigra	58 $\pm$ 3	29 $\pm$ 2
Vestibular nucleus	128 $\pm$ 5	66 $\pm$ 3
Cochlear nucleus	113 $\pm$ 7	51 $\pm$ 3
Superior olivary nucleus	133 $\pm$ 7	63 $\pm$ 4
Inferior colliculus	197 $\pm$ 10	103 $\pm$ 6
Superior colliculus	95 $\pm$ 5	55 $\pm$ 4
Pontine gray matter	62 $\pm$ 3	28 $\pm$ 1
Cerebellar cortex	57 $\pm$ 2	31 $\pm$ 2
Cerebellar nuclei	100 $\pm$ 4	45 $\pm$ 2
White matter		
Corpus callosum	40 $\pm$ 2	11 $\pm$ 1
Internal capsule	33 $\pm$ 2	13 $\pm$ 1
Cerebellar white matter	37 $\pm$ 2	12 $\pm$ 1

The values are the means $\pm$ standard errors from measurements made in the number of animals indicated in parentheses.

^a From Sokoloff et al. (1977).

^b From Kennedy et al. (1978)

at least for a time. The [¹⁴C]DG-6-P is formed in the cytosol and must be transported into the cisterns of the endoplasmic reticulum where the glucose-6-phosphatase resides, before the hydrolysis can occur. This compartmentalization provides a period of time before the effects of glucose-6-phosphatase become significant. If the experimental period is kept within 45 min, then the influence of glucose-6-phosphatase is negligible and can be ignored (Sokoloff et al. 1977; Sokoloff 1982). If, however, it is necessary to extend the experimental period to longer intervals, as, for example, in studies in man with positron emission tomography

(Phelps, et al. 1979), then it is necessary to account for the effects of glucose-6-phosphatase activity. This has been done by modifying slightly the original model to include a k_4^* , the rate constant for [^{14}C]DG-6-P hydrolysis by glucose-6-phosphatase, and deriving a modified operational equation that incorporates it (Huang et al. 1980; Sokoloff 1982).

Computerized Image-Processing

The regional localization obtained with the deoxyglucose method is achieved by the use of quantitative autoradiography. The autoradiographs contain an immense amount of information that cannot be practically recovered by manual densitometry or adequately represented by tabular presentation of the data. A computerized image-processing system has, therefore, been developed to analyze and transform the autoradiographs into color-coded pictorial maps of the rates of local glucose utilization throughout the CNS (Gooch et al. 1980). The autoradiographs are scanned automatically by a computer-controlled scanning microdensitometer. The optical density of each spot on the autoradiograph, from 25 to 100 μm as selected, is stored in a computer, converted to ^{14}C concentration on the basis of the optical densities of the calibrated ^{14}C plastic standards, and then converted to local rate of glucose utilization by solution of the operational equation. Colors are assigned to narrow ranges to the rates of glucose utilization, and the autoradiographs are then displayed in a color monitor in color along with a calibrated color scale for identifying the rate of glucose utilization in each spot of the autoradiograph from its color. The display is similar to that used by Lassen, Ingvar, and associates (Lassen et al. 1978) for regional cortical blood flow, but it represents the rates of glucose utilization in all parts of the brain down to a resolution of at least 100 μm . This image-processing system has been adapted for use with positron emission tomographic data obtained in man, but then, of course, the resolution is much less and limited by that of the tomography.

The [^{18}F]Fluorodeoxyglucose Technique

The deoxyglucose method was originally designed for use in animals with quantitative autoradiography and the radioactive isotopes most suitable for film autoradiography, ^{14}C or ^3H . Its basic physiological and biochemical principles apply, however, to man as well, and it is applicable to man provided the local tissue concentrations of isotope can be measured in the brain. Film autoradiography is a type of emission tomography that for obvious reasons cannot be used in man, but recent developments in computerized tomography have made it possible to determine local concentrations of γ -emitting isotopes in the cerebral tissues. The only possible γ -emitting isotopes that could be incorporated into 2-deoxyglucose are ^{11}C or ^{15}O , but the short half-lives of these isotopes present problems in the synthesis of the compounds. Alternatively, an analogue of 2-deoxyglucose with another γ -emitting isotope but with similar biochemical properties could be used. It is a common experience that the substitution of the very small atom, F, in place of a hydrogen at a judicious site in the molecule often does not alter the basic biochemical behavior of metabolic substrates. 2-[^{18}F]Fluoro-2-deoxy-D-glucose has been synthesized, found to retain the biochemical properties of 2-deoxyglucose, and used to measure cerebral glucose utilization in man by means of single photon emission tomography (Reivich et al. 1979). ^{18}F is actually a positron emitter, and the absorption of positrons in the tissues gives rise to two coincident annihilation γ -rays of equal energy traveling at almost 180° to each other. Positron emission tomography takes advantage of these coincident annihilation γ -rays and, therefore, is inherently capable of better spatial resolution than single photon tomography. The [^{18}F]fluorodeoxyglucose method is, therefore, now generally used with positron emission tomography (Phelps et al. 1979). Present positron emission tomography with [^{18}F]fluorodeoxyglucose is still relatively slow, and it may take up to 2 h to obtain sufficient counts for accurate measurements of local ^{18}F concentrations in all parts of the brain. Although low in brain, glucose-6-phosphatase activity is not zero, and its effect becomes significant after the first 45 min

after the pulse of tracer (Sokoloff 1982). It has, therefore, been necessary to modify the model to include a rate constant for the hydrolysis of the phosphorylated product by the glucose-6-phosphatase and to derive a new operational equation that takes this activity into account (Phelps et al. 1979; Huang et al. 1980; Sokoloff 1982). The [^{18}F]fluorodeoxyglucose technique for the measurement of local cerebral glucose utilization in man is now operational and in use for studies of the human brain in health and disease in a number of laboratories throughout the world.

Local Cerebral Protein Synthesis

The basic biochemical principles for the measurement of metabolic rates *in vivo*, which were so effectively applied in the deoxyglucose technique, also apply to other metabolic processes. A metabolic activity of broad interest is protein synthesis. This biochemical activity is not likely to reflect directly functional activity, at least not acutely, but it may well be involved in slower more gradual processes in the nervous system, such as growth and development, plasticity, regeneration and repair, response to drugs and hormones, and possibly learning and memory. Protein synthesis may also be altered in pathological states, such as brain tumors, mental retardation due to metabolic errors, aging and senility, Alzheimer's disease, Huntington's disease, and endocrine diseases.

A method for the measurement of local rates of protein synthesis in the nervous system is under development (Smith et al. 1980). Like the deoxyglucose method it is designed to achieve localization by quantitative autoradiography, although eventually it should be adaptable to positron emission tomography. Also like the deoxyglucose methods it is based on the same biochemical principles described above, but their application to the measurement of local cerebral protein synthesis may be far more complex because of still undefined properties and kinetics of the equilibration of the precursor amino acid pool in the tissue with the circulating amino acid pool in the plasma.

The two essential variables which must be determined in any quantitative radioactive assay

of the rate of a reaction are the amount of product formed and the integrated precursor specific activity over a measured interval of time. Because autoradiographic or emission tomographic techniques measure the concentration only of the radioisotope and not that of the radioactive product itself, it is essential that the method be so designed that the label in the tissue is confined only to the product molecule itself or, at least, in well-defined chemical species which can be quantified separately. This problem is mitigated by the use of carboxyl-labeled L-leucine, an essential amino acid which is prevalent in most proteins and which, other than incorporation into protein, has a single simple pathway of metabolic degradation. In the pathway of degradation, the amino acid is first transaminated and then rapidly decarboxylated; the label is then lost as radioactive CO_2 , which, because of dilution by the large pool of unlabeled CO_2 constantly generated by carbohydrate metabolism, the relatively slow rate of CO_2 fixation, and the rapid removal of CO_2 from brain tissue by the blood flow, is removed from the tissues. The label is then retained only in the product of the reaction, labeled protein, and the residual unincorporated amino acid. The concentration of free labeled amino acid can be calculated from the history of the plasma concentration and the kinetic constants for the equilibration of the tissue free amino acid pools and the plasma. The concentration of free labeled amino acid in the tissue can be minimized by its intravenous administration as a pulse at zero time and then waiting a sufficiently long time for the plasma and tissue to be cleared of labeled amino acid. One hour after the pulse the fraction of total radioactivity in the tissue that is in the free amino acid pool is small (about 10%) in the rat. In adult monkey and probably in man, however, it is large (30%–50%) because of the relatively slow rates of protein synthesis and of clearance of the free amino acid pools. The error in subtracting a poorly defined free amino acid pool concentration from a total concentration that is not a great deal larger may, of course, be enormous.

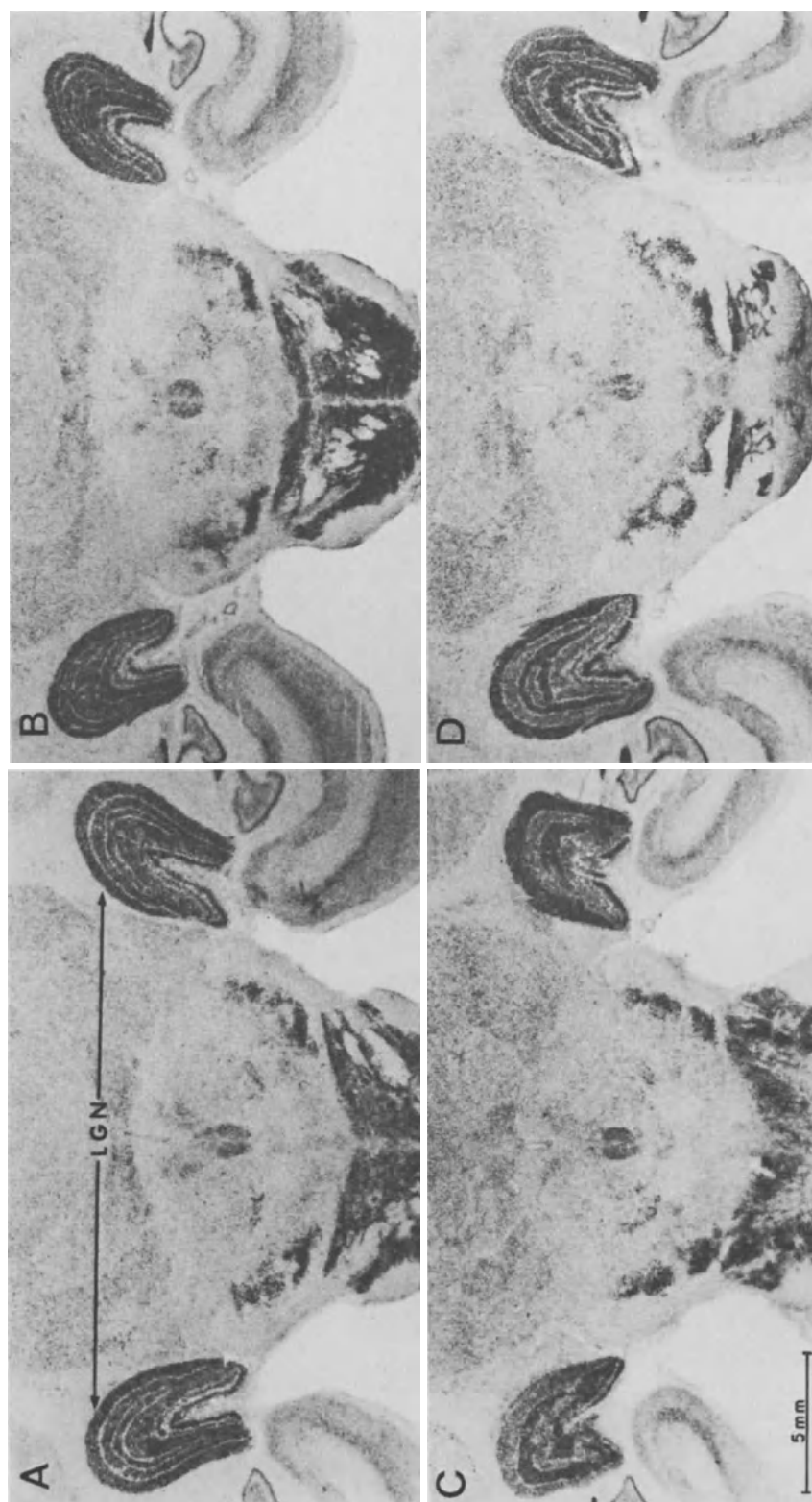
An even more difficult problem is the determination of the integrated precursor pool specific activity in the tissue from measurements in the plasma. To accomplish this, it is necessary to

know the kinetics of the equilibration of the precursor pool in the cells with that of the plasma. What makes this problem particularly perplexing is that there is evidence that amino acids in the cells are compartmentalized with only a fraction of the total intracellular amino acid content representative of the pool that serves as precursor for protein synthesis. Therefore, although it is relatively simple to measure the rate constants for the equilibration of the total amino acid pool with that of the plasma, there is little assurance that this pool reflects the kinetic behavior of the fraction of it that is the precursor for protein synthesis. A further complication is the possibility of significant dilution of the intracellular precursor amino acid pool by unlabeled amino acid derived from the slow but continuous turnover of the protein components of the cell. The magnitude of this potential dilution is even more difficult to evaluate. Nevertheless, studies are currently in progress in our laboratory that are designed to resolve these problems. If successful, they will not only determine the rate constants for the turnover of the true precursor pool but also the degree of admixture of unlabeled amino acids from protein breakdown with amino acids in the precursor pool.

Although the method is not yet fully developed, experiments with our first and most primitive model already indicate the potential usefulness for a technique that measures local cerebral protein synthesis. Model I is essentially the same as that for the deoxyglucose method; it assumes a single tissue pool of free amino acid, all of which equilibrates uniformly with the plasma and serves as the precursor pool for protein synthesis with no dilution by unlabeled amino acids derived from protein degradation (Smith et al. 1980). Although the quantitative values obtained with this version of the method are probably not accurate and may be underestimates of the true rates of L-leucine incorporation into protein, the results demonstrate that the rates of protein synthesis do change in specific regions of the brain in response to altered function. For example, in the rat, section of one hypoglossal nerve is followed by increased protein synthesis in the ipsilateral hypoglossal nucleus after a lag of close to 4 days (Agranoff et al. 1980). The increase reaches a maximum between 20%–30%, and protein synthesis does

not return to normal until regeneration and restoration of functional activity in the hypoglossal nerve is complete (C. B. Smith, unpublished observations).

The method has also been used to study plasticity in the binocular visual system of the newborn rhesus monkey (Kennedy et al. 1981). The outputs from the retinae of the two eyes are crossed approximately 50%, and the optic tracts terminate in the lateral geniculate ganglia in six discrete laminae, 1, 4, and 6, the sites of termination of the pathways from the contralateral retina, and 2, 3, and 5, the laminae supplied by the ipsilateral eye. The cells in these laminae project via the geniculocalcarine tracts to the ipsilateral striate cortex in such a way that the two retinal outputs for each point in the visual field converge to two adjacent cortical columns, one for each eye, for the same spot in the visual field. These are the ocular dominance columns, first described by Hubel and Wiesel (1968), and demonstrated autoradiographically by the [^{14}C]deoxyglucose method (Kennedy et al. 1976). The visual system of the newborn monkey exhibits plasticity. Chronic occlusion of one eye in a newborn monkey leads to widening of the ocular dominance columns for the functioning eye at the expense of the columns for the deprived eye until eventually the columns disappear, and the entire striate cortex is taken over to subserve the function of the undeprived eye (Hubel et al. 1977; Des Rosiers et al. 1978). Presumably the axonal terminals of the geniculocalcarine pathway for the functioning eye grow into, and take over, the synaptic connections in the adjacent column normally reserved for the deprived eye. If so, changes in protein synthesis required for axonal growth and sprouting could be involved, and the protein synthesis used for this process occurs in the cell bodies of origin of the pathway, i.e., in the lateral geniculate ganglia. Illustrated in Fig. 6 are the results of application of the local protein synthesis technique to this question. The laminae in the geniculate bodies are clearly visible and relatively uniform in the autoradiographs of the 25-day-old normal monkey. Acute monocular deprivation for 3 h does not alter the rates of protein synthesis in any of the laminae, including those for the deprived eye. Chronic monocular deprivation from 2 days to 25 days of age results in marked reductions in



protein synthesis in the laminae normally served by the deprived eye. Apparently chronic reduction in functional activity, in contrast to the acute state, results in a lowering of protein synthesis in the affected pathway. These results suggest that the loss of ocular dominance columns in the striate cortex for the chronically deprived eye is the result of depressed protein synthesis and deficient axonal growth in the geniculocalcarine pathways for the deprived eye.

It is hoped that measurement of local rates of protein synthesis in the nervous system may be useful for the study of normal and abnormal processes that may not be accessible to other autoradiographic and tomographic procedures, processes such as growth, maturation, plasticity, and actions of hormones. The full potential of this approach must, however, await the development of an accurate and reliable method.

References

- Agranoff BW, Smith CB, Sokoloff L (1980) Regional protein synthesis in rat brain after hypoglossal axotomy. *Trans Am Soc Neurochem* 11: 95
- Des Rosiers MH, Sakurada O, Jehle J, Shinohara M, Kennedy C, Sokoloff L (1978) Functional plasticity in the immature striate cortex of the monkey shown by the [^{14}C]deoxyglucose method. *Science* 200: 447–449
- Duffy TE, Cavazzuti M, Gregoire NM, Cruz NF, Kennedy C, Sokoloff L (1979) Regional cerebral glucose metabolism in newborn beagle dogs. *Trans Amer Soc Neurochem* 10: 171
- Frackowiak RSJ, Lenzi G-L, Jones T, Heather JD (1980) Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man using ^{15}O and positron emission tomography. *J Comput Assist Tomogr* 4: 727–736
- Gooch C, Rasband W, Sokoloff L (1980) Computerized densitometry and color coding of [^{14}C]deoxyglucose autoradiographs. *Ann Neurol* 7: 359–370
- Hawkins R, Phelps M, Huang SC, Kuhl D (1981) Effect of ischemia upon quantification of local cerebral metabolic rates for glucose with 2-(F-18)fluoro-deoxyglucose (FDG). *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S9–S10
- Hers HG (1957) *Le métabolisme du fructose*. Editions Arscia, Bruxelles, p 102
- Huang SC, Phelps ME, Hoffman EJ, Sideris K, Selin CJ, Kuhl DE (1980) Noninvasive determination of local cerebral metabolic rate of glucose in man. *Am J Physiol* 238: E69–E82
- Hubel DH, Wiesel TN (1968) Receptive fields and functional architecture of monkey striate cortex. *J Physiol* 195: 215–243
- Hubel DH, Wiesel TN, LeVay S (1977) Plasticity of ocular dominance columns in monkey striate cortex. *Phil Trans R Soc Lond B* 278: 377–409
- Kennedy C, Des Rosiers MH, Sakurada O, Shinohara M, Reivich M, Jehle JW, Sokoloff L (1976) Metabolic mapping of the primary visual system of the monkey by means of the autoradiographic [^{14}C]deoxyglucose technique. *Proc Natl Acad Sci USA* 73: 4230–4234
- Kennedy C, Sakurada O, Shinohara M, Jehle J, Sokoloff L (1978) Local cerebral glucose utilization in the normal conscious macaque monkey. *Ann Neurol* 4: 293–301
- Kennedy C, Suda S, Smith CB, Miyaoka M, Ito M, Sokoloff L (1981) Changes in protein synthesis underlying functional plasticity in immature monkey visual system. *Proc Natl Acad Sci USA* 78: 3950–3953
- Lassen NA, Ingvar DH, Skinhoj E (1978) Brain function and blood flow. *Sci Amer* 239: 62–71
- Phelps ME, Huang SC, Hoffman EJ, Selin C, Sokoloff L, Kuhl DE (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with (F-18)2-fluoro-2-deoxy-d-glucose: validation of method. *Ann Neurol* 6: 371–388
- Reivich M, Kuhl D, Wolf A, Greenberg J, Phelps M, Ido T, Casella V, Fowler J, Hoffman E, Alavi A, Som P, Sokoloff L (1979) The [^{18}F]fluoro-deoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
- Schuijer F, Orzi F, Suda S, Kennedy C, Sokoloff L (1981) The lumped constant for the [^{14}C]deoxyglucose method in hyperglycemic rats. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S63
- Smith CB, Davidsen L, Deibler G, Patlak C, Pettigrew K, Sokoloff L (1980) A method for the de-

Fig. 6A–D. Autoradiographs of coronal sections of monkey lateral geniculate nuclei (LGN) obtained with the [^{14}C]leucine method. The left side of the brain is on the *left side* of the autoradiographs. **A** Twenty-five-day-old monkey with intact binocular vision. **B** Twenty-five-day-old monkey with acute occlusion of the left eye. **C** Twenty-five-day-old monkey with chronic occlusion of right eye initiated on

2nd day of life. **D** Twenty-five-day-old monkey with chronic occlusion of left eye initiated on 2nd day of life. Note the decreased labeling and, therefore, rate of protein synthesis in the laminae 1, 4, and 6 of the lateral geniculate ganglion contralateral to the deprived eye and in laminae 2, 3, and 5 of the ganglion ipsilateral to the deprived eye (Kennedy et al. 1981)

- termination of local rates of protein synthesis in brain. *Trans Am Soc Neurochem* 11: 94
19. Sokoloff L (1978) Local cerebral energy metabolism: its relationships to local functional activity and blood flow. In: Purves MJ, Elliott K (eds) *Cerebral vascular smooth muscle and its control*, Ciba Found Symp 56: 171–197
 20. Sokoloff L (1979) The [^{14}C]deoxyglucose method: four years later. *Acta Neurol Scand [Suppl 70]* 60: 640–649
 21. Sokoloff L (1981) Localization of functional activity in the central nervous system by measurement of glucose utilization with radioactive deoxyglucose. *J Cereb Blood Flow Metabol* 1: 7–36
 22. Sokoloff L (1982) The radioactive deoxyglucose method. Theory, procedure, and applications for the measurement of local glucose utilization in the central nervous system. In: Agranoff BW, Aprison MH (eds) *Advances in neurochemistry*, vol. 4. Plenum, New York, pp 1–82
 23. Sokoloff L, Reivich M, Kennedy C, Des Rosiers MH, Patlak CS, Pettigrew KD, Sakurada O, Shinohara M (1977) The [^{14}C]deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure, and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
 24. Suda S, Shinohara M, Miyaoka M, Kennedy C, Sokoloff L (1981) Local cerebral glucose utilization in hypoglycemia. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S62

Experimental Application of Triple-Labeled Quantitative Autoradiography for Measurement of Cerebral Blood Flow, Glucose Metabolism, and Protein Biosynthesis

By G. Mies, W. Bodsch, W. Paschen, and K.-A. Hossmann

Introduction

During the past years autoradiographic techniques for measuring blood flow (Reivich et al. 1969; Sakurada et al. 1978) and metabolic activity (Sokoloff et al. 1977) have been of increasing interest in the study of various physiological and pathophysiological states. As long as the experimental procedure can be standardized, correlation of the different parameters is possible using single-label autoradiography, applied to different animals. However, considerable difficulties may be encountered when the parameters of interest change in an unpredictable way. In this case, simultaneous assessment of flow and metabolism in the same brain is a prerequisite for a meaningful evaluation.

Recently, double-tracer techniques for the measurement of cerebral blood flow and cerebral glucose utilization have been described (Lear et al. 1981; Mies et al. 1981a), using two isotopes with different half-life and energy spectrum. This technique has been validated and applied under a variety of pathological conditions such as experimental tumor development, brain infarction, spreading depression, and penicillin seizures (Mies et al. 1981b). A combination is possible with previously described fluorescence and bioluminescence methods for the assessment of biochemical substrates on intact brain slices (Paschen et al. 1981a). The additional autoradiographic assessment of regional protein biosynthesis requires the introduction of a third isotope. In the present investigation, the methodological requirements of triple-labeled autoradiography will be described and validated in various experimental situations. A more detailed description of this technique will be given elsewhere (Bodsch et al. to be published).

Methods

Theoretical Considerations

The objective of the present approach is the differentiation of three isotopes in such a way that three single-tracer autoradiograms with minimal contamination by the other tracers are obtained. The following isotopes are used: ^{131}I -iodo-antipyrine for measurement of cerebral blood flow, ^{14}C -deoxyglucose for the determination of glucose metabolism, and a mixture of five ^3H -amino acids for the assessment of protein biosynthesis.

^{131}I and ^{14}C are differentiated as has been described before (Mies et al. 1981a). In short, specific activity of ^{131}I -iodo-antipyrine is so high that the short-time exposure to an X-ray film for 24 h results in blackening of the film by the γ -emitting isotope without significant contamination by the other tracers. After 8–10 weeks ^{131}I has decayed. Autoradiograms obtained from a second exposure to a Kodak NMB film for 2 weeks represent ^{14}C radioactivity only because the gelatine protection layer of the film prevents significant contamination by tritium. Subsequently, the sections are washed for removal of ^{14}C -deoxyglucose and ^{14}C -deoxyglucose-6-phosphate. This procedure will also remove free ^3H -amino acids. A third exposure of the same sections for 2 weeks to a tritium-sensitive film (LKB) allows the selective visualization of incorporated amino acids, and represents protein biosynthesis.

Experimental Procedure

Animal Preparations

Triple-tracer autoradiographic measurements, and bioluminescent and fluorescent techniques were carried out in 20 BD IX rats and six stroke-prone spontaneously hypertensive rats

(courtesy of Prof. Yamori, Izumo, Japan). Animals were anesthetized with 0.8% halothane or by an intraperitoneal dose of 50 mg/kg pentobarbital (Nembutal). Both femoral arteries and veins were catheterized for arterial blood sampling, blood pressure monitoring, and application of drugs or tracers. After tracheotomy, rats were immobilized and artificially ventilated. Arterial blood gases were kept within physiological limits. Body temperature was adjusted to 37°C by means of controlled heating.

Experimental Groups

Control Group (Eight Rats). In four awake rats surgical procedure was restricted to cannulation of the four femoral vessels. Animals were allowed to recover from light halothane anesthesia for 2 h, and partially immobilized by means of loose-fitting plaster casts. The effect of halothane and barbiturate anesthesia was studied in two rats, respectively. Animals were prepared as described above.

Cortical Cryogenic Injury (Six Rats). Cortical cold lesions were performed according to Klatzo et al. (1980) 4, 9, 32, and 64 h prior to the autoradiographic study.

Brain Tumors (Five Rats). Experimental brain tumors were produced by stereotactical implantation of a glial cell clone into the right caudate putamen, with the rat under barbiturate anesthesia. After 3–4 weeks, animals developed spherical tumors at the implantation site. Investigations were carried out only in animals without neurological symptoms.

Stroke-Prone Spontaneously Hypertensive Rats (Six Rats). Animals were kindly provided by Prof. Yamori, Izumo, Japan. All rats exhibited cerebral infarctions with typical neurological symptoms, such as drowsiness and spastic paresis.

Experimental Schedule

In order to measure simultaneously cerebral glucose utilization, protein synthesis, blood flow, and the concentrations of glucose, ATP,

and NADH, experiments were carried out in the following way:

Time Course of Tracer Application

At the beginning of the experiments, a bolus of 10 $\mu\text{Ci}/100\text{ g}$ ^{14}C -deoxyglucose (spec. activity 300–350 mCi/mmol, NEN chemicals, Dreieichhain, FRG) was given intravenously for assessment of local glucose metabolism (Sokoloff et al. 1977). Arterial blood samples were collected at increasing intervals from 20 to 5 min immediately after injection. After centrifugation, plasma was stored on ice for determination of plasma ^{14}C -deoxyglucose and glucose concentrations.

For the determination of protein biosynthesis, 25 min later a mixture of five tritium-labeled amino acids was infused over 1 min followed by repeated arterial blood sampling to measure the ^3H -amino acid plasma specific activity profile (Bodsch and Hossmann, to be published). Forty-five minutes after the onset of measuring glucose consumption, cerebral blood flow was determined with ^{131}I -iodo-antipyrine according to the original autoradiographic method described by Sakurada et al. (1978). One hundred microcuries per 100 g ^{131}I -iodo-antipyrine (specific activity 7.45 mCi/mg; unbound iodide <2%; radiochemical purity >98%; NEN Chemicals, Dreieichhain, FRG) was infused over 1 min while arterial blood sampling was performed every 10 s. After 60 s, conscious animals were decapitated. The brains were dissected out as rapidly as possible and frozen in methylbutane cooled to -70°C . In anesthetized rats, the heads were frozen in situ in liquid nitrogen, starting 55 s after onset of ^{131}I -iodo-antipyrine infusion in order to allow the assessment of regional substrate concentrations of glucose, ATP, and NADH.

Measurement of Tracer Tissue Concentrations

Four different ^{131}I brain tissue standards were prepared with increasing doses of ^{131}I -iodo-antipyrine (100–550 μCi), which were allowed to circulate for 2 min. Brains were removed and sectioned into corresponding anatomical regions. The other hemisphere was frozen in -70°C cooled methylbutane while the dissected tissue was weighed and counted in a gamma

counter (Beckman) for the content of ^{131}I radioactivity per gram brain tissue.

A similar procedure was applied for quantitative ^{14}C autoradiography, using increasing amounts of ^{14}C -deoxyglucose per animal (20–100 μCi). The tracer was allowed to be incorporated for 45 min. In homologous tissue samples, ^{14}C concentration per gram brain was determined in one hemisphere by digestion in 0.3 ml hyamine hydroxide, and measurement of ^{14}C radioactivity with a scintillation counter (Beckman). Since the ^{14}C brain standards were equivalent to the experimental brains, they were also used to test different washing solutions for the removal of ^{14}C -deoxyglucose and ^{14}C -deoxyglucose-6-phosphate according to the strategy of triple tracer autoradiographic technique, described above.

Brains, ^{14}C , and ^{131}I standards were cut into 20- μm -thick sections in a cryostat (SLEE, Mainz, FRG) at -20°C , picked up by object holders, and dried immediately on a hot plate at 60°C to prevent diffusion of the tracers. Brain sections and tissue samples were also taken for the determination of biochemical substrates and quantitative assessment of protein biosynthesis (see below).

Brain sections and four different ^{14}C and ^{131}I standards were placed in a X-ray cassette and exposed to a NMB film for 24 h. Thereafter, brain sections were stored at 0°C until ^{131}I had decayed. After 8–10 weeks, the absence of ^{131}I in the sections was ascertained by repeated exposure of ^{131}I standards to X-ray film. Then the brain slices were exposed for the second time in order to carry out ^{14}C autoradiography. Thereafter, ^{14}C tracer and free ^3H -amino acids were removed from the sections using a washing solution, and sections were exposed to a tritium-sensitive film (LKB) for visualization of incorporated ^3H -amino acids into proteins.

Quantitative Evaluation of Cerebral Blood Flow

Calculation of cerebral blood flow was performed according to Sakurada et al. (1978), by

$$C_i^*(T) = \lambda \times K \int_0^T C_p^* e^{-K(T-t)} dt$$

where C_p^* is the arterial plasma concentration of ^{131}I -iodo-antipyrine, λ is the tissue blood partition coefficient (which is 0.8 for iodo-antipyrine), and $C_i^*(T)$ is the regional concentration of ^{131}I -iodo-antipyrine in brain tissue. Blood samples were counted during the first exposure period and expressed as nCi/ml blood, correcting the weighed probes for volume with $\sigma = 1.06$.

Quantitative Evaluation of Cerebral Glucose Utilization

Local glucose consumption was calculated according to the equation given by Sokoloff et al. (1977):

$$R_i = \frac{C_i^*(T) - k_1^* e^{-(k_2^* + k_3^*)T} \int_0^T C_p^* e^{(k_2^* + k_3^*)t} dt}{(\text{LC}) \times \left[\int_0^T C_p^*/C_p dt - e^{-(k_2^* + k_3^*)T} \int_0^T C_p^*/C_p e^{(k_2^* + k_3^*)t} dt \right]}$$

where C_p^* is the arterial plasma content of ^{14}C -deoxyglucose and C_p is the plasma glucose. LC is a lumped constant which was determined with 0.483. The following rate constants were used: $k_1^* = 0.189$, $k_2^* = 0.245$, and $k_3^* = 0.052$ for gray matter; and $k_1^* = 0.079$, $k_2^* = 0.133$, and $k_3^* = 0.02$ for white matter of the rat brain.

Quantitative Cerebral Protein Biosynthesis

The procedure for the quantitative evaluation of cerebral protein synthesis and the kinetical and mathematical treatment will be given in detail elsewhere (Bodsch and Hossmann, to be published). Briefly, the local rate of protein synthesis was determined after a 20-min period of incorporation of five tritium-labeled amino acids into proteins (^3H -amino acid mixture: phenylalanine, tyrosine, isoleucine, leucine, and methionine; total: 0.5 mCi/100 g). Between two brain sections microtissue samples were taken from various regions of the brain and analyzed for their content of free amino acids, aminoacyl tRNAs and proteins. Regional rates of protein synthesis (PSR) were expressed

as nanomoles of total amino acid incorporation/g per minute and calculated according to the equation

PSR nmol/g per minute =

$$\frac{q \times C_{pi}^* (T) \times mw_{aa}}{(C_{pi}/C_p) \times \int_0^T (C_{bi}^*/C_b) (t) e^{-(k_1+k_3+k_5)t} dt}$$

where C_{pi}^* is the radioactivity in the protein-bound amino acid i (dpm/g); C_{pi} is the concentration of the amino acid in the protein (nmol/g), and C_p the total amount of all amino acids in the protein (mmol/g). $(C_{bi}^*/C_b) (t)$ is the specific activity of the amino acid in the blood pool. k_1 , k_3 , and k_5 are the experimentally determined rate constants (min^{-1}) of amino acid influx, aminoacylation of tRNA, and incorporation of amino acid from aminoacyl tRNAs into proteins, respectively. $\overline{mw}_{aa}$ is the average molecular weight of amino acids, q is the amino acid distribution factor.

Regional Demonstration of Glucose and ATP Content

Brain sections adjacent to the ones used for autoradiography were processed for bioluminescent evaluation of glucose (Paschen et al. 1981b) and ATP concentration (Kogure and Alonso 1978). The cryostat sections were covered with frozen enzyme solutions which contained the necessary enzymes and co-enzymes for specific reaction with the substrate. After thawing the brain sections, the resulting light emission was recorded on a photographic film. Regional decrease of ATP and glucose was interpreted as indicative for regional deficiency of energy metabolism.

Regional Demonstration of NADH Fluorescence

The method used was described by Welsh and Rieder (1978). The tissue block from which brain sections were taken was illuminated in the cryostat with ultraviolet light at 360 nm and the fluorescence of tissue was photographed.

An increased intensity is emitted from regions with high concentrations of NADH, and reflects disturbances of the redox state of the brain.

Results

Validation of Isotope Separation

Validation of autoradiographic separations of ^{131}I and ^{14}C has been carried out recently (Mies et al. 1981b). As shown in Fig. 1, immediate exposure for 24 h of standard containing ^{131}I and ^{14}C resulted mainly in blackening of the film by γ -radiation. Eight to 10 weeks later, a second exposure for 14 days revealed only ^{14}C but no ^{131}I radioactivity. Figure 2 demonstrates the removal of ^{14}C from brain sections of animals which had received ^{14}C -deoxyglucose 45 min earlier. More than 90% of the ^{14}C concentrations could be washed out, resulting in negligible cross contamination during subsequent ^3H exposure. Using this procedure, three autoradiograms were obtained from the same brain section, and evaluated like single-tracer autoradiograms.

Animal Experiments

Control Experiments

In Fig. 3, the autoradiographic representation of cerebral blood flow, glucose utilization, and protein biosynthesis of the same brain section is shown. Coupling of flow, glucose metabolism, and protein biosynthesis generally is very close. Only in certain regions such as the pyramidal layer of the hippocampus, blood flow does not follow the metabolic pattern. When glucose consumption is compared with protein biosynthesis, it can be demonstrated that the location of protein synthesis is in the pericarya while glucose metabolism is increased in regions with high density of dentritic endings. For example, in the fascia dentata, the granular cells are clearly demarcated in the protein biosynthesis autoradiogram while glucose consumption is relatively low. On the other hand, the molecular layer of the hippocampus exhib-

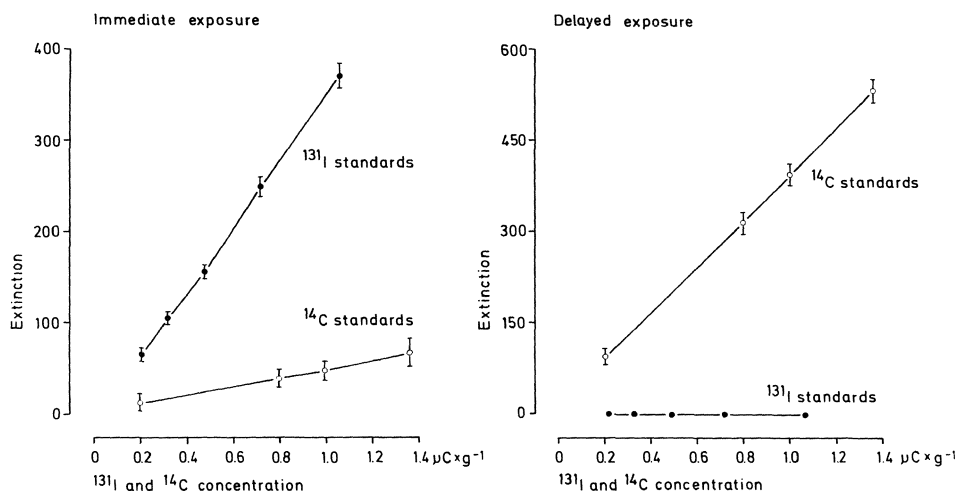


Fig. 1. Autoradiographic differentiation of ^{131}I and ^{14}C by means of standards. The first exposure was 24 h, representing almost exclusively ^{131}I radioactivi-

ty. Eight weeks later ^{131}I has decayed and blackening of the film results from ^{14}C only

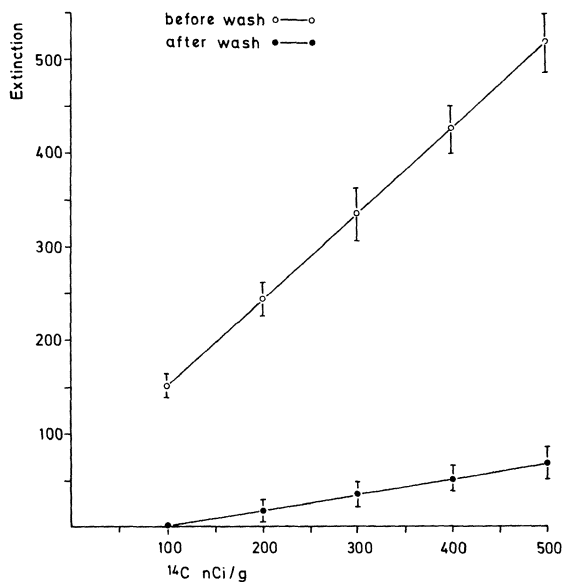


Fig. 2. Comparison of the ^{14}C content in brain sections before and after the removal of ^{14}C -deoxyglucose and ^{14}C -deoxyglucose-6-phosphate. ^{14}C radioactivity is expressed in optical density units (m OD) obtained after an exposure time of 3 weeks

its a high metabolic rate of glucose in comparison to the much lower protein biosynthesis.

Cold Injury of Cerebral Cortex

In Fig. 4, the relationship between blood flow, glucose consumption, and protein biosynthesis is demonstrated 4 h after cortical cryogenic injury. In the injured cortex, blood flow has arrested, and consequently a breakdown of glu-

cose and protein metabolism occurred. Reactive hyperemia demarcated the necrotic part of the lesion and extended into the cortex of the opposite hemisphere. This increase in blood flow, however, was not followed by a similar increase of metabolic rates. Between the zone of the necrotic lesion and the intact tissue, a small rim of increased glucose metabolism and protein biosynthesis was present which was not coupled to a similar rise in blood flow. It has

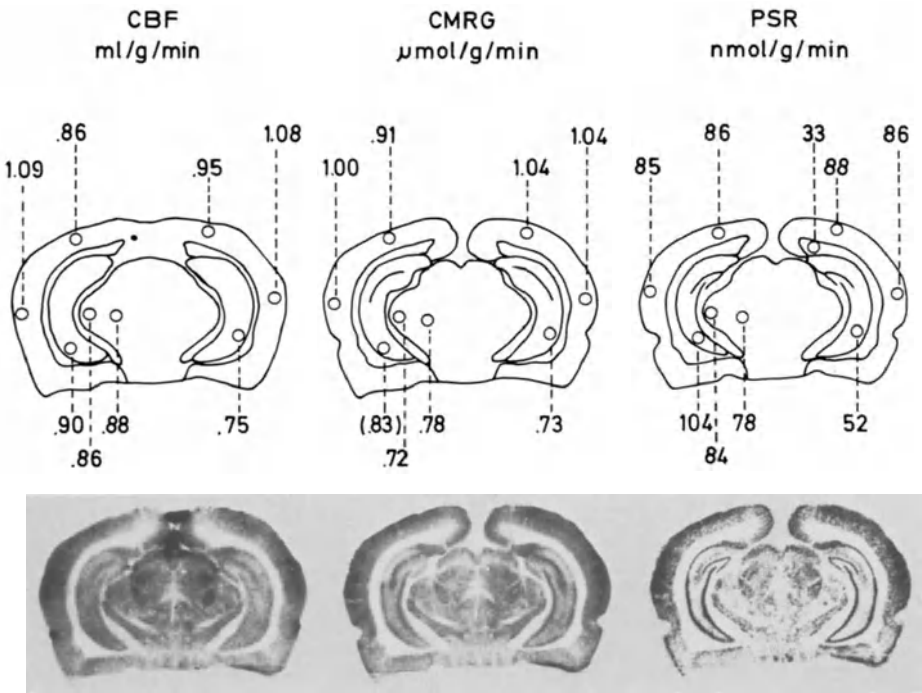


Fig. 3. Quantitative triple tracer autoradiography of blood flow with ^{131}I -iodo-antipyrine, glucose utilization with ^{14}C -deoxyglucose, and protein biosynthesis with a mixture of ^3H -labeled amino acids in an animal anesthetized with halothane. Autoradiograms are obtained from the same brain section

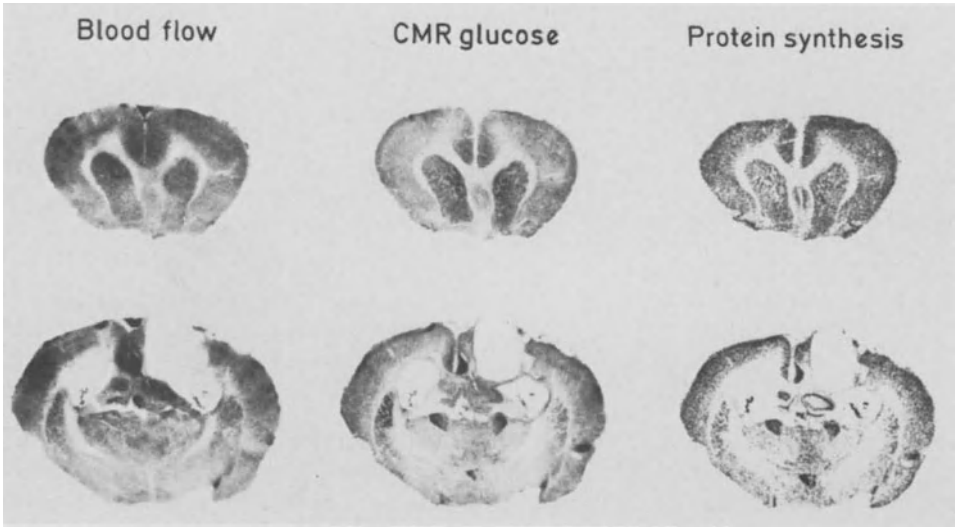


Fig. 4. Triple tracer autoradiography in a rat 4 h after cortical cryoinjury. Note the drastic increase of blood flow in the cortex which is not coupled to a similar rise in glucose and protein metabolism

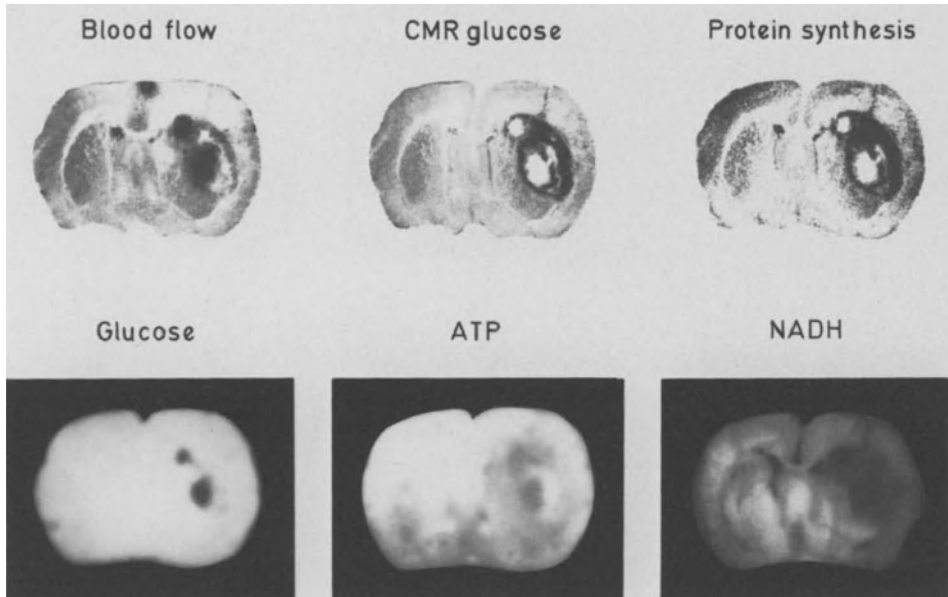


Fig. 5. Triple tracer autoradiography of blood flow, glucose metabolism, and protein synthesis combined with bioluminescent techniques for the regional assessment of glucose and ATP and with fluorescent technique for the regional demonstration of NADH

in experimental brain tumor of the rat. Note the high glucose utilization in the tumor due to stimulation of anaerobic glycolysis. ATP bioluminescence reveals a beginning failure of energy production in the tumor

been shown before that this is the region where the blood-brain barrier breaks down and from where edema fluid spreads into the surrounding white matter (Blasberg et al. 1980; Bodsch et al. 1982).

Brain Tumors

Following intracerebral implantation of clonized rat glioma cells into the right caudate putamen, spherical tumors developed which frequently exhibited a necrotic center as revealed by regional assessment of glucose and protein metabolism (Fig. 5). Increased ^{131}I -iodo-antipyrine radioactivity in this region reflects blood volume rather than blood flow because in the tumors large sinusoidal vessels are present. In the solid parts of the tumor a distinct increase of glucose consumption and protein biosynthesis could be observed which was not coupled to a similar increase of blood flow. It is a well-known fact that glycolysis of tumor tissue is preferentially anaerobic. The limiting factor for the metabolic activity in the tumors, therefore, is glucose rather than oxygen availability. Since

anaerobic glycolysis yields only $2\text{ }\mu\text{mol ATP}/\mu\text{mol glucose}$, it is not surprising that the concentration of ATP was reduced. This did not interfere, however, with protein biosynthesis. The energy requirements for protein biosynthesis are about $0.1\text{ }\mu\text{mol ATP}$ at a protein biosynthesis rate of $50\text{ nmol/g per minute}$, which is only a small fraction of ATP production, even when anaerobic glycolysis is the preferred pathway of tumor energy metabolism.

Stroke-Prone Spontaneously Hypertensive Rats

In most regions, a close correlation existed between blood flow, glucose, and protein metabolism (Fig. 6). Only in the anterior lateral nucleus of the thalamus metabolic rates were not coupled to flow. A large cyst resulting from a chronic infarct was sharply demarcated against the surrounding tissue. Unlike in acute focal ischemia or cold injury, no rim of increased glucose metabolism and protein biosynthesis could be detected.

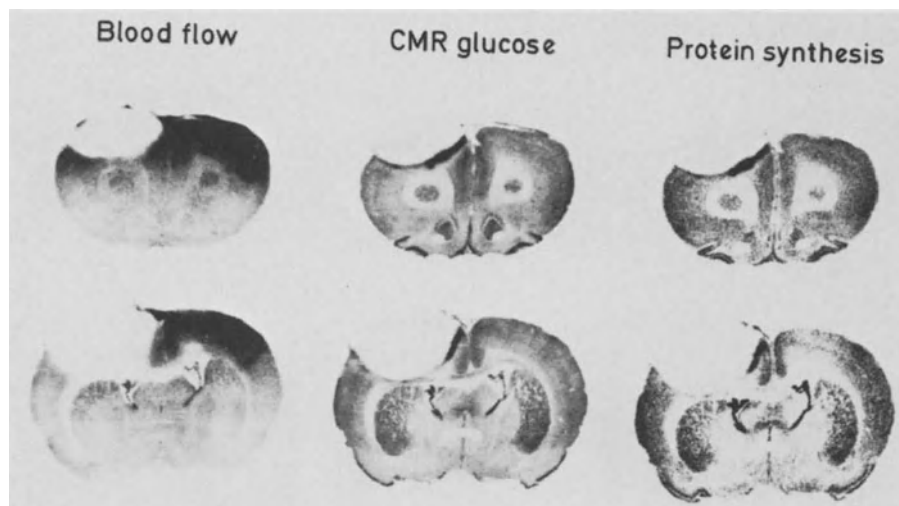


Fig. 6. Triple tracer autoradiography in a stroke-prone spontaneously hypertensive rat (courtesy of Prof. Yamori, Izumo, Japan). A large cyst results

from chronic infarction. Note the absence of hypermetabolism around the infarct

Discussion

In this study, a triple-labeled autoradiographic technique is presented for the simultaneous measurement of cerebral blood flow, glucose consumption, and protein biosynthesis in the same animal. With this method it is possible to correlate these three parameters on a regional basis because blood flow, glucose metabolism and protein synthesis can be assessed on the same brain section. Moreover, the combination of triple-labeled autoradiography with bioluminescent and fluorescent techniques for the assessment of biochemical substrates contributes further to the understanding of the pathophysiology of the brain.

However, there are certain methodological restrictions when this technique is applied under pathophysiological conditions. The separation of ^{131}I and ^{14}C does not involve any problems in normal animals because increase of blood flow is generally coupled to an increase of glucose consumption. However, interference of ^{131}I autoradiography by ^{14}C disintegrations may occur in areas in which flow is not coupled to stimulated glucose metabolism, i. e., in which ^{131}I -iodo-antipyrine concentration is low. In practice, this problem is of minor importance: as shown in Fig. 4, the small hypermetabolic

zone at the margin of cold lesion does not cause relevant ^{14}C contamination of the ^{131}I iodo-antipyrine autoradiogram.

The emitted low-energy particles of tritium should not present a major problem for cross-contamination of ^{14}C autoradiography. Only during prolonged exposure for ^{14}C , regions with high amounts of free or incorporated ^3H -amino acids could interfere, but tritium contamination can always be prevented by using thin plastic foils for the absorption of β -radiation. Under normal conditions, cross-contamination due to tritium is negligible, as shown in Fig. 3. For example, the granular layer of the fascia dentata is clearly demarcated in the ^3H autoradiogram whereas this structure is not present in the ^{14}C autoradiogram.

A more serious problem is possible cross-contamination by ^{14}C during the exposure for ^3H autoradiography. Tritium-sensitive films preferentially visualize low-energy radiation because these films are not protected by a gelatine layer. This reduces the ^3H self-absorption between the brain section and the photo emulsion, and enhances tritium sensitivity in relation to ^{14}C . However, since this does not prevent ^{14}C cross-contamination, ^{14}C -deoxyglucose and ^{14}C -deoxyglucose-6-phosphate were removed from the brain slices by a washing procedure. Exposure of such sections leads on-

ly to minimal blackening of the tritium-sensitive film by the remaining ^{14}C radioactivity.

The application of the triple-labeled autoradiographic technique involves a few methodological assumptions which must be considered. The methods used for measuring glucose consumption and amino acid incorporation require a certain period during which the metabolic substrates accumulate. Under physiological and steady-state pathophysiological conditions, the time lag between the assessment of flow and metabolic rate is probably not important, but it may introduce serious errors when non-steady-state phenomena are examined. Another aspect is the validity of the autoradiographic methods per se. For quantitative evaluation of glucose and protein synthesis, the kinetic constants must be known. Under pathophysiological conditions these constants may be determined in separate studies, assuming that the kinetics of the tracers always behave similarly under the same experimental setup. Finally, the measurement of blood flow requires an immediate arrest of systemic blood circulation. When triple-labeled autoradiography is combined with bioluminescent and fluorescent techniques, however, it is necessary to freeze the brain tissue *in situ* in order to prevent further metabolism of substrates, i.e., glucose and ATP. During this procedure, tissue concentration of ^{131}I -iodo-antipyrine may decrease in those regions of the brain in which circulatory arrest is delayed. Consequently, a slightly lower blood flow would be measured in such parts of the brain.

Even with these methodological limitations in mind, the advantage of the present technique for the simultaneous investigation of regional blood flow and metabolism is evident. In the normal brain, a close coupling of function, blood flow, and metabolism has been reported before. However, with the present technique it becomes evident that within a given brain region, e.g., the hippocampus, local blood flow, glucose metabolism, and protein biosynthesis are distinctly dissociated even under normal physiological conditions. This is not surprising as far as the dissociation between glucose utilization and incorporation of amino acids is concerned because the cellular sites of these metabolic activities are not the same. However, there is also a dissociation between hippocam-

pal blood flow and metabolic activity in the different cell layers. This may reflect a different regulation of blood flow in the hippocampus with the priority of arterial blood supply to the whole region rather than to anatomically defined structures.

The enormous advantage of multiparametric investigations in the same animal can also be demonstrated in experimental brain tumor studies. Brain tumors in general do not appear as a homogeneous tissue mass. Since the relationship between solid, mitotic, and necrotic parts of the tumors is not a constant one, measurements of global or regional investigations in different animals will fail to give relevant information. For example, the interpretation of the blood flow autoradiogram from Fig. 5 could lead to the assumption that a solid tumor grows in the right hemisphere. The comparison with the other parameters from this figure clearly shows that in the center of the tumor biochemical substrates have been used up and metabolic activities have ceased. Even the tumor mass itself exhibits a heterogeneous state of glucose and protein synthesis which locally is not always paralleled by the same increase of blood flow. This is apparently due to the stimulation of anaerobic glycolysis and explains the patchy appearance of ATP concentration around the tumor center.

Another important aspect for using triple-tracer autoradiography in the same animal is the possibility of studying the effect of brain lesions on the surrounding parenchyma, e.g., the respective border zones between pathological and healthy tissue, as well as the interaction on remote intact brain regions.

Following cortical cryogenic injury (Fig. 4), the hyperemia in the opposite cortex can be easily identified as "luxury perfusion", which is not coupled to a similar rise in glucose consumption or protein biosynthesis. On the other hand, glucose consumption and protein synthesis distinctly increase in a small rim around the necrotic center of the cold lesion in which blood flow has not changed. Such a dissociation, however, is not present around chronic infarcts in stroke-prone spontaneously hypertensive rats, indicating that coupling between flow and metabolism recovers in the chronic stage of brain lesions.

These findings underline the usefulness of re-

gional multiparametric methods for simultaneous assessment of several parameters in the same animal. Detailed analysis of the complex hemodynamic and metabolic processes during pathophysiological events require the use of methods with a maximum of resolution. As has been pointed out, coupling or uncoupling between flow and metabolism may vary even within the same pathological area. For a reliable evaluation and sensible interpretation of pathophysiological processes in the brain, triple tracer autoradiography combined with bioluminescent and fluorescent techniques, therefore, appears to be a most useful tool.

References

1. Blasberg RG, Gazendam J, Patlak CS et al. (1980) Quantitative autoradiographic studies of brain edema and a comparison of multi-isotope autoradiographic techniques. In: Cervós-Navarro J, Ferszt R (eds) *Advances in neurology*, vol 28: Brain edema. Raven, New York, pp 255–270
2. Bodsch W, Hossmann K-A (to be published) A quantitative regional analysis of amino acids involved in rat brain protein synthesis by high performance liquid chromatography. *J Neurochem* 40: 371–382
3. Bodsch W, Hürter T, Hossmann K-A (1982) Immunohistochemical method for quantitative evaluation of vasogenic brain edema following cold injury of rat brain. *Brain Res* 249: 111–121
4. Bodsch W, Mies G, Hossmann K-A (to be published) Simultaneous determination of local cerebral rates of protein synthesis, glucose utilization and blood flow by multi-autoradiographic techniques. *Proc Natl Acad Sci*
5. Klatzo I, Chui E, Fujiwara K et al. (1980) Resolution of vasogenic brain edema. *Adv Neurol* 28: 359–373
6. Kogure K, Alonso OF (1978) A pictorial representation of endogenous brain ATP by a bioluminescent method. *Brain Res* 154: 273–284
7. Lear JL, Jones SC, Greenberg JH, Fedora TJ, Reivich M (1981) Use of ^{123}I and ^{14}C in a double radionuclide autoradiographic technique for simultaneous measurement of LCBF and LCMRgl. *Stroke* 12: 589–597
8. Mies G, Niebuhr I, Hossmann K-A (1981a) Simultaneous measurement of blood flow and glucose metabolism by autoradiographic techniques. *Stroke* 12: 581–588
9. Mies G, Niebuhr I, Hossmann K-A (1981b) A double tracer autoradiographic technique for simultaneous measurement of cerebral blood flow and cerebral metabolism in rats. *Eur Neurol* 28: 188–193
10. Paschen W, Djuricic B, Bosma H-J et al. (1981a) Regional blood flow and regional distribution of biochemical substrates in experimental stroke of gerbils. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S 174–S 175
11. Paschen W, Niebuhr I, Hossmann K-A (1981b) A bioluminescence method for the demonstration of regional glucose distribution in brain slices. *J Neurochem* 36: 513–517
12. Reivich M, Jehle J, Sokoloff L et al. (1969) Measurement of regional cerebral blood flow with antipyrine- ^{14}C in awake cats. *J Appl Physiol* 27: 296–300
13. Sakurada O, Kennedy C, Jehle J et al. (1978) Measurement of local cerebral blood flow with iodo (^{14}C) antipyrine. *Am J Physiol* 234: H59–66
14. Sokoloff L, Reivich M, Kennedy C et al. (1977) The (^{14}C) deoxyglucose method for the measurement of local cerebral glucose utilization: Theory, procedure, and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
15. Welsh FA, Rieder W (1978) Evaluation of in situ freezing of cat brain by NADH fluorescence. *J Neurochem* 31: 299–309

Methodology

(Chairman: T. Jones)

New Design Concepts for Quantitative Positron Emission Computed Tomography of the Brain

By E. J. Hoffman, J. B. Barton, M. E. Phelps, and S.-C. Huang

The purpose of this chapter is to provide an analysis of some of the new design concepts for instrumentation in positron tomography as it specifically relates to studies of the brain. Since positron tomography is an imaging modality it is useful to have a picture of the state of the art of instrumentation and a picture of the possible improvement that could be provided by future instruments. This improvement can be seen in simulations of brain images, which were created by digitizing the gray and white matter distributions of actual brain slices, assigning isotope concentrations to the structures (a 4 to 1 gray/white ratio), and then imposing the resolution blurring and statistical noise of an actual tomograph upon this ideal data (Mazziotta et al. 1981; Phelps et al. 1982). Examples of such a simulation are shown in Fig. 1. The lower two images are comparable in resolution to present-

day tomographs with 9–12 mm FWHM resolution (Hoffman et al. 1981, 1982). The upper two images constitute the design goal for future positron tomography systems with intrinsic resolutions of 2–4 mm FWHM. The complexity and small size of many of the structures in the brain make it obvious that the highest possible resolution should be a primary design goal for any brain imaging system.

New Design Concepts

Image quality in positron tomography is primarily a function of the statistical signal to noise ratio in the data (number of counts per image) and the resolution. Present-day tomographs with multiple planes and collection of

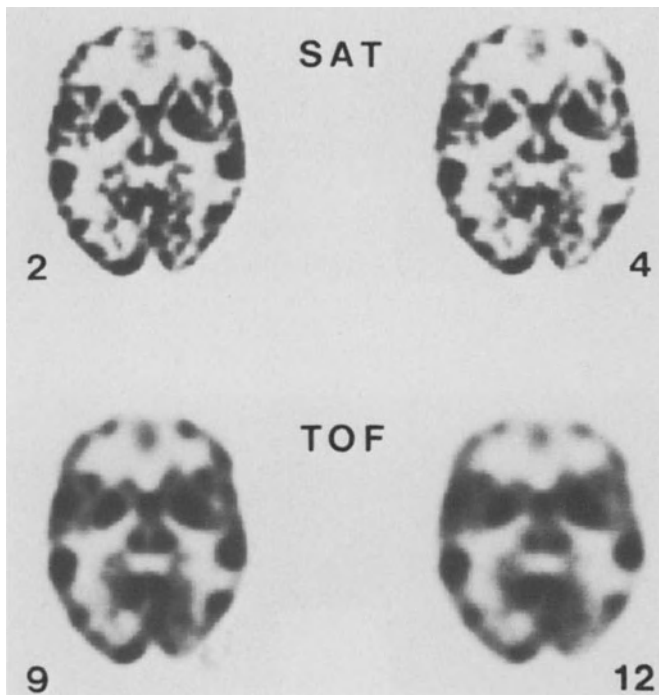


Fig. 1. Simulations of FDG brain scans in which the white and gray matter distribution of an actual brain slice is digitized with a video camera, the gray structures are set equal to a concentration of 4, the white regions are set equal to 1, and scan profile data are created from this close to perfect data set. The scan profile data are then blurred with the resolution spread of a hypothetical tomograph, and Poisson noise is added to simulate the statistics of an actual scan. In the upper two figures the intrinsic simulated resolutions are 2 and 4 mm with one million counts per image. In the lower images the intrinsic resolutions are 9 and 12 mm with 20 million counts per image

cross-plane data are accumulating a large fraction of the available events, and little improvement is available in terms of simply increasing system sensitivity. Therefore, new design concepts have been channeled in the direction of improving the quality of measurement of each event rather than attempting to increase the number of actual accumulated events. Two approaches to accomplishing this end are being actively pursued. These approaches are the time of flight technique (TOF) (Allemand et al. 1980; Mullani et al. 1980; Snyder et al. 1981) and the signal amplification technique (SAT) (Phelps et al. 1981; 1982).

Time of Flight

In positron tomography, the data set is a series of scans about a cross section as a function of angle. In the data set, highly detailed activity distributions are obtained along the direction of the scan profile and no information is obtained about the position of the activity between the two coincident detectors. In TOF a very fast detector (CsF) is used to time the difference in the arrival time of the annihilation photons at the two detectors, and since the photons travel at a known velocity (the speed of light) the time difference gives the position of the event between detectors. Unfortunately, the timing accuracy is presently in the order of 0.4 ns FWHM, which translates into 6 cm FWHM spatial resolution (Allemand et al. 1980). If the timing resolution were 10–20 times better, there would no longer be a need to perform a reconstruction since the data could simply be placed in the correct location from a knowledge of the detector pairs involved and the arrival time difference of the photons. The improved quality of the data in this limiting case would be equivalent to an increase in sensitivity of about a factor of 20 for an object the size of the human head.

In order to evaluate the possibility of achieving an improvement in resolution with CsF by a factor of 10–20, the resolution limit of CsF can be evaluated relative to the intrinsic properties of the material. These properties are the refractive index of the crystal and the decay time and yield of scintillation photons (Allemand et al. 1980). Since the refractive index of CsF for

scintillation light is about 1.5 in the crystal, the scintillation light travels at two-thirds the speed of light in a vacuum, and thus the timing depends on the depth of interaction of the annihilation photon. If the gamma ray interacts at the front surface of the crystal, the apparent arrival time of the photon is later than if the interaction is toward the back of the crystal because of the difference of the velocity of light in the crystal compared with the velocity of the gamma ray. The probability of the depth of interaction was estimated using the attenuation coefficient of CsF and the assumption of a single interaction per photon. The resolution spread caused by this phenomenon is shown in Fig. 2 for three thicknesses of crystal. The resolution spread due to photon yield and decay time is based on the statistical probability of the production of 1, 2, or 3 photoelectrons in the photomultiplier, assuming all time jitter is due to that time spread (Lynch 1966). The resolution spread due to this factor is shown for each of these cases in Fig. 2. Also included in Fig. 2 are possible net resolutions based on the combinations of timing and crystal thicknesses indicated. Since timing on one photoelectron and using a 3-cm thick crystal may be sacrificing too much efficiency and performance stability, the limit in resolution is more likely to be 2.7–2.8 cm or about 3 cm when the noise of the measurement electronics is added.

Since the TOF resolution is not adequate to directly measure the position of activity, the information from the time of flight is used to add additional constraints to the data to aid in the reconstruction (Snyder et al. 1981). The net effect of the TOF measurement is an improvement in the statistical quality of the image. For a 30-cm diameter uniform distribution of activity and 6-cm TOF resolution, the improvement in image quality is equivalent to a factor of 4.4 in system sensitivity. The improvement is proportional to the size of the activity distribution (the larger the distribution, the greater the improvement). The improvement is also proportional to the improvement in the TOF resolution. In the range under consideration here, the improvements are linearly related to these factors (Snyder et al. 1981).

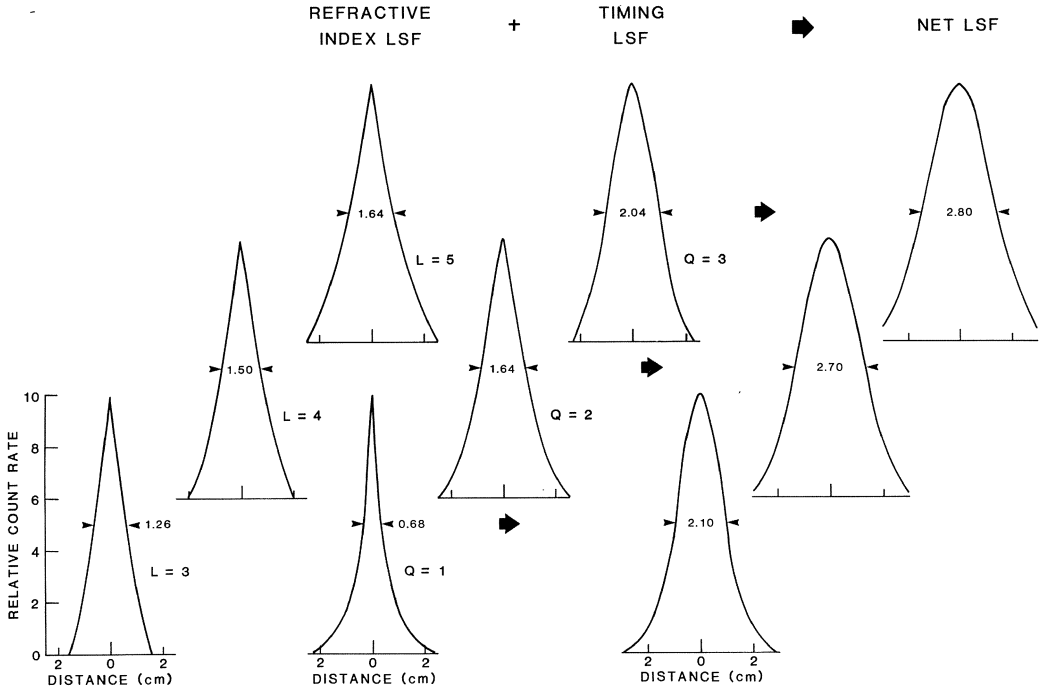


Fig. 2. Time of flight resolution limits for CsF. Estimates of the spatial resolution limits of TOF for CsF were made on the basis of the difference of the speed of light of the scintillation photons in the crystal relative to the speed of a gamma ray, which is unaffected by the refractive index of a medium. The line spread functions were calculated for 3-, 4-, and 5-cm-deep

(L) crystals. The resolution spread due to the statistics of the light emission was calculated assuming the time trigger was on the 1st, 2nd, or 3rd electron (Q). The crystal was assumed to have a 40% photofraction, and all events above 100 keV were included. The net LSFs are the convolution of the first and second LSF in each row

Signal Amplification Technique

The time of flight technique improves data quality by improving the localization of information along the line between detectors, while SAT improves data quality by improving the localization of information along the data profile by using very narrow detectors. That is, the high-frequency components (small objects) of the signal are amplified because they are detected much more clearly with the narrow detector. The detectors that are employed are much narrower than the expected resolution of the final image.

It has been the trend in tomography to try to have image resolution as close as possible to the resolution of the detection system. Thus, if the intrinsic resolution of a detector pair is 1 cm, the usual goal is 1.1 cm or better in the final image. The reconstruction technique required to obtain 1.1 cm resolution from a 1-cm

FWHM detector system will amplify spatial frequencies in the data that are just barely detected by a 1-cm FWHM detector system. Generally, most of the apparent data available at those frequencies are actually statistical fluctuations. Thus, the reconstruction technique is primarily amplifying noise at those frequencies. With SAT very narrow detectors are used and the reconstruction technique may only attempt to achieve one-half the detector resolution in the image. This means that the frequencies that are amplified by the reconstruction mathematics are only those that are strongly detected by the narrow detectors. The signal to noise ratio is very good at the cutoff frequency of the reconstruction filter, therefore, although the noise is still in the data, the signal has been amplified by the detector, and the noise has been suppressed by the relatively low cutoff frequency employed in the reconstruction.

As an example of the effect of SAT, consider

the consequences of modifying the detectors on the NeuroECAT (Hoffman et al. 1981) from 17 mm to 4 mm wide (there would be four times the number of detectors on the modified system). The resolution of the detector pairs would change from 10 mm to 2.5 mm. If the noise per pixel were required to be the same for both systems, the resolution of the 4-mm system would be 6 mm compared with 11.7 mm for the NeuroECAT. Essentially, the higher noise level that would have been anticipated for the higher resolution system was reduced by using a lower resolution reconstruction technique, which sacrificed some of the resolution to obtain a good signal to noise ratio. Alternatively, if 11.7-mm resolution is adequate, the 4-mm system can yield a 11.7 mm image with a signal to noise ratio equivalent to a system with three times the signal to noise ratio of the NeuroECAT. In many cases, the number of counts available will be adequate to give better than the 6-mm resolution stated above with a potential of achieving 3- to 4-mm FWHM resolution for images obtained with high statistics.

The SAT approach is possible because of a relatively new detector material called bismuth germanate (BGO) (Nestor and Huang 1975; Cho and Farukhi 1977). This detector has three times the stopping power for 511 keV photons of the NaI(Tl) detector (which is comparable to CsF in stopping power). This fact allows the localization of the event in a surprisingly narrow detector (Ricci et al. 1982; Derenzo 1981). Previous attempts to use narrow NaI(Tl) detectors (8 mm wide) in tomographs have led to redesigning of the tomographs because of low efficiency and scatter of the radiation between detectors making it difficult to accurately position each event (Derenzo et al. 1979; Eriksson et al. 1980). Some of the important properties of three narrow BGO detectors (2.5, 3.5, and 4.5 mm wide) are summarized in Table 1. The system efficiency was estimated by measuring the narrow detector efficiency relative to a pair of ECAT detectors and then calculating the total system efficiency. Within the experimental error, there is no loss of efficiency relative to 9.5-mm wide BGO detectors (Derenzo 1981) for the 4.5-mm detectors. There is a small loss with the 3.5-mm detectors and the loss is significant for the 2.5-mm detectors. In all cases crosstalk or scatter between crystals is small. In

Table 1. Physical properties of narrow BGO detectors

Property	Detector width		
	2.5 mm	3.5 mm	4.5 mm
Resolution (FWHM)	2.0 mm	2.4 mm	2.9 mm
Cross talk	8%	7%	2%
System efficiency ^a	68%	92%	100%

^a Relative to system with 9.5-mm-wide BGO detectors

summary, the BGO detector allows very good localization of each event, giving high resolution (2.4 mm) with very little loss in efficiency.

Comparison of TOF and SAT

In this comparison of the near term application of SAT and TOF, the estimates of the design parameters come from typical systems in the literature (Blaine et al. 1982; Mullani et al. 1982). In order to obtain the excellent timing characteristics required for TOF, developers of these systems have chosen cylindrical detectors with sizes matched to high-performance photomultipliers. The detector containers and reflector materials require a relatively large amount of space compared with BGO. CsF is very hygroscopic and requires a hermetically sealed environment. BGO resembles quartz, is just as inert, and only requires a light tight environment. Because of these factors, the current designs of TOF systems are employing relatively large detectors which are not very closely packed. The cylindrical shape and large dead spaces between the crystals of the TOF system and the lower stopping power of the CsF all reduce the efficiency of TOF systems relative to closely packed BGO systems.

In order to compare a TOF system with an SAT system, I have chosen the detector sizes and spacings of the TOF system being developed by Mullani et al. (1982). The intrinsic efficiencies of both the BGO and CsF detectors were taken from the work of Derenzo (Derenzo and Riles 1982). These physical parameters are listed in Table 2. It is assumed that the geometry of both systems is identical except for those differences required by the detector. The intrinsic efficiency of the BGO detector is given in the De-

Table 2. Physical and geometric efficiency factors: BGO VS CSF

Factor	BGO	CSF
Detector size (mm) (width $\times$ height $\times$ depth)	$4 \times 18 \times 30$	18 diameter $\times 50$
Intrinsic efficiency	0.71	0.68
Coincidence efficiency	0.504	0.462
Packing fraction	0.9	0.68
Geometric efficiency	0.81	0.428
Total efficiency	0.408	0.198

renzo work, while the efficiency used for the CsF detector is that of a larger more efficient crystal (20 mm $\times$ 30 $\times$ 50 mm). The intrinsic coincidence efficiency is given by the square of the intrinsic efficiency since both photons must register. The packing fraction is the fraction of the 18-mm high detector ring that is occupied by the detector. The geometric efficiency is the square of the packing fraction. The comparison of these sensitivities indicates that the BGO system is slightly more than twice the sensitivity of the CsF system.

This means that the TOF improvement must be at least a factor of 2 to break even with the sensitivity of the BGO system. The area containing the activity will be about equal to an 18-cm-diameter circle in a brain-imaging system; thus the TOF improvement will be $\frac{18}{30}$ times that of

the 30-cm circle noted above. The improvement was a factor of 4.4 for the 30-cm circle, giving a factor of 2.64 improvement for an 18-cm circle. Because of the factors in Table 2, the CsF already has a reduced sensitivity giving an apparent net improvement over the BGO system of only a factor of 1.28. This value applies to the present 6-cm TOF resolution. If the resolution of a system could approach the limit of 3 cm, the apparent sensitivity could double to 2.56.

It should be noted that the improvement was calculated by Snyder (Snyder et al. 1981) for a uniform distribution of activity. For other distributions, Snyder (1982) has shown the improvement to be smaller. The intrinsic efficiency value was for a larger crystal; the actual efficiency is smaller. Considering these factors, the effective sensitivity of the two designs is actually very similar for a brain-sized object.

Now consider the SAT effect of the 4-mm wide BGO detector. The intrinsic resolution of that detector is between 2 and 3 mm. The intrinsic resolution of an 18-mm-diameter detector is about 9 mm. The significance of this fact is seen in Fig. 1. The image in the lower left-hand corner has an intrinsic resolution of 9 mm and 20 million counts. The upper images have 2-mm and 4-mm FWHM intrinsic resolution and only one million counts. It is obvious that the upper images are far superior to the lower images, even though they only contain 5% of the counts.

The major difficulty of the TOF approach is that it is very difficult to attain the high resolution of the SAT approach. This is due to the stopping power of the CsF crystal, which will rapidly lose efficiency at significantly smaller sizes, and the stringent requirements on the crystal shape and size and the type of photomultiplier for a TOF system. Either type of change causes an effective efficiency loss either due to less stopping power or poorer timing characteristics. The SAT approach can give similar characteristics to TOF in terms of noise reduction, but it also has the flexibility of very high resolution when adequate counts are available.

To provide perspective on the effect of SAT in speed and quality of brain imaging, one can extrapolate from the experience with the NeuroECAT. Figure 1 is essentially a simulation of an FDG image and if 4-mm detectors were put in the NeuroECAT it would require about 1 min to accumulate a one-million-count image from a 10-mCi injection of FDG. Thus if one is willing to take 20 min on a scan, this quality of scan can be attained with only 5% of the uptake of FDG or if 12-mm resolution is adequate, high-quality scans can be achieved with activities in the order of 5 nCi/ml.

It is clear from these considerations that the SAT approach is appropriate for a positron tomograph designed for brain imaging. Also when one considers the actual size of the structures containing activity in a scan of any part of the body, it is apparent that these structures are rarely much larger in area than the brain. Consequently, the effectiveness of the TOF technique is still an open question even for the imaging of the torso.

Implementation of SAT

While the SAT approach to positron tomography may seem attractive from a theoretical point of view, it will require significant developmental work to provide an attractive method of implementation of SAT in actual practice. A typical tomograph will require about 400–500 narrow BGO detectors per detector plane and three to five planes. Typical photomultiplier tubes are much wider than the detectors; thus there is a difficult packing problem in coupling the photomultipliers to the detectors in an adequate way.

Presently Available Methods of Implementing SAT

Methods are available for implementing SAT with presently available technology. However, the general impression of the methods is that they are cumbersome, and in the long run there must be something better. In order to make SAT worthwhile for a brain system, the detector size must be significantly smaller than those used previously (less than 5 mm wide, 3 mm FWHM intrinsic resolution).

One possible method of achieving the above goal is shown in Fig.3. This technique employs 19-mm diameter photomultiplier tubes and 4.5-mm BGO detectors on 4.9-mm centers. Each photomultiplier is optically coupled to three crystals, sharing the outside crystals with the neighboring photomultipliers and viewing the central crystal alone. The crystal of interaction is identified by a simple coding scheme in which the central detector is identified by signals from single photomultipliers and the shared or outside crystals identified by signals from two adjacent photomultipliers. This system was analyzed by Ricci et al. (1982). While the system does work, it has the drawback that it is difficult to implement for more than two planes and the failure of a single photomultiplier affects the operation of five neighboring detectors, potentially causing serious artefacts.

Recently, a 10-mm diameter photomultiplier (Hamamatsu R1635) became available, which may be adequate for application to positron

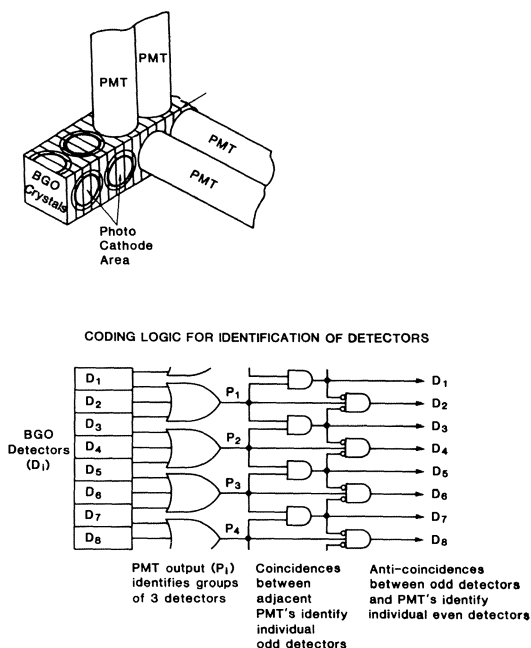


Fig.3. Diagram of one possible implementation of SAT. Each photomultiplier views three crystals, sharing each outside crystal with the adjacent photomultiplier. The crystal is identified as a shared crystal when signals are seen in adjacent photomultipliers and as unshared when only one photomultiplier has a signal. The possible physical arrangement and possible logic schema are indicated

tomography. This tube would have the advantage that one tube could be coupled to each crystal in a staggered array on the outside of the detector ring, allowing as many planes as desired, and the loss of a single tube would affect only the coincidences involving that detector. Of course this requires twice as many photomultipliers as the coding scheme described above. Additionally, the problem of adequate timing with such a small photocathode may degrade the performance of such a system. The problem of light output from BGO has been investigated theoretically by Derenzo and Riles (1982) and experimentally and theoretically by Barton et al. (1982). These investigators have concluded that significant increases in the light yield of BGO can be attained by simple optimization of detector shape, surface treatment, and reflective coating. Thus, the small photomultiplier is a viable approach for implementing SAT.

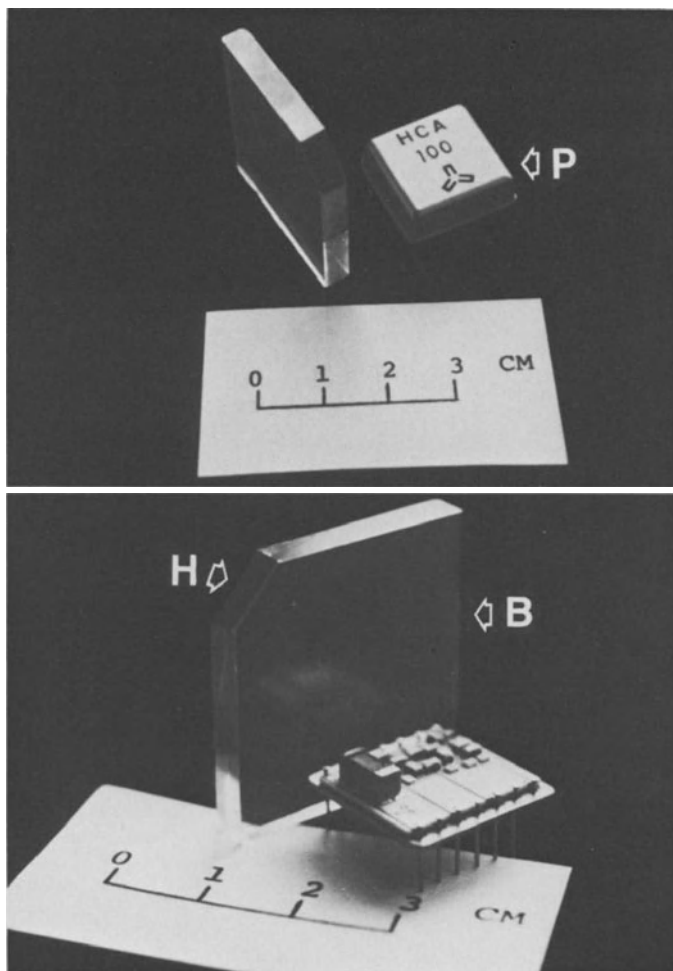


Fig. 4. Photograph of HgI_2 detector (H) coupled to a BGO crystal (B) and the low-noise preamp (P) with and without cover

Future Implementation of SAT

The above methods are not totally satisfactory solutions for SAT implementation. An alternative method was sought by investigators at UCLA for the identification of the crystal of interaction with the coincident gamma ray. The general line of investigation concentrated on the concept of a solid state photodiode or photodetector to identify the crystal and a large photomultiplier to provide the timing information for groups of detectors. The light sensitivity of most photodetectors was low, so the investigation also involved research into methods of improving the light yield of BGO. Most previous work in this area involved scintillators with relatively high light yield and very energetic particles (Bateman 1969 a, b, c; Bateman and

Ozsau 1973; Keil 1968; Webb and McIntyre 1976). Generally the leakage current and sensitivity of those materials generally considered to be photodetectors was not adequate for any application with BGO.

Attention was then turned to a material that was not generally considered to be a photodetector, but was generally employed as an X-ray and gamma ray detector, mercuric iodide (HgI_2). Investigators at the University of Southern California had recently developed techniques for manufacturing high-resolution X-ray detectors from HgI_2 (Dabrowski et al. 1981) and had developed very low-noise preamplifiers to accomplish this (Iwanczyk et al. 1981). HgI_2 is a crystalline material of very high resistivity (leakage current in the picoampere range as a detector) and very high absorption for the

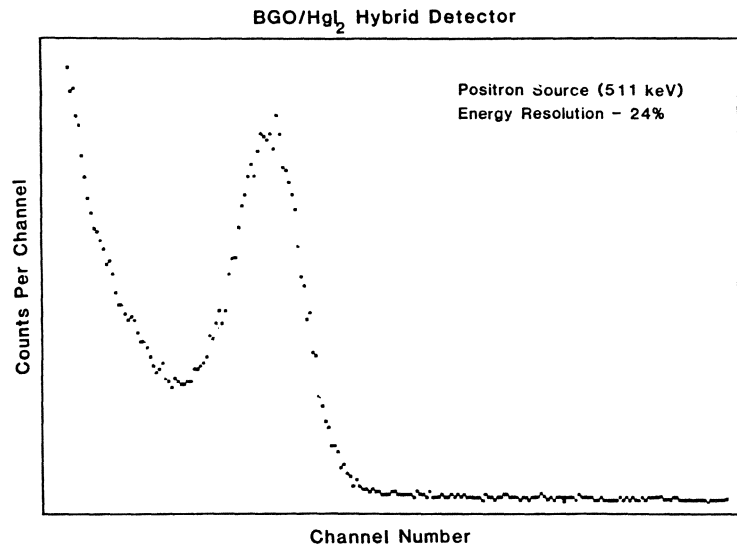


Fig. 5. Pulse-height spectrum from a hybrid HgI₂-BGO detector of the 511-keV gamma ray from a positron emitter

light emitted from the BGO crystal. The crystal has optimal resolution when it is thin and of small dimensions ideal for coupling to a narrow BGO detector.

Figure 4 shows photographs of the HgI₂ attached to a 4.5-mm-wide BGO crystal. The low-noise preamplifier is shown for size comparison. No effort has yet been made to miniaturize the preamplifier. Preliminary measurements with this system (Barton et al. 1982) show a surprisingly good energy resolution for 511-keV gamma rays on the first attempt, with no optimization of the coupling medium, detector size, or electronics. An energy spectrum from a BGO crystal as measured by a HgI₂ detector is shown in Fig. 5. Note that the energy resolution is 24%, which is almost as good as the 20% resolution generally anticipated for BGO with a photomultiplier. The timing resolution was poor, in the order of 100 ns. However, the task of the HgI₂ will only be to identify which of a small number of crystals was struck by a gamma ray. All the fast timing will be done by a photomultiplier. If there is no improvement in timing, data loss due to the timing of the HgI₂ will be only 10% of the loss experienced in the multiplexing logic of the ECAT (Phelps et al. 1978) or NeuroECAT (Hoffman et al. 1981) and those devices have very satisfactory performances in those areas.

Figure 6 shows a conceptual drawing of the basic detector unit employing the HgI₂ photo-detector. The unit consists of: (1) a relatively large photomultiplier coupled to several narrow BGO detectors. The photomultiplier will supply the fast signals for coincidence timing. (2) A set of HgI₂ detectors, with each detector coupled to an individual BGO detector with the signal transmitted to a remote preamplifier. The HgI₂ detector will identify the crystal of interaction. A positron tomograph for the brain would consist of about 40–50 of these units per ring. The resolution of such a unit will be limited only by the ability of the BGO to absorb the 511-keV photons within the first crystal that is struck. The HgI₂ detectors can easily be as thin as 2 mm, which is probably narrower than is reasonable when all other resolution degrading factors acting in the measurement are taken into consideration.

Conclusion

The work on the use of the HgI₂-BGO hybrid detector has just begun as a collaborative effort between investigators at UCLA and USC. There are many practical problems to be solved, but these problems seem to be ones of developing techniques rather than of basic lim-

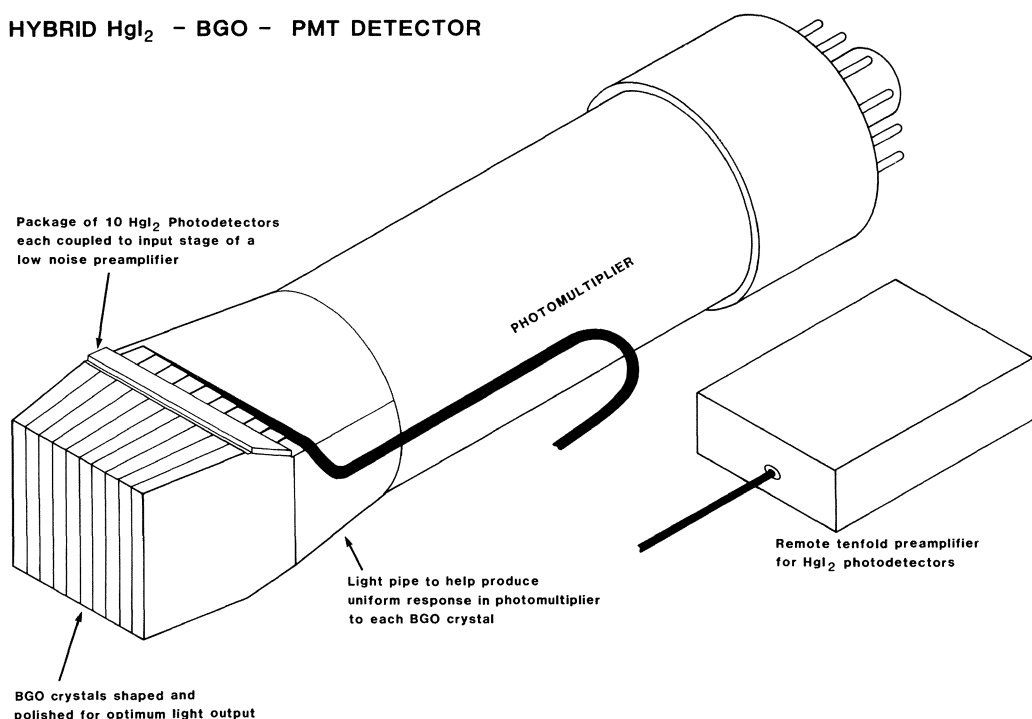
HYBRID HgI_2 - BGO - PMT DETECTOR

Fig. 6. Conceptual drawing of the basic HgI_2 -BGO detector unit for a positron tomograph. The unit consists of a relatively large photomultiplier for coinci-

dence timing for a group of BGO crystals and one HgI_2 photodetector per BGO detector to identify which detector was involved in the coincidence

itations in the method. This concept has the potential of pushing SAT and positron tomography to its limits. At least until someone develops a method to combine both SAT and TOF.

Acknowledgments. We wish to thank R. Carson, D. Plummer, A. Ricci, G. Low, and A. Meadors for technical assistance and M. Griswold for illustrations. This work was supported in part by Department of Energy contract DE-AM06-76-SF000012 and NIH grants USPH57-ROI-GM24839 and 1-PO1-ns15654.

References

1. Allemand R, Gresset C, Vacher J (1980) Potential advantages of cesium fluoride scintillator for a time of flight positron camera. *J Nucl Med* 2: 153-155
2. Barton JB, Hoffman EJ, Iwanczyk IS et al. (1982) High-resolution detection system for positron tomography. *IEEE Nuclear Science Symposium*, Washington, DC
3. Bateman JE (1969a) Some recent results with a photodiode-organic scintillator combination used as a detector for high-energy charged particles. *Nucl Instrum Meth* 71: 269
4. Bateman JE (1969b) Some new scintillator-photodiode detectors for high-energy charged particles. *Nucl Instrum Meth* 67: 93
5. Bateman JE (1969c) A solid state scintillation detector for high-energy charged particles. *Nucl Instrum Meth* 71: 261
6. Bateman JE, Ozsai FE (1973) A cryogenic scintillator-photodiode detector for penetrating charged particles. *Nucl Instrum Meth* 108: 403-407
7. Blaine J, Ficke D, Hitchens R et al. (1982) Data acquisition aspects of super-pett. *IEEE Trans Nucl Sci* NS-29: 544-547
8. Cho ZH, Farukhi MR (1977) New bismuth germanate crystal - a potential detector for positron camera applications. *J Nucl Med* 18: 840-844
9. Dabrowski AJ, Iwanczyk JS, Barton JB et al. (1981) Performance of room temperature mercuric iodide (HgI_2) detectors in the ultralow-energy X-ray region. *IEEE Trans Nucl Sci* NS-28: 536-540
10. Derenzo SE (1981) Monte Carlo calculations of the detection efficiency of arrays of NaI(Tl) ,

- BGO, CsF, Ge, and plastic detectors for 511-keV photons. *IEEE Trans Nucl Sci* NS-28: 131–136
11. Derenzo SE, Riles JK (1982) Monte Carlo calculations of the optical coupling between bismuth germanate crystals and photomultiplier tubes. *IEEE Trans Nucl Sci* NS-29: 191–195
 12. Derenzo SE, Budinger TF, Cahoon JL et al. (1979) The Donner 280-crystal high-resolution positron tomograph. *IEEE Trans Nucl Sci* NS-26: 2790–2793
 13. Derenzo SE, Budinger TF, Huessman RH et al. (1981) Imaging properties of a positron tomograph with 280 BGO crystals. *IEEE Trans Nucl Sci* NS-28: 81–89
 14. Eriksson L, Bohm C, Bergstrom C et al. (1980) One-year experience with a high-resolution ring detector positron camera system: present status and future plans. *IEEE Trans Nucl Sci* NS-27: 435–439
 15. Hoffman EJ, Phelps ME, Huang SC et al. (1981) A new tomograph for quantitative emission computed tomography of the brain. *IEEE Trans Nucl Sci* NS-28: 99–103
 16. Hoffman EJ, Phelps ME, Huang SC et al. (1982) Evaluating the performance of multiplane tomographs designed for brain imaging. *IEEE Trans Nucl Sci* NS-29: 469–473
 17. Iwanczyk JS, Dabrowski AJ, Huth GC et al. (1981) A study of low-noise preamplifier systems for use with room temperature mercuric iodide (HgI₂) X-ray detectors. *IEEE Trans Nucl Sci* NS-28: 579–582
 18. Keil G (1968) Gamma-ray spectroscopy with a scintillator-photodiode combination. *Nucl Instrum Meth* 66: 167
 19. Lynch FJ (1966) Improved timing with NaI(Tl). *IEEE Trans Nucl Sci* NS-13: 140–147
 20. Mazziotta JC, Phelps ME, Plummer D, et al. (1981) Quantitation in positron emission computed tomography: 5. physical-anatomical effects. *J Comput Assist Tomogr* 5: 734–743
 21. Mullani NA, Markham J, Ter-Pogossian MM (1980) Feasibility of time-of-flight reconstruction in positron tomography. *J Nucl Med* 21: 1095–1097
 22. Mullani NA, Wong W, Hartz R et al. (1982) Sensitivity improvement of TOFPET by the utilization of Inter-slice coincidences. *IEEE Trans Nucl Sci* NS-29: 479–483
 23. Nestor OH, Huang CY (1975) Bismuth Germanate: a high-Z gamma ray and charged particle detector. *IEEE Trans Nucl Sci* NS 22: 68–71
 24. Phelps ME, Hoffman EJ, Huang SC et al. (1978) ECAT: a new computerized tomographic imaging system for positron emitting radiopharmaceuticals. *J Nucl Med* 19: 635–647
 25. Phelps ME, Hoffman EJ, Huang et al. (1981) Positron computed tomography – present and future design alternatives. *Medical Radionuclide Imaging 1980*, vol I, IAEA-SM-247/203: 199–230
 26. Phelps ME, Huang SC, Hoffman EJ et al. (1982) An analysis of signal amplification using small detectors in positron emission tomography. *J Comput Assist Tomogr* 6: 551–565
 27. Ricci AR, Hoffman EJ, Phelps ME et al. (1982) Investigation of a technique for providing a pseudo-continuous detector ring for positron tomography. *IEEE Trans Nucl Sci* NS-29: 452–456
 28. Snyder DL, Thomas LJ, Ter-Pogossian MM (1981) A mathematical model for positron-emission tomography systems having time-of-flight measurements. *IEEE Trans Nucl Sci* NS-28: 3575–3583
 29. Snyder DL (1982) Some noise comparisons of data-collection arrays for emission tomography systems having time-of-flight measurements. *IEEE Trans Nucl Sci* NS-29: 1029–1033
 30. Webb PP, McIntyre RJ (1976) Large area reach-through avalanche diodes for X-ray spectroscopy. *IEEE Trans Nucl Sci* NS-23: 138–144

Design Characteristics of a Multiring Positron Camera System for Emission Tomography of the Brain

By L. Eriksson, C. Bohm, M. Bergström, K. Ericson, T. Greitz, G. Blomqvist, J. Litton, L. Widén, P. Hansen, S. Holte, and H. Stjernberg

Introduction

Since 1976 we have been interested in the combination of radionuclide tracer techniques and positron camera devices for regional metabolic studies of the brain as a function of time. Parallel efforts have been made to produce ^{11}C -glucose, to design positron camera systems with high spatial resolution, high sensitivity, and high signal-to-noise ratios (SNR), and to develop metabolic models for ^{11}C -glucose. Positron camera systems with the above design goals are feasible due to the availability of BGO scintillator crystals. The high density of BGO scintillators gives an acceptable data collection efficiency even for the small crystal considered here. With the wobbling technique, suggested by Bohm et al. (1978), a high sampling density can be obtained, resulting in a reconstructed resolution close to the intrinsic detector channel resolution. In this paper we describe the design characteristics of a four-ring positron camera system, PC384-B, and will show results from its present clinical use.

System Design

Gantry

PC384-B has four detector rings with a ring diameter of 48 cm. Each ring has 96 BGO detectors, subdivided into 12 cassettes, each cassette containing eight detectors. The cassettes are housed in a special assembly for easy access for service and replacements. To exclude radiation from parts of the patient outside the region of interest, annular lead shields are fitted around the patient port. The lead thickness is 5 cm over the crystal area on the patient side, tapering off to 2.6 cm at the aperture radius. The average lead thickness on the off patient side is 3.6 cm.

The septa collimator assembly fits inside the cassette frame aperture. Each septum is made of lead 7 mm thick. The septa collimators are used to minimize random and scatter coincidence contributions. The detector assembly and the septa collimator unit perform a continuous wobbling motion. The wobble diameter is 15 mm. The maximum speed is set to 70–80 rpm. The speed used in routine work is 30–60 rpm, and is set via the software.

Detectors

Each cassette contains eight BGO detectors, including scintillation crystals, PM tubes, voltage dividers, and buffer amplifiers. The size of the BGO scintillators is $12 \times 20 \times 30$ mm, giving a geometrical resolution of 6 mm and, with a Cu-64 line source, a 6.5-mm intrinsic resolution. The detectors are separated by lead wedges approximately 3 mm wide at the front, giving a crystal packing fraction of 80%. The eight anode signals of each cassette can drive $50\ \Omega$ at a 500-mV amplitude. The pulse width is set to approximately $1\ \mu\text{s}$. The PM tubes mounted on the crystals are Hamamatsu R647-01.

All mechanical motions of the positron camera, the wobble motion, and patient positioning of the couch including axial wobbling are carried out under computer control.

Electronic System

An overall block diagram of the electronic system is shown in Fig. 1. The anode signals from the 4×96 detectors are analyzed in a discriminator stage. The discriminators operate with a leading edge timing technique using an energy window of 200–700 keV. The output signals (ECL-logic) are organized into groups of 16 and are encoded to give a group signal and a 4-

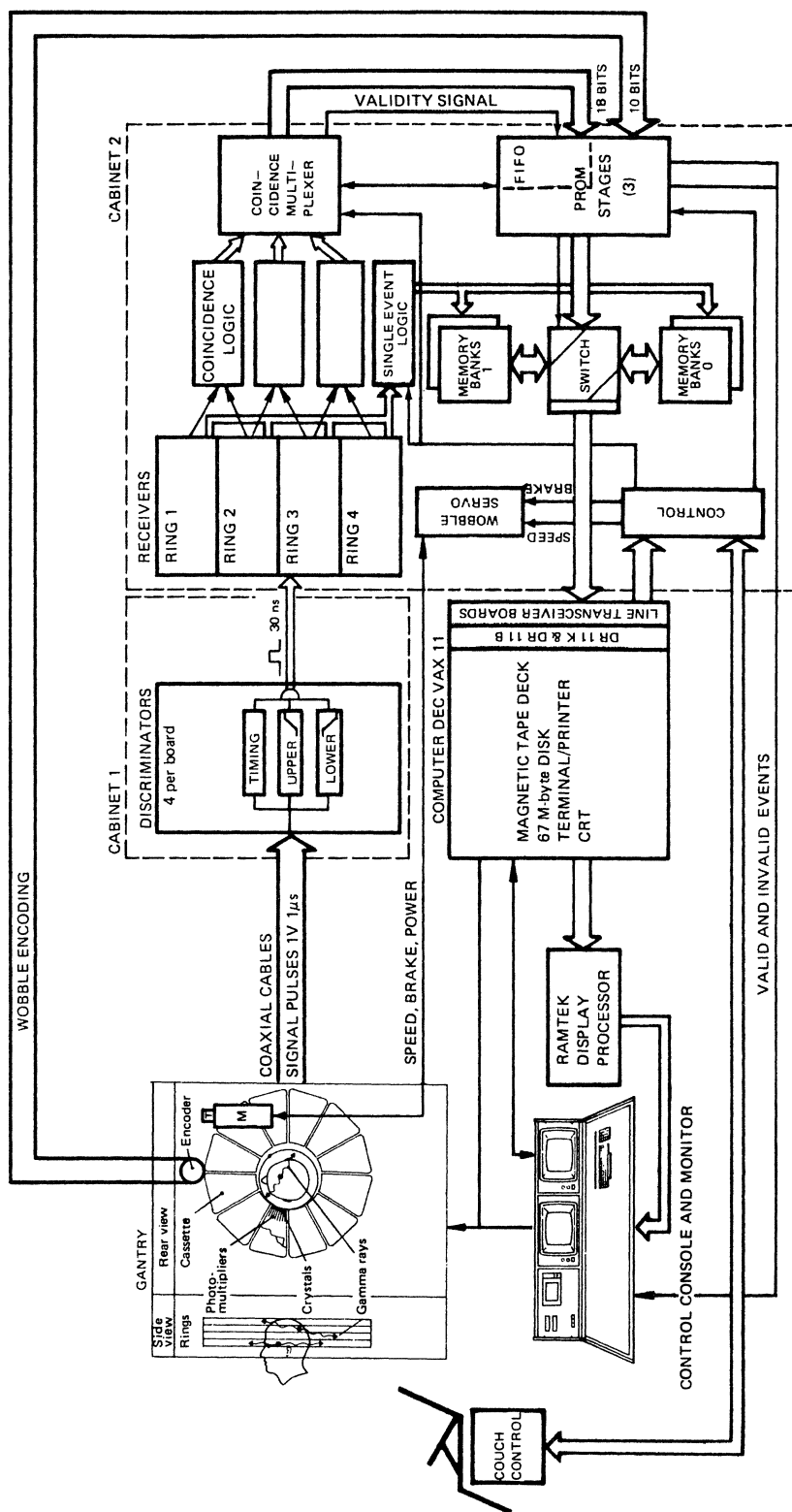


Fig. 1. Overall block diagram of the PC384-B electronic system

bit address code. When a coincidence is detected between two 16-group signals the 16 input encoders are latched and the address code can be read. A coincident event requires the 2×4 bits from the two participating encoders plus 4 bits for defining the two group signals and 4 bits for defining the actual ring combination, resulting in a 16-bit word. This word is passed through the 64-word-deep FIFO memory to regularize the coincidence rate. This leads to a reduced deadtime of the system. The maximum rate of the FIFO is about 10 MHz, giving a system deadtime of the order of 100 ns. The FIFO feeds the preprocessing stage, composed of different PROM stages. First data originating from data channels outside a selected image field are discarded. Next the bit structure of valid events is rearranged into projection angle and projection position. The presence of a valid event will latch the wobble encoder information. The projection angle and the encoder angle enters a look-up table giving the product of the wobble radius and the sine of the difference of the two angles. This gives the wobble displacement, and in the next stage this is added to the stationary detection channel position, giving the true detection channel position, which in turn is used for determining what memory position should be used to store the event. The detector pairs of the ring are organized into 18 parallel detection channels for each of the 96 projection angles. The wobble will spread each of these sensing positions into 12 additional positions with a width of 1.25 mm. An outcoming event is now specified by (a) the ring combination, (b) the projection angle ranging from 1 to 96, and (c) the projection position, ranging from 1 to 256.

The total bit size of a valid event is 18. The PROM stage directly interfaces one of two buffer memories of 256 kbytes each. While one memory bank is accumulating data, the other is transferring data to the computer. The switchings between banks is carried out by the computer. Data transferred to the computer is stored on disk together with a file header containing all the adequate information regarding the patient and examination, for example, the examination starting time, date, measurement times for coincidences and singles, and the camera status, such as the width of the coincidence window and the energy threshold of the

discriminators. Since the detector efficiencies will change with time, data from the last normalization measurement is included in the raw data files. These are performed in a stationary mode, which means that the amount of information is considerably less than data from a normal high-resolution scan. The single count rates from both the normalization and the actual measurement are also included in the data file. Single events are measured simultaneously with but independently of the coincidence counting. The count rates of the 4×96 discriminator outputs are measured via a multiplexing system, and the result is stored in one of two separate buffer memories of 512 locations, 16 bits deep. The singles are needed to estimate the random coincidence contribution and to monitor the overall positron camera status.

Computer

The PC384-B is run on a VAX 11/780 computer with a 512-kbyte memory, VAX/VMS operating system, and a 67-Mbyte disk. All programs, including patient file management, data

Table 1: Control file

CAR	:FDG	! carrier
ISO	:F-18	! isotope
QES	:INFARKT 1	! question
ACT	:7.3	! activity
ASS	:SCHMIDT	! assistant
DOC	:ILSEN	! physician
INP	:ARM	! injection place
INT	:BOL	! injection type
MTM 0	:180	! measurement time
MTM 0-		
MTM 10	:300	
MTM 10-		
ZDI 0	:0	! couch displacement in millimeters
ZDI 0-		
NME	:18	! number of measurements
POR	:R	! patient orientation
PAN	:20	! patient angle
NRV	:30	! wobble speed in revolutions per minute
NPO	:0	! number of wobble positions
		! NPO = 0 means continuous wobbling

listing, application programs, image reconstruction programs, and physiological function programs are all available in high-level language (FORTRAN 77). When input data are required the camera control program first consults a control file, before asking the operator. The questioning sessions are thus reduced to a minimum by the elimination of standard answers. All specification concerning the measurements can be made from these files, for example, measurement times, the position of the chair at different measurements, and the wobble speed. An example of such a file used for dynamic FDG studies is shown in Table 1.

On the left side the mnemonics of the different variables are shown: MTM is the measurement time, ZDI are the different displacements of the couch at the different measurements, NME is the number of measurements, and NRV is the wobbling speed, 30 rpm.

In the actual example the first ten (0–9) measurements are performed with a measurement time of 180 s followed by a sequence of eight measurements run with a 300-s measurement time. Since $ZDI=0$ for all measurements, the chair is not moved between the different positions.

Results

Spatial resolution is defined as the ability to separate adjacent structures under optimal conditions. The spatial resolution is usually specified by the point spread function or the line spread function. To determine the PSF very thin tubes of activity positioned perpendicular to the imaging plane were measured (Fig. 2a). The result for Ga-68 is 8.4 mm FWHM. With a sharper filter and Cu-64 activity, 7.7 mm FWHM was obtained, which is the limiting resolution of the system. The resolution varies slightly with the distance approximately 10% between the center ($R=0$) and $R=10$ cm tangentially.

Spatial invariance or uniformity of image data exists when the pixel values are quantitatively correct irrespective of the position within the imaged object. This is easiest measured with a phantom filled with a uniform concentration of radionuclides. In the resulting image the pixel

values are plotted along a strip (Fig. 2b). A straight horizontal line is fitted to the data points in a least squares sense and the goodness of fit will show the homogeneity. An additional run test gives a more detailed analysis of the degree of homogeneity. A run test on the fit in Fig. 2b shows no statistical deviation.

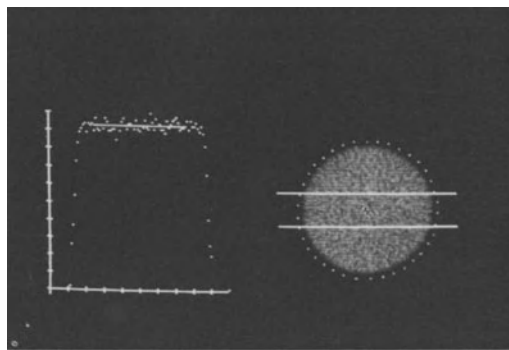
Various configurations of small objects have been used to demonstrate the spatial resolution in positron cameras. A phantom described by Derenzo et al. (1979) contains six sectors with thin holes drilled in a plastic cylinder with a diameter of 20 cm. The hole diameters are one-fourth the distances between the holes, which are 25, 20, 16, 14, 12, and 10 mm. All sectors are resolved with the present positron camera system. A different type of phantom uses bar structures with separations (center-to-center) between activity regions of 25, 18, 14, and 10 mm. All sectors are fully resolved. The number of counts is 100 million for the Derenzo phantom and 200 million for the bar phantom. The images are shown in Figs. 2c, 2d. The advantage of these phantoms is the ease with which a rough estimate of the resolution can be made.

The slice thickness defines the response in the axial direction. For in-plane slices the response depends on the crystal size. For cross-slices the ring separation, the ring diameter, and the septa shield will, in addition to the crystal size, have a strong influence. The geometry also varies considerably between the center and the periphery of the image field. The slice thickness is usually measured with thin plates of activity positioned at different distances from the image center, parallel to the image field. For in-plane slices the FWHM is 12 mm and for cross-plane slices the thickness varies from 9 mm at the center to 14 mm 10 cm from the center. To improve the quantification with cross-slice imaging we can include a wobbling in the axial direction, allowing direct planes and cross-planes in an alternating way sense the same geometrical region. A second step is to avoid cross-slice summation and instead rely on a direct volume reconstruction.

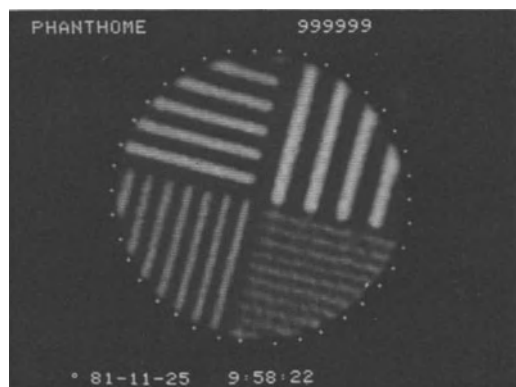
Dead time losses are mostly due to triple coincidence reduction, i.e., the presence of a triple event in the coincidence logic system is discarded. The system response is almost linear up to $1 \mu\text{Ci/cc}$ for a 20-cm diameter water phan-



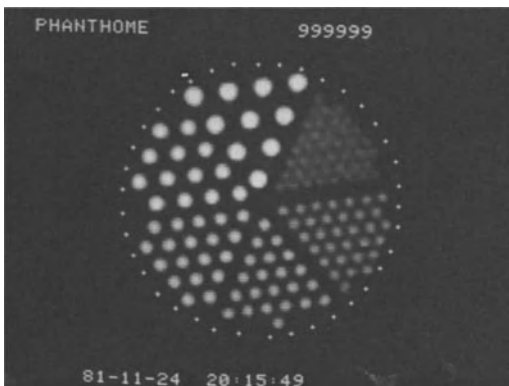
a



b



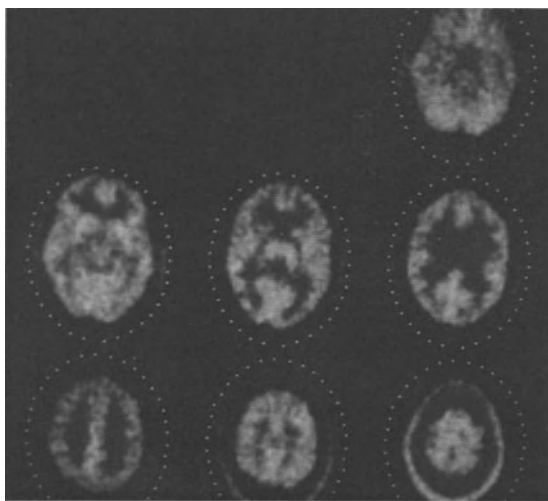
c



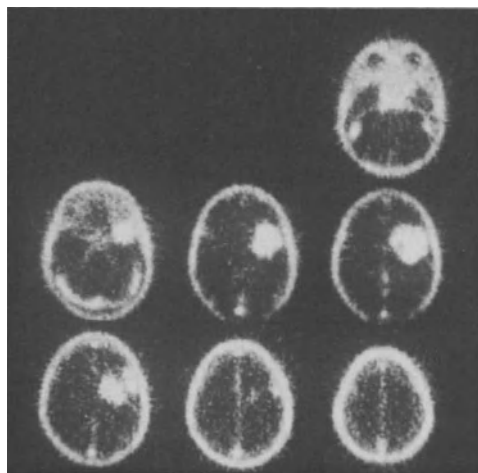
d

Fig. 2a-d. Results from various tests with the positron camera. **a** Spatial resolution test with thin tubes with activity. **b** Homogeneity test with a 20-cm-diameter cylinder phantom. **c** Image of the Derenzo phantom. **d** Image of the bar phantom

ameter cylinder phantom. **c** Image of the Derenzo phantom. **d** Image of the bar phantom



a



b

Fig. 3a, b. Result from two clinical investigations. **a** Summation image from a ^{11}C -glucose study.

b Summation image from a dynamic Ga-68 EDTA study

tom and is then flattened out. However, quantitative correct values can be obtained by using a simple dead time correction factor in the image reconstruction program. This correction gives accurate values up to around $3 \mu\text{Ci/ml}$. However, at this high activity, with around five to ten times more random than true coincidences the image quality is degraded.

The sensitivity denotes the total count rate of the system, excluding random and scatter coincidences, for a specified amount of activity concentration. The usual measure is performed with a 20-cm-diameter cylinder containing a uniform concentration of activity specified as counts/s/ μCi per milliliter. The present figures for PC384-B are 25000 c/s for direct planes, 35000 c/s for cross-plane, and 205000 c/s for the total system. The sensitivity depends on the slice thickness, which must be remembered when comparing the imaging properties of different systems.

Several hundred investigations with Ga-68 EDTA have been performed since the completion of the instrument in 1981. An example of an infarct study is shown in Fig. 3 b. The infarct is nicely shown in five of the seven simultaneously recorded slices. This is a summation of a dynamic study in order to determine the isotope accumulation in the infarct as a function of time. Figure 3 a shows an example of a ^{11}C -glucose study. Usually a sequence of 16 images is recorded in order to give input values of the metabolic model. The image is a summation of the ten last measurements and has approximately 3–4 million counts per image.

Conclusion

The use of small BGO scintillators together with a continuous wobbling motion give uniform high-resolution images with high sensitivity and high contrast. The system is simple to run with several standard routines being adequate for most experimental situations. A new standard routine to fulfill special needs is easy to create. Our clinical experience with PC384-B is good, with very few breakdowns, easy maintenance, and reliable operation even for demanding dynamic function studies.

Acknowledgments. This work has been supported by the Swedish Technical Board of Development, the Swedish Medical Research Council, and the Swedish Natural Science Research Council.

References

1. Bohm C, Eriksson L, Bergstrom M et al. (1978) A computer-assisted ring detector positron camera system for reconstruction tomography of the brain. *IEEE Trans Nucl Sci* NS-25: 624–637
2. Derenzo S, Budinger TF, Cahoon JL et al. (1979) The Donner 280-crystal high-resolution positron tomograph. *IEEE Trans Nucl Sci* NS-26: 2790–2793

Design Concepts and Performances of HEADTOME, a Multiring Hybrid Emission Tomograph for the Brain

By I. Kanno, K. Uemura, Y. Miura, and S. Miura

Introduction

In order to meet an increasing clinical demand for a tomographic measurement of cerebral blood flow (CBF), a prototype HEADTOME was originally designed to measure a dynamic tomogram of inert gas clearance of the brain. To avoid the necessity for a cyclotron, we designed the prototype HEADTOME to include a hybrid capability for measuring both positron and single photon emissions. The prototype HEADTOME was a single-ring tomograph using 64 NaI detectors. Because of the primary demand for a dynamic study, high efficiency dominated other design characteristics. Therefore, a small-diameter detector ring was chosen to provide high efficiency. However, this caused difficulties in obtaining a uniformity of spatial resolution and slice thickness over the field of view (FOV) and in accurately correcting for scattered coincidences. These difficulties resulted in a different partial volume effect over the FOV and low accuracy.

HEADTOME II resulted from the development of HEADTOME to provide three NaI detector rings while maintaining the hybrid capability. Recently we have designed a new positron tomograph – HEADTOME III – which is dedicated to positron studies and uses small, deep BGO crystals arrayed on the larger diameter to achieve high sensitivity and high accuracy. HEADTOME III is the goal of the HEADTOME series – providing accurate measurements of pathophysiological functions of the brain at a reasonable cost. This paper describes the design and performance characteristics of HEADTOME II as a positron emission tomograph and HEADTOME III.

Design of HEADTOME II

HEADTOME II was basically the same design as the prototype HEADTOME (Kanno et al. 1981) except it was expanded to three rings to increase its clinical usefulness by enabling it to measure the whole brain simultaneously. This expansion is an important feature for dynamic studies. The tomograph consists of three rings of 64 NaI crystals. Each crystal, which measures $16 \times 28 \times 70$ mm, is encapsulated in an $18 \times 30 \times 72$ -mm aluminium can and is directly photocoupled to a $\frac{3}{4}$ -in. photomultiplier tube (PMT) (R1450, Hamamatsu). A lead septum tapering to 2 mm thickness at the inner face is placed between the crystals. Detector-to-detector spacing around the ring is 20.6 mm. The three rings are separated by a 5-mm-thick lead plate; the ring-to-ring spacing is 35 mm. The thickness of the lead annulus used to shield the detector rings from photons emerging from outside of FOV is 7 cm at the front face of the gantry and 4 cm at the rear. Four units of collimators are mounted on an upright shaft in the gantry which can be rotated to allow alignment of a single unit within the detector ring. The shaft, which has four rotating slots at the bottom, is hung on a cantilever. The cantilever is fixed on the same base as the detector ring (Fig. 1). Two collimators are for single photon emission imaging; the other two are for positron emission imaging. Of the two positron collimators, a high-sensitivity collimator was designed to measure a cross slice as well as a direct slice, and a high quantitation collimator a direct slice only (Fig. 2).

In order to achieve fine linear sampling, detector motion of wobble and rotation were performed. The rotation movement was made by rotating the ring by one-half of the detector-to-detector spacing. This reduces the linear sampling distance to one-half of the detector-to-detector spacing. The wobble motion was defined by small circular movements of the

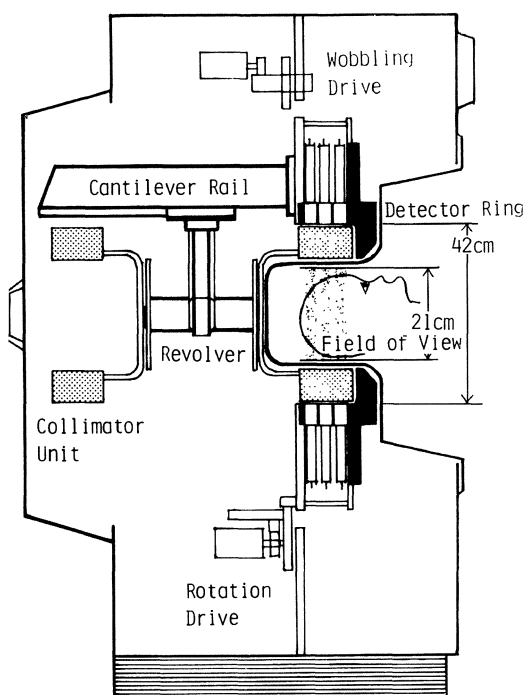


Fig. 1. Cross-section sketch of HEADTOME II gantry. Three rings of 42 cm diameter each containing 64 NaI crystals are fixed on the rotation base. Four collimators are held on the revolving shaft, which is suspended on the cantilever. One of the collimators is chosen inside the NaI rings. The planar field of view is 21 cm in diameter while the axial field of view covers 10 cm

ring, each circular movement being carried out discretely every one-fifth of the circle. The diameter of the wobble circle was determined as three-eighths of the effective detector-to-detector spacing, as measured experimentally at a depth of 1.0 cm below the crystal surface. Thus, the wobble diameter was defined to be 8.1 mm. The combined wobble and rotation give an average linear sampling distance of 3.2 mm. For dynamic studies, only the rotation is carried out. For static studies, the wobble and the rotation are used together. The combination of the wobble and rotation is an optimum choice when considering the scan time and effective linear sampling distance. The average linear sampling distance, when employing the rotation and the wobble, is fine enough to match the intrinsic resolution of the current detector pair, which was measured as 9.5 mm FWHM.

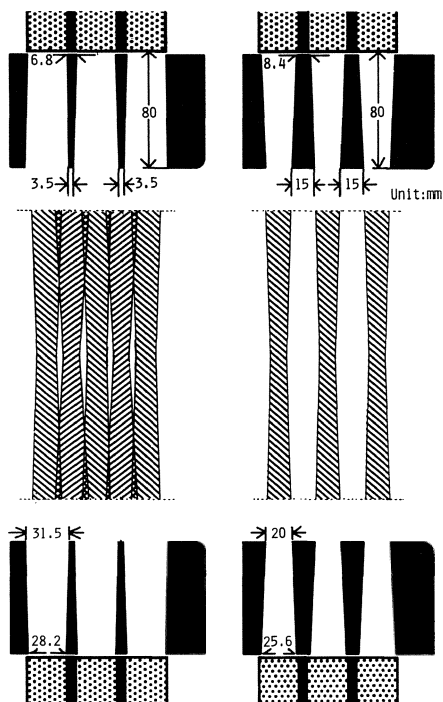


Fig. 2. Two slice collimators of HEADTOME II for positron imaging. A high-sensitivity collimator (*left*) was designed to measure three direct slices and two cross slices. A high-quantitation collimator (*right*) was designed to measure three direct slices only. The tapering of inter-ring lead annulars was chosen to provide a good compromise between sensitivity and uniformity of slice thickness

Signals from the PMT were fed into an amplifier and a timing single channel analyzer (TSCA) with a leading edge timing discriminator. The time resolution measured by one pair of the detector assemblies was 2.8 ns FWHM with the energy discriminator set at a threshold of 100 keV. The time resolution was slightly improved by increasing the threshold of the energy discriminator. In routine studies we normally use a 100- to 700-KeV energy window. TSCA outputs were ORed in groups of eight detectors. Eight group signals for a direct slice or 16 group signals for a cross slice were sent to a coincidence detection circuit. Coincidence detection was achieved on one gate per slice in order to simplify the coincidence gate. However, the one-gate method had a large dead time due mainly to high single event rates per gate. Coincidence line data were corrected for an uneven efficiency of detection line, scattered

coincidence, accidental coincidence, and the attenuation effect of the object. During each patient study we acquired three sets of scans, a blank scan of a ring source without an object, a transmission scan of a ring source with the object in place and an emission scan of the object. The correction for scattered and accidental coincidences was based on the assumption that these phenomena resulted in an even distribution of misplaced events throughout and beyond the operational field of view of the tomogram, and that coincidence events recorded outside this would correspond to scattered and accidental events alone. These false coincidences were determined and subtracted from the area within the field of view of the tomogram. The subtraction was performed at each scan position (two rotation points times five wobble points) and at each slice (three direct slices plus two cross slices). The blank scan and the transmission scan were corrected for the geometric thickness of the ring source for each coincidence line. The emission scan was normalized by dividing by the blank scan to correct for the uneven sampling efficiency of each coincidence line. The transmission scan was divided by the blank scan and converted to attenuation factors. These factors were smoothed in order to reduce the statistical noise arising from the transmission scan and multiplied by the normalized emission scan to correct for the attenuation effect. The last two calculations, i.e., the smoothing of the attenuation factors and their multiplications, were carried out, projection by projection, after interpolating the coincidence lines into paralleled and equally spaced projections.

Table 1. Slice thickness and sensitivity of HEADTOME II

	HS ^a direct	HS ^a cross	HQ ^b direct
Slice thickness ^c			
At the center	15.0	12.0	11.0
9 cm from center	18.0	23.5	14.5
Sensitivity ^d	27.5	36.5	25.3

^a HS: high-sensitivity collimator.

^b HQ: high-quantitation collimator.

^c FWHM (mm) measured by Na-22 point source.

^d 10³ cps/μCi/ml measured by Ga-68 solution in 20-cm diameter pool

Performance of HEADTOME II

HEADTOME II has been used since October 1981. Performance as a positron emission tomograph was, in some respects, sacrificed in order to optimize for single photon emission tomography. Performances in the single photon mode have been reported elsewhere (Hirose et al. 1982). Therefore we confine ourselves here to its function as a positron emission tomograph.

Spatial resolution was measured using a Ga-68 line source in a 20-cm-diameter lucite cylinder at various distances from the center of the FOV. At each position of the line source the spatial resolution was obtained along both a radial and a tangential direction. At the center of the FOV the resolution was 10 mm FWHM and 20 mm FWTM. Spatial resolution was increased by ~ 15%–20% along the tangential direction, but by less than 10% along the radial direction at 9 cm from the center. The difference in spatial resolution between the direct slice and the cross slice was negligible. In addition we did not find significant differences in resolution between the high-sensitivity collimator and the high-quantitation collimator.

Slice thickness was measured using a point source of Na-22. This was placed in a 20-cm-diameter water pool and scanned in an axial direction at 4-mm intervals. Slice thickness varied by more than 20% from the center to the periphery of the FOV in any slice, for both direct and cross slices for each collimator (Table 1). This difference of slice thickness between the center and the periphery resulted from the small diameter of the detector ring of the system. In the cross slice of the high sensitivity collimator, the slice thickness at the periphery was more than double that at the center.

System sensitivity, measured using a standard method employing a 20-cm-diameter uniform activity pool, was 27.5 × 10³, 36.5 × 10³, and 25.3 × 10³ cps/μCi per milliliter within the direct slice and the cross slice of the high-sensitivity collimator, and the direct slice of the high-quantitation collimator, respectively (Table 1). These results are from scans which had been corrected for false coincidences as described above.

The accuracy of the system was evaluated using a phantom containing twelve 4-cm-square bot-

tles inside the 20-cm water pool. Each bottle contained a different concentration of Ga-68 in solution. Three randomly arrayed concentration patterns were measured and the mean pixel value in a 1.5-cm square corresponding to each bottle was plotted as a function of activity concentration of each bottle (Fig.3). The departure of the data points from the line of identity was probably the result of inaccurate correction for the scattered coincidences. Therefore, the assumption that the scattered coincidence distribution can be approximated by the level outside the FOV is not entirely satisfactory for small-diameter ring systems.

Design of HEADTOME III

We have recently redesigned HEADTOME II to give increased accuracy and efficiency in the positron emission scan. HEADTOME III employs small, deep BGOs and a relatively large ring diameter to satisfy these requirements. The tomograph consists of three BGO rings with a diameter of 75 cm. Each ring has 160 BGOs with dimensions $13.4 \times 25 \times 40$ mm. The BGO is directly photocoupled to a $\frac{1}{2}$ -in. PMT and assembled within magnetic shielding. The detector assembly is made up of discrete components to allow easy exchange. The detector-to-detector spacing is 14.7 mm, which results in detector packing ratio of 91%. Ring-to-ring axial spacing is 30 mm. The axial FOV covers 8.5 cm.

The collimator system for defining slices consists of a fixed part and a removable part. The fixed part is a high-sensitivity collimator – the removable part is a high-accuracy collimator. The inner diameter of the high-sensitivity collimator is 50 cm. Thus, the system includes the option of recovering a whole body tomogram when used in the high-sensitivity mode. The 30-cm (inner diameter) high-accuracy collimator can be easily attached or removed from the inside of the high-sensitivity collimator. The use of the high-accuracy collimator will improve the quantitation of brain studies. Both collimators are designed to acquire five slices with a uniform geometry over the FOV with a minimum scattered coincidence rate (Tanaka et al. 1982).

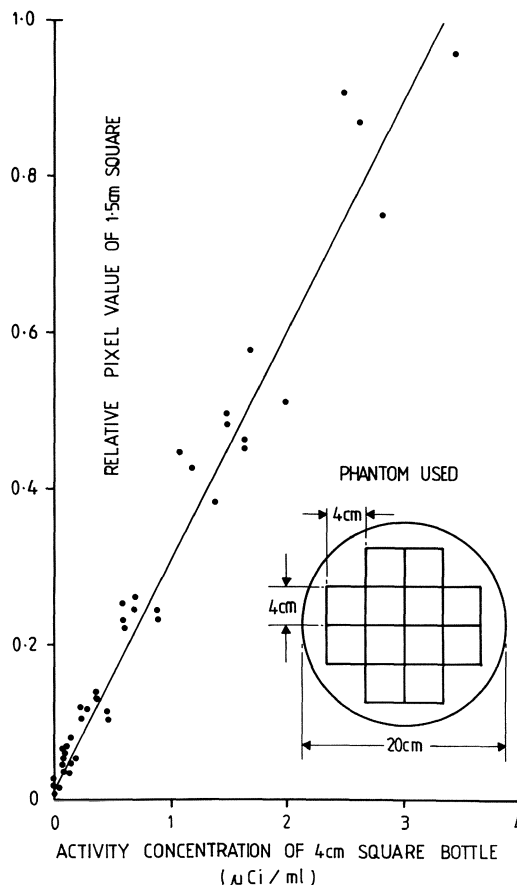


Fig. 3. Evaluation of linearity of HEADTOME II. A phantom array of twelve 4-cm-square bottles containing solutions of Ga-68 of varying concentrations was scanned in a 20-cm-diameter water pool. From tomograms of three different patterns, mean pixel values from 1.5-cm-square areas were plotted as a function of activity concentration in the bottle. The high-sensitivity collimator and direct slices only were used

In order to perform a high-resolution scan 160 5×8 -mm rectangular tungsten bars are inserted to narrow the BGO openings using a motor drive. The intrinsic resolution of the BGO detector, measured as 8.0 mm FWHM, is then reduced to 5.8 mm FWHM.

Scan motions to be employed in the HEADTOME III are basically the same as HEADTOME II, namely a half rotation and a wobble. The wobble diameter is 5.5 mm. This configuration gives an average linear sampling dis-

Table 2. Design parameters of HEADTOME III

Number of rings and slices	3 Rings 5 slices
Diameter of the ring	75 cm
Number of crystals	160 BGOs per ring
BGO dimensions	13.4 × 25 × 40 mm
BGO packing fraction	91%
Narrowing collimator	5 × 8-mm tungsten bar
BGO opening	
Without narrowing collimator	13.4 mm
With narrowing collimator	9.7 mm
Patient aperture	
High sensitivity mode	50 cm
High accuracy mode	30 cm
Planar field of view	
High sensitivity mode	40 cm
High accuracy mode	25 cm
Axial field of view	8.5 cm
Scan motion	Half rotation and wobble
Rotation	½ BGO spacing
Wobble	Five points of 5.5 mm diameter

tance of 1.8 mm. Dead time during mechanical motion is designed to be as short as possible so as not to reduce the effective sensitivity in the dynamic mode. The half rotation takes 0.2 s and one point of wobble 0.2 s.

Electronic configurations are as follows: PMT outputs are fed to the TSCAs with a constant fraction timing discriminator. The measured time resolution of the detector assembly is 6.6 ns FWHM. The outputs of the 160 TSCAs

of each ring are divided into ten groups of 16 adjacent detectors. Coincidence fan is achieved with the opposing seven groups. The number of coincidence gates is 35 per slice.

The design parameters of the HEADTOME III are summarized in Table 2. The estimated performance characteristics are: (1) spatial resolution of 8.5 mm FWHM without the narrowing collimator and 6 mm FWHM with it, (2) a slice thickness of 14 mm FWHM, (3) the uniformity of these planer and axial resolutions being kept to less than 10% within the 10-cm radius of the FOV, and (4) the sensitivity for true coincidences of 50×10^3 cps/ μ Ci per milliliter without use of the narrowing collimator and 15×10^3 cps/ μ Ci per milliliter with it.

The HEADTOME III will be ready for clinical use in April 1983.

References

1. Hirose Y, Ikeda Y, Higashi K et al. (1982) A hybrid emission CT – HEADTOME II. *IEEE Trans Nucl Sci* NS-29: 520–523
2. Kanno I, Uemura K, Miura S et al. (1981) HEADTOME: A hybrid emission tomograph for single photon and positron imaging of the brain. *J Comput Assist Tomogr* 5: 216–226
3. Tanaka E, Nahara N, Tomitani T et al. (1982) Analytical study on the performance of multi-layer positron computed tomography scanner. *J Comput Assist Tomogr* 6: 350–364

Radiochemicals

(Chairman: Y. L. Yamamoto)

Labeled Compounds for Positron Emission Tomography

By A. P. Wolf and J. S. Fowler

The development of accelerator-produced radionuclides is undergoing a rapid if not exponential increase worldwide. The positron emitters carbon-11, nitrogen-13, oxygen-15, and fluorine-18 are enjoying ever increasing attention and are being actively utilized in clinical research and application. The advent of the small cyclotron that is easy to operate and maintain and can be readily installed in university hospitals, and the more recent general availability of positron emission transaxial tomographs of sufficient resolution and sensitivity, taken in conjunction with the development of a few new radiopharmaceuticals labeled with positron emitters, have made possible new research and clinical application in neurology, cardiology, and oncology. However, these efforts still require a multi- and interdisciplinary approach. The lower radiation doses and possibilities for short-term serial studies are part of the advantage of short half-life radionuclides. Biomolecules of all sorts can be labeled with positron emitters without alteration of their biochemical and physiologic activity. By virtue of the annihilation radiation from the positron emitter they can be located and quantitated in a small volume element of tissue. Present efforts are focused mainly on measuring metabolic rates in the brain and in the myocardium. Assessing receptor-site action in the brain and heart with labeled neuroleptics is under intensive development. Noninvasive methods that will allow us to evaluate the dynamics of such action in disease states and to quantitate the response to therapy can be expected in the near future. Protein synthesis and cell proliferation rates measured *in vivo* are under study. Synthetic techniques and the development of both remotely operated and automated procedures yielding positron emitter labeled biomolecules of all sorts, including simple alcohols, sugars, amino acids, steroids, alkaloids, and a host of drugs, make possible a wholly new area in nuclear medicine.

From 1939 through 1976 (Christman and Karlstrom 1975; Karlstrom and Christman 1978) the total number of papers and abstracts describing compounds labeled with the positron emitters ^{11}C , ^{13}N , ^{15}O , and ^{18}F numbered less than 200. In the ensuing 6 years, another 200 have appeared in the literature. This is clearly indicative of an increasing interest in these compounds. The application of radiopharmaceuticals and the techniques of nuclear medicine in the traditional fields of neurology and cardiology is currently under intense exploitation. The positron-emitting radiopharmaceuticals have been especially successful in application in the study of function and metabolism, e.g., the study of complex physiologic and biochemical processes *in vivo*. ^{11}C , ^{13}N , and ^{15}O are all isotopes of the elements of life and have the unique advantage that their incorporation into naturally occurring biomolecules or drugs of various classes does not alter the compounds' defined courses in living systems. Labeling organic compounds with other radionuclides often changes the transport properties of the compound, and usually profoundly affects the biochemical course *in vivo*. This is not to say that such labeling cannot be useful, but it does require a thorough study of the various biological and biochemical aspects of its use if it is to be used to probe a specific function or metabolic pathway. This becomes particularly significant if one is studying such a function in a pathologic state where specific pathways are often even less clearly understood. This contrast can be exemplified by considering the application of ^{11}C -glucose (Raichle et al. 1979), ^{14}C -2-deoxyglucose (Sokoloff et al. 1977), ^{11}C -2-deoxyglucose (MacGregor et al. 1981; Reivich et al. 1982), and ^{18}F -2-deoxy-2-fluoro-D-glucose (Reivich et al. 1979). Validation of the use of the glucose analogs as quantitative indicators of glucose metabolism was necessary and a substantial literature documenting details is in evidence (Phelps 1981). If, instead of

a β^+ -emitter labeled deoxyglucose analog, a technetium-labeled analog were suggested, then resultant altered structure, charge state, electron density, stereochemistry, etc., would require a complete restudy of all parameters. We are fortunate in having the positron emitter ^{18}F available, since in many cases it can be used as a label without profound changes in organism response. Its size and predominant monovalent character allow one to use it as a substitute for hydrogen or hydroxyl. Nevertheless, it must always be borne in mind that its electron density and chemical nature is such that it cannot be assumed to be a perfect mimic for hydrogen in positions in a specific molecule that are assumed to be noninteractive sites.

Positron emitter-labeled radiopharmaceuticals should be considered in the context of their application in biologic and medical research. Indeed, their preparation in isolation is a sterile exercise. The three areas that interact and overlap in biological and medical research and medical application of positron emitters are instrumentation (positron emission tomography encompassing physics, electronics, mathematics, computer science, etc.), medicine (nuclear medicine and those disciplines such as neurology and cardiology that derive benefit from the techniques, etc.), and chemistry (production of radionuclides, synthesis, basic biochemistry, pharmacology, etc.). It will be the purpose of this chapter to place one of those areas, chemistry, in perspective and to provide some information on application in research and practice. It is not our intention to provide an exhaustive review but rather to give a general overview. The topics considered in the area of chemistry are the accelerators required to produce the radionuclides, targetry, and the precursors required for compound preparation, the compounds that need to be prepared, and finally a short comment on application, especially of glucose analogs and neuroleptics, classes of compounds that have given strong impetus to the development of positron emitters and positron emission tomography.

Accelerators

The subject of accelerator production of medical nuclides has been the subject of a number

of reviews (Tilbury and Laughlin 1974; Wolf 1977; Silvester and Waters 1979; Ruth and Wolf 1979; Beaver et al. 1979). In recent years accelerator manufacturers have made a real effort to provide a product that is more amenable to routine use. The "small" machines of today constitute a major departure from the complex machines available just 5 years ago. The cyclotron is still the only particle accelerator sold for radionuclide production for medical purposes. Two other accelerator types, the Van de Graaff and the linac, could also be considered, but manufacturers have not yet addressed the question of providing sufficient particle energy and beam current for radionuclide production in the quantities needed for routine clinical application. A fourth approach might be the use of an electron linac in carrying out photonuclear reactions for radionuclide production. In this latter case, however, cost and capacity are deterrents to foreseeable meaningful application.

It is this question of routine clinical use in addition to needed clinical research that is really determining. Clinical use is defined here as being a patient-limiting factor not a machine-limiting factor. The facility must be capable of providing sufficient radionuclide and tomographic capability to service the patient load, be it a mix of required radionuclides and diagnostic procedures or be it five to ten glucose metabolic studies per day. There is no question that today the only accelerator meeting this requirement in a hospital environment is a cyclotron. However, available particle energy from beam current, particle type, and target material will determine which radionuclides can be efficiently prepared and which can be prepared in sufficient quantity for a clinical program. With the ever increasing interest in accelerator-produced radionuclides, it may well be that other particle accelerator types will become competitive.

Table 1 lists four of the major positron emitters and some of their properties. Over 80% of the literature to date is concerned with only ^{11}C and ^{18}F . Thus, the following discussion is limited to an accelerator that can be used effectively for these two radionuclides plus ^{13}N and ^{15}O . To be sure, larger accelerators can produce not only these radionuclides but many more. However, attendant problems in siting, housing, and

Table 1. Positron emitters for biomolecules

Radionuclide	Half-life (min)	Atoms/Ci	Ci/mol	End-point positron energy (MeV)	% β^+ decay
Carbon-11	20.4	6.5×10^{13}	9.2×10^9	0.97	99 + %
Nitrogen-13	9.96	3.2×10^{13}	1.9×10^{10}	1.19	100%
Oxygen-15	2.07	6.6×10^{12}	9.1×10^{10}	1.7	100%
Fluorine-18	109.7	3.5×10^{14}	1.7×10^9	0.635	97%

Table 2. Small cyclotrons suitable for ^{11}C , ^{13}N , ^{15}O , and ^{18}F production

Company	The Cyclotron Corporation (TCC)	Japan Steel Works (JSW)	Scanditronix	Sumitomo CGR-MeV
Model	CP 16 ^b (CS 16 ^c)	JSW 168	RNP-16	Cypris 325
Proton energy	16 (16)	16 (17) ^e	16	13.6
Deuteron energy	8 ^d (8)	8 (10)	8	7.5 ~ 8.0
Available external beam current ^a	50 μA	50 μA	50 μA	50 μA

^a The current given is the minimum guaranteed current. Some of the cyclotrons will exceed this current, especially for protons.

^b The CP 16 is a negative ion machine. The others are positive ion machines.

^c The CS-16 is still available from TCC and is a four-particle positive ion machine.

^d Deuterons are an option on the CP 16.

^e A slightly larger version with 17 MeV protons and 10 MeV deuterons is also available

maintenance of operation increase the complexity and cost of the facility.

A listing of available "small" accelerators is given in Table 2. These can be considered as state of the art and suitable for production of ^{11}C , ^{13}N , ^{15}O , and ^{18}F .

While each of the machines listed has numerous positive features and some deficiencies, they all attempt to incorporate design features combining simplicity of operation, reliability, and compact structure. The manufacturers also provide targetry and remote systems for precursor production of a limited number of compounds and will provide details on request. These optional items vary in cost and level of operator control and sophistication. It should be emphasized, however, that there are other machines available with greater power and capacity from the four manufacturers. There is little question that as experience is gained with this new generation of machines the trends toward automation will evolve toward minimal operator intervention. Some mention might be made of the situation in the East Bloc countries. At present, the Leningrad Accelerator

Table 3. Production reactions for positron emitters

Radionuclide	Nuclear reaction
Carbon-11	$^{10}\text{B}(\text{d},\text{n})^{11}\text{C}$ $^{11}\text{B}(\text{p},\text{n})^{11}\text{C}$ $^{14}\text{N}(\text{p},\alpha)^{11}\text{C}$
Nitrogen-13	$^{12}\text{C}(\text{d},\text{n})^{13}\text{N}$ $^{16}\text{O}(\text{p},\alpha)^{13}\text{N}$ $^{13}\text{C}(\text{p},\text{n})^{13}\text{N}$
Oxygen-15	$^{14}\text{N}(\text{d},\text{n})^{15}\text{O}$ $^{15}\text{N}(\text{p},\text{n})^{15}\text{O}$
Fluorine-18	$^{18}\text{O}(\text{p},\text{n})^{18}\text{F}$ $^{20}\text{Ne}(\text{d},\alpha)^{18}\text{F}$

Factory (Technabsport) has constructed one medical cyclotron approximately equivalent to a TCC CS 22 or a CGR MeV 520. It is installed in Turku, Finland. They have announced their intention to build a machine similar in capability to those in Table 2; however, no details are available.

Whichever accelerator is chosen, the single salient consideration should be that it is to be viewed as a source of reagent, and in the con-

Table 4. Carbon-11 and fluorine-18 yields^a

	Saturation yield (mCi/ μ A)		Theoretical yield for 1-h bombardment (mCi/ μ A)		Production (mCi/ μ A)	
	8 MeV	14 MeV	8 MeV	14 MeV	8 MeV	14 MeV
Carbon-11-CO ₂ ¹⁴ N(p, α) ¹¹ C	36	130	31	113	28	102
Fluorine-18-F ₂ ¹⁸ O(p,n) ¹⁸ F	110	216	35	69	(17) ^b	(35) ^b
²⁰ Ne(d, α) ¹⁸ F	46	84	15	27	11	19

^a Yields are based on full energy on target. Production yields take into account all factors affecting practical yield.

^b Expected but not as yet realized in practice

text of this article, a quantity of protons and deuterons to be able to make a sufficient amount of the four positron emitters in question.

Targetry and Precursors

The choice of nuclear reaction for the production of a particular radionuclide is often a matter of individual preference. Yield and convenience, based on particle energy and current availability, usually dictate the choice. There are many reactions available. However, the reactions most commonly used are listed in Table 3. Yields are not listed, since these depend on the target design, length of bombardment, and nature of precursor required. Nevertheless, the machines listed in Table 2 have the capacity to allow ample production of the required positron emitters. Economical use of time in a clinical environment dictates a maximum 1- to 2-h bombardment to produce the required amount of activity. Carbon, oxygen, and nitrogen are no problem. ¹⁸F production, however, requires careful target design in order to allow adequate ¹⁸F production as [¹⁸F]F₂. Production of aqueous H¹⁸F poses less of a problem. An alternate approach to these radionuclides involves the use of a machine delivering protons only, in combination with the enriched nuclides required for (p, n) reactions. The current costs of the enriched nuclides are such as to be prohibitive in the case of the ¹⁸O: ¹⁸F system unless a method for recovery of the target gas can be devised. Production of fluo-

ride ion from an H₂¹⁸O target is, however, economically feasible since the recovery of the water as the radiolysis product poses less of a problem. While it is outside the province of this article to go into details of production, a comparison of theoretical and production methods for carbon and fluorine is given in Table 4. It is immediately apparent that for ¹⁸F the saturation yield and the yield realized in a 1- to 2-h bombardment are very different. It is just this type of consideration that is determining in a routine use situation.

The production capacity of the small cyclotrons operating at external beam energies of 16-MeV protons and 8-MeV deuterons is adequate for any presently conceived in-house program involving the four positron emitters. However, there is no question that, at least in the case of ¹⁸F, a somewhat higher deuteron energy (~10–12 MeV) would be useful.

The question of machine parameters is intimately related to the problem of precursor production. Precursor yields always need to be maximized because subsequent steps in an organic synthesis may result in a substantial decrease in radiochemical yield. The preparation of precursors has received extensive review in the literature (Wolf and Redvanly 1977; Straatmann 1977; Palmer et al. 1977). The commercial companies offer apparatus for the remote production of the more conventional precursors. Such devices can, in some cases (e.g., for ¹¹CO and ¹¹CO₂) be built as easily and at lower cost in the local shop. Table 5 lists some of the most common precursors and compounds that are either used directly in a diagnostic procedure or are used in synthetic procedures. All of

Table 5. Some simple β^+ -emitter-labeled compounds for

synthesis and biological studies	
Radionuclide	Precursor
Carbon-11	^{11}CO , $^{11}\text{CO}_2$, H^{11}CN , H^{11}CHO , $^{11}\text{CH}_3\text{I}$, $^{11}\text{COCl}_2$
Fluorine-18	$^{18}\text{F}]\text{F}_2$, H^{18}F (anhydrous) H^{18}F (aqueous), $\text{CH}_3\text{CO}_2^{18}\text{F}$
Nitrogen-13	$^{13}\text{NH}_3$, $^{13}\text{NO}_2^-$, $^{13}\text{NO}_3^-$
Oxygen-15	$^{15}\text{O}]\text{O}_2$, H_2^{15}O , C^{15}O , $^{15}\text{O}]\text{CO}_2$

the substances listed are available at end of bombardment (EOB) or shortly thereafter, and remotely operated devices for this production have been reported in the literature. As the need for routine delivery of compounds becomes more common, fully automated production under microprocessor control will become mandatory so that operator intervention is bypassed. This is now within our technologic capabilities and such devices will be reported in the literature in the next several years. The key elements making this possible are the new generation of partially automated small cyclotrons and the now existing thorough understanding of the chemical systems involved in the preparation of some of the radionuclides and simple precursors.

Radiopharmaceuticals

The foregoing discussion on cyclotrons and precursors makes it clear that an in-house facility is to be preferred if positron emitters are to be used in the clinical environment. Such use is mandatory for the compounds of ^{15}O and ^{13}N . ^{11}C compounds could, in principle, be supplied to users who are no more than 1 h removed from the source. However, this would require production of very large quantities of material and near perfect coordination between supplier, shipper, user, and subject in order that the material be effectively used. ^{18}F compounds, however, can be shipped reasonable distances. For example, we have been delivering ^{18}F -2-deoxy-2-fluoro-D-glucose ($2\text{-}^{18}\text{F}$ FDG) to the University of Pennsylvania Medical School, Philadelphia; Massachusetts General Hospital,

Boston; Harvard Medical School, Boston; and the NIH, Bethesda, for research purposes and for use in research in human subjects. Production is by completely remote operation (Fowler et al. 1981). Thus, in going from a half-life of 20 min to a half-life of 110 min, what is marginal at best becomes possible in terms of regional delivery of positron emitter labeled compounds. The concept of a regional facility for diagnostic and other applications thus has two distinct components. Patients and research subjects must go to the facility if ^{11}C , ^{13}N , and ^{15}O are to be used. The application of ^{18}F , however, can be accomplished both at the site of the accelerator and within a radius defined primarily by transportation availability. (However, even here, the many regulations, local and regional transport requirements, and bureaucratic obfuscation may make the transport of individuals to the particular site the only effective alternative.)

The special problems relating to positron emitter labeled radiopharmaceuticals thus far addressed have covered the accelerators required for production of the radionuclide and the targetry and precursor preparation. The final steps involve the preparation of the radiopharmaceutical itself and its application.

Radiopharmaceuticals labeled with the four positron emitters under consideration have been reviewed in depth (Wolf and Redvanly 1977; Straatmann 1977; Palmer et al. 1977; Wolf et al. 1973; Wolf and Fowler 1979; Welch and Tewson 1979; First Int'l. Symp. 1976; Second Int'l. Symp. 1978; Third Int'l. Symp. 1980). A recently completed monograph contains a comprehensive discussion of ^{11}C , ^{13}N , and ^{18}F labeled compounds (Fowler and Wolf 1982). A distinction must be made between the development of these compounds and the preparation of these compounds in a routine fashion. As noted previously, a few of the positron emitters that have seen extensive use in the past and continue to be studied and used today, principally ^{11}CO , $^{11}\text{CO}_2$, $^{13}\text{NH}_3$, $^{15}\text{O}\text{-CO}_2$, and H_2^{15}O , are easily prepared. In some cases commercial devices for remote preparation are available, and new devices for preparation of the somewhat more complex precursors (e.g., $^{11}\text{CH}_3\text{I}$) have been reported (Iwata et al. 1979). The increasing awareness over the past several years that the more complex radiopharmaceuticals

have potential utility in clinical medicine has provided impetus for developing routine and remote methods for some of the more useful compounds. The "*in vivo* measurement of transport and metabolic processes using radio-tracers" (Gallagher et al. 1979) and the ability to probe receptor populations, enzyme levels, and other biochemical and functional activities can be exemplified by research and application in heart and brain.

The well-known metabolic substrate for the myocardium, palmitic acid, was first labeled with ^{11}C in 1969 (Welch 1969) by the carbonation of a Grignard reagent. Subsequently, this ^{11}C -labeled compound has been studied extensively (Poe et al. 1975; Weiss et al. 1976; Hoffman et al. 1977; Goldstein et al. 1980; Ter-Pogossian et al. 1980; Lerch et al. 1981; Schelbert et al. 1982). The relationship of structure to *in vivo* scintigraphic distribution pattern has been studied for a large number of carboxylic acids (Winstead et al. 1973). More recently, ^{18}F -labeled analog fatty acids have been prepared and studied (Knust et al. 1979). The preparation of ^{11}C -labeled palmitic acid has been carried out in a remotely operated apparatus at Washington University, St. Louis, Mo., since 1976 and is used for delivery of compound several times a week on a purely routine basis. The carbonation of a Grignard is a reaction particularly well suited for use in radiopharmaceutical production, since it is a one-step reaction usually resulting in a high yield, is rapid, and yields a product readily purified. Should palmitic and other fatty acids labeled with ^{11}C prove to be valuable for routine use in the clinic, a truly automated production device can be constructed.

The upsurge in interest in small cyclotrons was due in part to recent success in the quantitative measurement of regional brain glucose metabolism *in vivo* and essentially noninvasively in humans (Reivich et al. 1979). Positron emitters also can be of use to quantitate regional fractional extraction of oxygen (Raichle 1979; Lenzi et al. 1978) and blood flow and blood volume (Raichle 1979). New and active areas of interest utilizing ^{11}C and ^{18}F compounds involve receptor site mapping and the study of tumor growth rate.

The compounds (other than the labeled ^{15}O and ^{11}C gases) required for this present work

are in most cases considerably more difficult to develop and prepare. The use of labeled glucose and some of its analogs is a case in point. The preparation of randomly, but not necessarily uniformly, labeled glucose was described in 1971 (Lifton and Welch 1971; Wolf 1971). A biosynthetic method involving plant materials was used. While demonstrating that ^{11}C -glucose could be made rapidly, problems that are general for biosynthetic methods involved the necessary rapid separation from plant residues, particularly the removal of pyrogens and other physiologically active compounds. The use of algae to prepare the ^{11}C -labeled sugar galactose has been described (Palmer and Goulding 1974). A recently reported synthesis of ^{11}C -glucose utilizing algae as the biosynthetic medium may have some advantages over the plant method (Ehrin et al. 1980). More recently a chemical synthesis of ^{11}C -glucose and ^{11}C -mannose labeled at carbon-1 has been reported (Shiue and Wolf 1981). Since glucose is ubiquitous in mammals and dilution occurs immediately on injection, questions of specific activity do not arise. However, chemical yield is of consequence. The advantages and disadvantages of utilizing ^{11}C -glucose for regional glucose metabolism measurements have been addressed (Raichle 1979). With the development of the Sokoloff-Reivich model for measuring regional brain glucose metabolism (Sokoloff et al. 1977), the need for labeled 2-deoxy-D-glucose or a labeled analog arose. A decision was made to develop the preparation of 2- ^{18}F FDG since the longer half-life would be a distinct advantage in what was essentially a steady-state method and ^{18}F would provide the limiting resolution of all the positron emitters that could be used as potential labels. (All positron tomographs existing today have resolutions considerably poorer than the limits set by the radial range decay characteristic of ^{18}F . A new synthetic method using ^{18}F -labeled elemental fluorine was developed (Ido et al. 1977) and applied to labeling the 2- ^{18}F FDG (Ido et al. 1978; Fowler et al. 1981). Subsequently, a synthesis for the "parent" ^{11}C -2-deoxy-D-glucose was also devised (MacGregor et al. 1981; Shiue et al. 1979a; Shiue et al. 1978). The ^{11}C -2-deoxy-D-glucose has the distinct advantage of allowing serial studies on the same day in the same individual. Thus drug intervention studies and

other experiments can be readily carried out. The first use in humans as well as the feasibility of carrying out multiple syntheses and human studies in a single day is described by the BNL-Univ. of Penn. group (Reivich et al. 1982). Preliminary reports of the synthesis of 2-¹⁸F-DG using fluoride ion have appeared recently (Levy et al. 1982; Tewson and Gould 1982). Considerable improvement in the 2-¹⁸F-DG synthesis has been achieved utilizing a remote synthesis system (Fowler et al. 1981). Higher radiochemical yields (~20%) have been achieved by using ¹⁸F-labeled acetyl hypofluorite in the 2-¹⁸F-DG synthesis (Shiue et al. 1982) and production yields of 35–45 mCi of product (at delivery) from 300 mCi of [¹⁸F]F₂ (at EOB) are routine. Nevertheless, a synthesis utilizing fluoride ion would be preferred because of the potentially higher yield obtainable in most nucleophilic displacement reactions. In addition, the potential specific activity can be very much higher. A synthesis for ¹⁸F-3-fluoro-3-deoxy-D-glucose has been reported (Tewson et al. 1978). However, here again several new problems arise regarding the applicability to the Sokoloff-Reivich model because $V_{\max}$ is considerably smaller than that for glucose and the Michaelis-Menten constant is considerably greater than for glucose. The ¹¹C-deoxy sugar is chemically identical with the compound used to develop the original model. Although the shorter half-life of ¹¹C will pose some difficulty when it is used in human studies because of limitations on the amount that can be injected, coupled with the low sensitivity of positron tomographs and the time course of the study, multiple-slice positron emission tomographs which have a small aperture and are designed strictly for brain studies will alleviate many of the problems related to speed and sensitivity. Generally speaking, however, ¹¹F-labeled compounds are easier to use than ¹¹C-labeled compounds in a clinical situation.

The foregoing detailed description of the sugars was given in order to emphasize the many problems that arise with regard to organic radiopharmaceuticals labeled with positron emitters. While the majority of studies with 2-¹⁸F-DG have involved the study of regional brain glucose metabolism, it has also been applied to the study of myocardial metabolism (Schelbert et al. 1982) and tumor growth (Som

et al. 1980). Along this line, the 2-¹⁸F-DG method provides a striking example of the potential value in developing β^+ -emitter-labeled compounds which can be used to extend the techniques of neurochemical autoradiography (Rainbow et al. 1982) to humans.

One of the most challenging and active areas of research in the development of β^+ -emitter-labeled radiotracers relates to their use in studying brain neurotransmitter receptors in humans. Studies using tritium-labeled receptor ligands of very high specific activity in conjunction with quantitative autoradiographic and pharmacological techniques have served to identify molecules which have a particular affinity for various neurotransmitter receptors. Extension of these studies to humans requires not only that these receptor specific molecular structures be labeled with β^+ -emitters, but also that the pattern of their biodistribution *in vivo* reflects receptor binding and that models be developed which allow the extraction of quantitative information from PETT data. One of the most interesting receptor systems from the standpoint of its implication in various disease states is the dopamine receptor (Seeman, 1980). Although dopamine itself was labeled with ¹¹C more than a decade ago (Christman et al. 1970), its use in the study of brain dopamine receptors is precluded by its inability to cross the blood-brain barrier. 3,4-Dihydroxyphenylalanine, a precursor of dopamine which crosses the blood-brain barrier, has been labeled with ¹¹C (Reiffers et al. 1977) and ¹⁸F and the behavior of the fluoro analog studied in animals (Firnau et al. 1973; Garnett et al. 1980). Perhaps the most widely sought after β^+ -emitter-labeled dopamine receptor ligand is spiroperidol, a butyrophenone neuroleptic drug which acts as a dopamine antagonist and has a very high affinity for the dopamine receptor. Since spiroperidol contains fluorine, routes to both ¹¹C- and ¹⁸F-labeled spiroperidol have been sought. The synthesis of high specific activity ¹⁸F-spiroperidol as well as haloperidol has been described using the triazene decomposition method and ¹⁸F-labeled fluoride as the labeled precursors (Tewson et al. 1981; Tewson et al. 1980; Maeda et al. 1981). This method, in contrast to the Schiemann reaction which was used earlier to prepare ¹⁸F-haloperidol (Digenis et al. 1981), provides a potentially very high specific activi-

ty product but suffers the disadvantages of low chemical yield and production of complex reaction mixtures. A route to no-carrier-added ^{11}C -labeled spiroperidol which uses ^{11}C -cyanide has been devised and provides this labeled dopamine receptor antagonist in a 20%–25% radiochemical yield (Fowler et al. 1982). The specific activity of this NCA radiotracer was determined by radioreceptor assay to be 2.6 Ci/ μmol (EOB). Studies in rats with ^{11}C -spiroperidol have shown that specific binding is attained at early times, suggesting that the half-life of ^{11}C is compatible with its use to probe the dopamine receptor (Arnett et al., to be published). Recently, serial studies with ^{11}C -spiroperidol using unanesthetized, conditioned baboons as subjects and pharmacological intervention have shown that with PETT VI in the high-resolution mode (~ 7 mm) regional localization can be detected in spite of the small brain size (Arnett and Wolf 1982, unpublished results). Here again it should be reemphasized that one of the great advantages with ^{11}C is that of being able to do serial studies in the same subject at short time intervals. Other dopamine receptor ligands which have been prepared recently include radiobrominated spiroperidol (Kulmala et al. 1981) and ^{11}C -labeled pimozone (Crouzel et al. 1980).

At the present time a rather large number of agonists and antagonists of neurotransmitter receptors are under investigation. For example, benzodiazepine receptors have been studied in the live baboon using ^{11}C -flunitrazepam (Comar et al. 1979), and a synthesis has been reported labeling the opiate agonist, etorphine, with ^{11}C (Maziere et al. 1981a). Recently, myocardial acetylcholine-muscarine receptors have been imaged using ^{11}C -labeled quinuclidinyl benzilate methiodide (Maziere et al. 1981b). The use of β^+ -emitter-labeled receptor ligands might prove especially fruitful if the effectiveness of drug regimens could be quantitatively evaluated in humans. The application to psychiatric disorders is of particular interest. It should be kept in mind, however, that the exquisite sensitivity of autoradiographic methods in animals using tritium cannot be matched with PETT because of the ultimate limiting resolution of the method.

Interest in the synthesis of β^+ -emitter-labeled amino acids has been stimulated by the recent

development of a model for quantitating regional brain protein synthesis (Smith et al. 1980). This particular model was developed with 1- ^{14}C -leucine, and its extension to humans requires that amino acids be synthesized in high specific activity and that the radiolabel be ^{11}C in the carboxyl carbon. The synthesis of 1- ^{11}C -L-leucine and the use of PETT and the model developed for autoradiographic studies has been recently applied to the measurement of regional brain protein synthesis in humans (Barrio et al. 1982a). Brain protein synthesis has also been measured using methyl- ^{11}C -methionine (Comar et al. 1976). D,L-Mixtures of high specific activity ^{11}C -amino acids have been synthesized by the Strecker synthesis (Washburn et al. 1979) and by the carboxylation of α -lithioisonitriles (Vaalburg et al. 1976). A recent breakthrough in the synthesis of ^{11}C -amino acids has been the HPLC resolution of D,L-valine using reverse phase HPLC column and a chiral mobile phase (Washburn et al. 1982). D,L-Tryptophan has been resolved using a bovine serum albumin bound to a sepharose column (Wu et al. 1981). Another resolution technique uses oxidative deamination with immobilized L- and D-amino acid oxidase (Digenis et al. 1979). These rapid resolution methods offer the possibility of doing studies on amino acid metabolism using the physiologically active L-isomer, thus reducing background activity as well as the radiation dose resulting from the D-isomer.

The synthesis of amino acids by enzymatic methods is of continuing interest and is the subject of a recent review (Gelbard 1981). Early methods focused on ^{13}N labels, but with the need for a longer half-life, ^{11}C is receiving increasing attention. For example, a method for the rapid enzymatic synthesis of ^{11}C -aspartic acid reported more than a decade ago (Hara et al. 1971) has been recently refined using purified, immobilized enzymes (Barrio et al. 1982b). Fluorine-18 may also prove to be useful as a label, especially in the high molecular weight amino acids such as tryptophan (Atkins et al. 1972), if the labeled amino acid is a good substitute for the naturally occurring amino acid in the process, etc., being studied. When imaging or localization of a tumor (etc.) is of interest, the requirements for the position of an "unnatural" label or for a structure identical or

nearly identical to what is involved in a biochemical process are less demanding than when quantitative information on specific biochemical or metabolic processes is sought.

While the major synthetic effort in recent years has centered on saccharides, neuroleptics, and to a lesser extent, amino acids, active efforts in labeling other classes of compounds have been in progress. Uptake of polyamines in the prostate and prostate tumors has been demonstrated (Welch et al. 1977; Winstead et al. 1980).

^{11}C -labeled alkyl monoamines which are sequestered by the capillary endothelium of the lung might be used as diagnostic agents for the biogenic amine-regulating function of the lung (Fowler et al. 1976a, b). Another area of interest is the labeling of purine, pyrimidines, nucleotides, and nucleosides (Fowler et al. 1973; Vine et al. 1979; Abrams et al. 1979; Shiue et al. 1979b; Christman et al. 1972; Crawford et al. 1978). The feasibility of using ^{18}F -5-fluorouridine to measure tissue proliferation via its incorporation into RNA is a promising technique for studying brain tumor growth rate and its response to chemotherapy (Crawford et al. 1982).

Conclusion

The combination of positron emission tomography and organic radiopharmaceuticals labeled with positron emitters is presently growing at an exponential rate. For many years accelerator-produced radionuclides played only a minor role in nuclear medicine and other medical disciplines. There is no question that on a clinical nuclear medicine procedure basis, technetium-99m is still the most important radionuclide. It is, however, also clear that accelerator-produced radionuclides, particularly thallium-201 and gallium-67 (iodine-123 in Europe), are rapidly gaining in importance. Where do the positron emitters ^{11}C , ^{13}N , ^{15}O , and ^{18}F fit in this picture? At present, they are not having their major impact in the more traditional areas of interest in nuclear medicine practice, but rather the efforts of the nuclear medicine physicians, physicists, and chemists have brought this multi- and interdisciplinary field to the attention of other medical specialties, e.g., neurology, cardiology, and oncology. Indeed, there is an upsurge in basic research in

the use of these positron emitters in biology, physiology, and biochemistry, a trend that the authors view as a necessity if the full potential of the varied disciplines is to be applied to medical practice. The key to the combination of nuclear medicine positron instrumentation and chemistry and medical application is the ability to probe the physiology and biochemistry of humans in normal and in pathologic states in a quantitative manner. It has already been demonstrated that regional brain glucose metabolism can be quantitated. The quantitation of regional cerebral blood flow at the same level of accuracy is not far off. This technique alone is opening up new areas of diagnostic sophistication in psychiatric disorders, stroke, and epilepsy, and may prove to be of utility in Huntington's disease, tardive dyskinesia, growth rates in brain tumors, senile dementia, and other disease states. In cardiology, functional and metabolic activity of small volumes of the myocardium can be probed, once suitable models have been perfected. As resolution of the tomographs increase, smaller volume elements can come under scrutiny. Perhaps the assessment of function in 1–2 cm^3 of tissue is the best that one can do today, but this will be reduced to 0.05–0.1 cm^3 in the not too distant future. In the field of oncology, the quantitation of drug regimen with suitable compounds is possible, as is the noninvasive quantitation of tumor growth rates or tumor ablation rates.

All of this depends on the development of suitable radiopharmaceuticals, models that allow evaluation of ingress and egress of the compound, and its manipulation by concurrent drug therapies in the particular volume element and continual improvement of the positron tomographic instrumentation. Single photon tomography as a modality should not be overlooked as being complementary and also allowing the use of a wider variety of radionuclides in labeled compounds, albeit radionuclides that are foreign to the physiology and biochemistry of humans.

As the use of β^+ -emitter-labeled radiotracers proliferates, questions regarding the biological consequences of the decay event itself have begun to be addressed. The decay of a β^+ -emitter leaves behind a multiply charged atom which leaves the site originally binding the β^+ -emitter-labeled radiotracer. The molecular "debris"

left behind by this decay event can be in the form of a totally fragmented molecule or a highly excited molecular species which can then form a new molecule which may or may not influence its surroundings in a deleterious way. While the chemical and biological consequences of β -decay have been reviewed in detail (Halpern and Stocklin 1977), the biological consequences of β^+ -decay have only recently received attention (Kassis et al. 1981). As the use of β^+ -emitters proliferates in medical procedures, this area must receive attention. This is particularly apparent when one considers the fact that sites connected with compounds labeled with positron emitters can include receptors, nuclear material, and other physiologically active areas, to which damage could lead to irreversible changes.

The cyclotron is no longer an instrument requiring elaborate support facilities for its operation and maintenance. It has already been developed to the point where it can be operated by the nonspecialist. True automation of operation of the accelerator and the production of radiopharmaceuticals of proven utility is actively being developed here and abroad. The routine delivery of compounds such as ^{11}C -palmitate and ^{18}F -2-deoxy-2-fluoro-D-glucose is a reality today, allowing daily diagnostic procedures to be carried out in those centers with the available facilities. To be sure, it has not been developed to the point where a hospital pharmacy can be called upon to provide this service, but this can become a reality soon.

Acknowledgment. This research was carried out at Brookhaven National Laboratory in part by contract DE-AC02-76CH00016 with the U.S. Department of Energy and supported by its Office of Basic Energy Sciences, and the Office of Health and Environmental Research. The authors are indebted to Grune and Stratton, the publishers of Seminars in Nuclear Medicine, for allowing them to use in substantial part the material published in Wolf, A. P., Seminars in Nuclear Medicine XI, 2-12 as the basis for this article.

References

1. Abrams DN, Knaus EE, Wiebe LI et al. (1979) ^{18}F -5-fluoro-2'-deoxyuridine: a potential diagnostic agent for tumor metabolism. *J Label Compd Radiopharm* 16: 155-156
2. Arnett CD, Fowler JS, Wolf AP et al. (to be published) Specific binding of [^{11}C] spiroperidol in rat brain in vivo. *J Neurochem*
3. Atkins HL, Christman DR, Fowler JS et al. (1972) ^{18}F -labeled 5- and 6-fluorotryptophan. *J Nucl Med* 13: 713-719
4. Barrio JR, Phelps ME, Huang S-C et al. (1982a) Positron-emitting labeled L-amino acids for measurement of protein synthesis. *Trans Am Nucl Soc* 41: 17-18
5. Barrio JR, Egbert JE, Henze A et al. (1982b) L-[^{11}C] aspartic acid: enzymatic synthesis, myocardial uptake and metabolism. *J Med Chem* 25: 93-96
6. Beaver JE, Fernandez-Rubio F, Nash D et al. (1979) Radionuclide production aspects of medical cyclotrons. *IEEE Trans Nucl Sci* NS-26: 1703-1706
7. Christman DR, Karlstrom KI (eds) (1975) Accelerator-produced nuclides for use in biology and medicine, vol. I. A Bibliography 1939-1973. Brookhaven National Laboratory 50448-I, 1975. (Available from National Technical Information Service, U.S. Department of Commerce)
8. Christman DR, Hoyte RM, Wolf AP (1970) Organic radiopharmaceuticals labeled with isotopes of short half-life. I. Dopamine hydrochloride- ^{11}C . *J Nucl Med* 11: 474-478
9. Christman DR, Crawford EJ, Friedkin M et al. (1972) Detection of RNA synthesis in intact organisms with positron-emitting (methyl- ^{11}C) thymidine. *Proc Natl Acad Sci USA* 69: 988-989
10. Comar D, Cartron JC, Maziere M et al. (1976) Labeling and metabolism of methionine-methyl- ^{11}C . *Eur J Nucl Med* 1: 11-14
11. Comar D, Maziere M, Godot JM et al. (1979) Visualization of ^{11}C -flunitrazepam displacement in the brain of the live baboon. *Nature* 280: 329-331
12. Crawford EJ, Christman D, Atkins H et al. (1978) Scintigraphy with positron-emitting compounds. I. Carbon-11-labeled thymidine and thymidylate. *Int J Nucl Med Biol* 5: 61-69
13. Crawford EJ, Friedkin M, Wolf AP et al. (1982) ^{18}F -5-fluorouridine, a new probe for measuring the proliferation of tissue in vivo. *Advances in Enzyme Regulation* 20: 3-22
14. Crouzel C, Mestelan G, Kraus E et al. (1980) Synthesis of a ^{11}C -labeled neuroleptic drug: pimozide. *Int J Appl Radiat Isot* 31: 545-548
15. Digenis GA, Casey DL, Wesner DA et al. (1979) Preparation of optically active ^{11}C -amino acids. *J Nucl Med* 20: 662
16. Digenis GA, Vincent SH, Kook CS et al. (1981) Tissue distribution studies of [^{18}F] haloperidol,

- [^{18}F]- β -(4-fluorobenzoyl) propionic acid and [^{82}Br] bromperidol by external scintigraphy. *J Pharm Sci* 70: 985–989
17. Ehrin E, Westman E, Nilsson SO et al. (1980) A convenient method for production of ^{11}C -labeled glucose. *J Label Compd Radiopharm* 17: 453–461
 18. Firna G, Nahmias C, Garnett S (1973) The preparation of [^{18}F] 5-fluoro-DOPA with reactor-produced fluorine-18. *J Appl Radiat Isotope* 24: 182–184
 19. First International Symposium on Radiopharmaceutical Chemistry, Upton, NY, (1976). *J Label Compd Radiopharm* 13 (Abstracts)
 20. Fowler JS, Wolf AP (1982) The synthesis of carbon-11, fluorine-18 and nitrogen-13 labeled radiotracers for biomedical applications. NAS-NS-3101, National Academy of Sciences, National Research Council, National Technical Information Service, 1982
 21. Fowler JS, Finn RD, Lambrecht RM et al. (1973) The synthesis of 5-fluorouracil- ^{18}F . *J Nucl Med* 14: 63–64
 22. Fowler JS, Gallagher BM, MacGregor RR et al. (1976a) Carbon-11-labeled aliphatic amines in lung uptake and metabolism studies. Potential for dynamic measurements in vivo. *J Pharmacol Exp Ther* 198: 133–145
 23. Fowler JS, Gallagher BM, MacGregor RR et al. (1976b) ^{11}C -labeled octylamine a potential diagnostic agent for lung structure and function. *J Nucl Med* 17: 752–754
 24. Fowler JS, Gallagher BM, MacGregor RR et al. (1977) ^{11}C -serotonin a tracer for the pulmonary endothelial extraction of serotonin. *J Label Compd Radiopharm* 13: 194–195
 25. Fowler JS, MacGregor RR, Wolf AP et al. (1981) A shielded synthesis system for production of 2-deoxy-2-[^{18}F]fluoro-D-glucose. *J Nucl Med* 22: 376–380
 26. Fowler JS, Arnett CD, Wolf AP et al. (1982) [^{11}C] Spiroperidol: synthesis, specific activity determination and biodistribution in mice. *J Nucl Med* 23: 437–445
 27. Gallagher BM, Fowler JS, MacGregor RR et al. (1979) In vivo measurement of transport and metabolic processes using radiotracers. In: Colombetti SG (ed) *Principles of radiopharmacology*, Vol II. Cleveland, CRC, 135–154
 28. Garnett ES, Firna G, Nahmias C et al. (1980) Blood-brain barrier transport and cerebral utilization of dopa in living monkeys. *Am J Physiol* 238: R318–R327
 29. Gelbard AS (1981) Biosynthetic methods for incorporating positron-emitting radionuclides into compounds of biomedical interest. *J Label Compd Radiopharm* 18: 933–945
 30. Goldstein RA, Klein MS, Welch MJ et al. (1980) External assessment of myocardial metabolism with C-11 palmitate in vivo. *J Nucl Med* 21: 342–348
 31. Halpern A, Stocklin G (1977) Chemical and biological consequences of β -decay. Parts 1 and 2. *Rad and Environ Biophysics* 14: 167–183, 257–274
 32. Hara T, Taylor C, Lembares N et al. (1971) L-Aspartic acid-4- ^{11}C as a tumor-scanning agent. *J Nucl Med* 12: 361–362
 33. Hoffman EJ, Phelps ME, Weiss ES et al. (1977) Transaxial tomographic imaging of canine myocardium with ^{11}C -palmitic acid. *J Nucl Med* 18: 57–61
 34. Ido T, Wan CN, Fowler JS et al. (1977) Fluorination with F_2 . A convenient synthesis of 2-deoxy-2-fluoro-D-glucose. *J Org Chem* 42: 2341–2342
 35. Ido T, Wan CN, Casella V et al. (1978) Labeled 2-deoxy-D-glucose analogs. ^{18}F -labeled 2-deoxy-2-fluoro-D-glucose, 2-deoxy-2-fluoro-D-mannose, and ^{14}C -2-deoxy-2-fluoro-D-glucose. *J Label Compd Radiopharm* 14: 175–183
 36. Iwata R, Ido T, Saji H (1979) A remote controlled synthesis of ^{11}C -iodomethane for the practical preparation of ^{11}C -labeled radiopharmaceuticals. *J Appl Radiat Isotope* 30: 194–196
 37. Karlstrom KI, Christman DR (eds) (1978) Accelerator-produced nuclides for use in biology and medicine, vol. II. A bibliography 1974–1976. Brookhaven National Laboratory 50448-II, 1978. (Available from National Technical Information Service, U.S. Department of Commerce)
 38. Kassiss AI, Adelstein SJ, Fowler JS et al. (1981) Uptake and radiotoxicity of F-18-2-fluoro-2-deoxyglucose in mammalian cells. *J Nucl Med* 22: 45
 39. Knust EJ, Kupfernagel CH, Stocklin G (1979) Long chain F-18 fatty acids for the study of regional metabolism in heart and liver. *J Nucl Med* 20: 1170–1175
 40. Kulmala HK, Huang CC, Dinerstein RJ et al. (1981) Specific in vivo binding of ^{77}Br -p-bromospiroperidol in rat brain: a potential tool for gamma ray imaging. *Life Sci* 28: 1911–1916
 41. Lenzi GS, Jones T, McKenzie CG et al. (1978) Study of regional cerebral metabolism and blood flow relationships in man using the method of continuously inhaling oxygen-15 and oxygen-15 labeled carbon dioxide. *J Neurol Neurosurg Psychiatry* 41: 1–10
 42. Lerch RA, Ambos HD, Bergmann SR et al. (1981) Localization of viable, ischemic myocardium by positron-emission tomography with ^{11}C -Palmitate. *Circulation* 64: 689–699
 43. Levy S, Livni E, Elmaleh DR (1982) A new synthesis of 2-deoxy-2-[^{18}F]fluoro-D-glucose with anhydrous [^{18}F]fluoride. *J Nucl Med* 23: P107
 44. Lifton JF, Welch MJ (1971) Preparation of glucose labeled with 20-minute half-lived carbon-11. *Radiat Res* 45: 35–40
 45. MacGregor RR, Fowler JS, Wolf AP et al. (1981) A synthesis of ^{11}C -2-deoxy-D-glucose for regional metabolic studies. *J Nucl Med* 22: 800–803

46. Maeda M, Tewson TJ, Welch M (1981) Synthesis of high specific activity ^{18}F spiroperidol for dopamine receptor studies. *J Label Compd Radiopharm* 18: 102–103
47. Maziere M, Godot JM, Berger G et al. (1981a) ^{11}C -labeled etorphine for in vivo studies of opiate receptors in brain. *J Radioanal Chem* 62: 279–284
48. Maziere M, Comar D, Godot JM (1981b) In vivo characterization of myocardium muscarinic receptors by positron emission tomography. *Life Sci* 29: 2391–2397
49. Palmer AJ, Goulding RW (1974) Biosynthetic preparation of ^{11}C -labelled galactose, glycerol and mannitol. *J Label Compd Radiopharm* 10: 627–635
50. Palmer AJ, Clark JC, Goulding RW (1977) The preparation of fluorine-18-labelled radiopharmaceuticals. In: Welch MJ (ed) *Radiopharmaceuticals and other compounds labelled with short-lived radionuclides*. Pergamon, New York, pp. 53–67
51. Phelps ME (1981) Positron computed tomography of cerebral glucose metabolism in man: theory and application in nuclear medicine. *Sem Nucl Med* 11: 32–49
52. Poe NP, Robinson GD Jr, MacDonald NS (1975) Myocardial extraction of labelled long chain fatty acid analogs. *Proc Soc Exp Biol Med* 118: 215–218
53. Raichle ME (1979) Quantitative in vivo autoradiography with positron emission tomography. *Brain Res Rev* 1: 47–68
54. Raichle ME, Larson KB, Phelps ME et al. (1979) In vivo measurement of brain glucose transport and metabolism employing glucose- ^{11}C . *Am J Physiol* 228: 1936–1948
55. Rainbow TC, Biegan A, Bleisch WV (1982) Quantitative autoradiography of neurochemicals. *Life Sci* 30: 1769–1774
56. Reiffers S, Beerling E, Vaalburg W et al. (1977) Synthesis of carbon-11-labelled Dopa. *J Label Compd Radiopharm* 13: 198–199
57. Reivich M, Kuhl D, Wolf A et al. (1979) The ^{18}F fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
58. Reivich M, Alavi A, Wolf A et al. (1982) The use of 2-deoxy-D $[1-^{11}\text{C}]$ glucose for the determination of local cerebral glucose metabolism in humans: variation within and between subjects. *J Cereb Blood Flow Metab* 2: 307–319
59. Ruth TJ, Wolf AP (1979) “Small” accelerator, radionuclide and radiopharmaceutical production. *IEEE Trans Nucl Sci* NS-26: 1710–1712
60. Schelbert HR, Henze E, Phelps ME et al. (1982) Assessment of regional myocardial ischemia by positron-emission computed tomography. *Am Heart J* 103: 588–597
61. Second International Symposium on Radiopharmaceutical Chemistry, Oxford, England (1978). *J Label Compd Radiopharm* 16 (Abstracts)
62. Seeman P (1980) Brain dopamine receptors. *Pharmacol Rev* 32: 229–313
63. Shiue C-Y, Wolf AP (1981) The synthesis of $1-[^{11}\text{C}]\text{-D-glucose}$ for the measurement of brain glucose metabolism. *J Nucl Med* 22: 58
64. Shiue C-Y, MacGregor RR, Lade RE et al. (1978) The synthesis of $1-^{11}\text{C}\text{-2-deoxy-D-glucose}$ (C-11-2-DG) for measuring regional brain glucose metabolism in vivo. *J Nucl Med* 19: 676–677
65. Shiue C-Y, MacGregor RR, Lade RE et al. (1979a) A new synthesis of 2-deoxy-D-*arabino*-hexose (1-deoxy-D-glucose). *Carb Res* 74: 323–326
66. Shiue C-Y, Fowler JS, MacGregor RR et al. (1979b) The synthesis of $^{18}\text{F}\text{-5-fluorouridine}$ as a probe for measuring RNA synthesis and tumor growth rates in vivo. In: *Radiopharmaceuticals II*. Society of Nuclear Medicine, New York, pp. 259–264
67. Shiue C-Y, Salvadori PA, Wolf AP et al. (1982) A new improved synthesis of 2-deoxy- $2-[^{18}\text{F}]\text{fluoro-D-glucose}$ from $^{18}\text{F}\text{-acetyl hypofluorite}$. *J Nucl Med* 23: 899–903
68. Silvester DJ, Waters SS (1979) Radionuclide production. In: *Radiopharmaceuticals II*. Society of Nuclear Medicine, New York, 727–744
69. Smith CB, Davidsen L, Deibler G et al. (1980) A method for determination of local rates of protein synthesis in brain. *Trans Am Soc of Neurochem* 11: 94
70. Sokoloff L, Reivich M, Kennedy C et al. (1977) The ^{14}C deoxyglucose method for the measurement of local cerebral glucose utilization. *J Neurochem* 28: 897–916
71. Som P, Atkins HL, Bandoypadhyay D et al. (1980) A fluorinated glucose analog, 2-fluoro-2-deoxy-D-glucose (F-18): nontoxic tracer for rapid tumor detection. *J Nucl Med* 21: 670–675
72. Straatmann M (1977) A look at ^{13}N and ^{15}O . In: Welch MJ (ed) *Radiopharmaceuticals and other compounds labelled with short-lived radionuclides*. Pergamon, New York, pp. 13–21
73. Straatmann MG, Welch MJ (1973) The liquid chromatographic purification of carbon-11-labeled glucose. *J Appl Radiat Isotope* 24: 234–236
74. Ter-Pogossian MM, Klein MS, Markham J et al. (1980) Regional assessment of myocardial metabolic integrity in vivo by positron-emission tomography with ^{11}C -labeled palmitate. *Circulation* 61: 242–255
75. Tewson TJ, Gould KL (1982) Synthesis of 2-deoxy-2-fluoroglucose using fluoride ion as a source of fluorine. *J Nucl Med* 23: P109
76. Tewson TJ, Welch MJ, Raichle ME (1978) ^{18}F -labeled 3-deoxy-3-fluoro-D-glucose: synthesis and preliminary biodistribution data. *J Nucl Med* 19: 1339–1345
77. Tewson TJ, Raichle ME, Welch MJ (1980) Pre-

- liminary studies with ^{18}F -spiroperidol. A radioligand for in vivo studies of the dopamine receptors. *Brain Res* 192: 291–295
78. Tewson TJ, Maeda M, Welch MJ (1981) Preparation of no carrier added ^{18}F -aryl fluorides: scope and conditions for the reaction. *J Label Compd Radiopharm* 18: 21–23
79. Third International Symposium on Radiopharmaceutical Chemistry, St. Louis, MO (1980). *J Label Compd Radiopharm* 18 (Abstracts)
80. Tilbury RS, Laughlin JS (1974) Cyclotron production and radioactive isotopes for medical use. *Semin Nucl Med* 4: 245–257
81. Vaalburg N, Beerling E, van der Molen HP et al. (1976) Preparation of carbon-11-labelled phenylalanine and phenylglycine by a new amino acid synthesis. *J Appl Radiat Isotope* 27: 153–157
82. Vine EN, Young D, Vine WH et al. (1979) An improved synthesis of ^{18}F -5-fluorouracil. *J Appl Radiat Isotope* 30: 401–405
83. Washburn LC, Sun TT, Byrd BL et al. (1979) High level production of C-11 carboxyl labeled amino acids. In: *Radiopharmaceuticals II*. Society of Nuclear Medicine, New York, pp. 767–777
84. Washburn LC, Sun TT, Byrd BL et al. (1982) Production of L-[1- ^{11}C]valine by HPLC resolution. *J Nucl Med* 23: 29–33
85. Weiss ES, Hoffman EJ, Phelps ME et al. (1976) External detection and visualization of myocardial ischemia with ^{11}C -substrates in vivo and in vitro. *Circ Res* 39: 24–32
86. Welch MJ (1969) Role of the chemist in nuclear medicine. In: Quinn JL III (ed) *Yearbook in nuclear medicine*, vol. IV. Year Book Medical Publishers, Chicago, pp. 6–18
87. Welch MJ, Tewson TJ (1979) Radiopharmaceuticals for neurological studies. *Radiopharmaceuticals II*. New York Society of Nuclear Medicine, 201–219
88. Welch MJ, Coleman RE, Straatmann MG et al. (1977) Labeled methylated polyamine analogs: uptake in prostate and tumor in animal models. *J Nucl Med* 18: 74–78
89. Winstead MB, Lamb JF, Winchell HS (1973) Relationship of chemical structure to in vivo scintigraphic distribution patterns of ^{11}C compounds. I. ^{11}C -carboxylates. *J Nucl Med* 14: 747–754
90. Winstead MB, Dischino DD, Munder NA et al. (1980) Relationship of molecular structure to in vivo distribution of carbon-11-labeled compounds. VI. Carbon-11-labeled aliphatic diamines. *Eur J Nucl Med* 5: 165–169
91. Wolf AP (1971) The preparation of organic radiopharmaceuticals containing isotopes of short half-life. *Radioisotopy* 12: 499–518
92. Wolf AP (1977) Medical cyclotrons, vol I, Medical radionuclide imaging, IAEA, Vienna, 343–354
93. Wolf AP, Redvanly CS (1977) Carbon-11 and radiopharmaceuticals. In: Welch MJ (ed) *Radiopharmaceuticals and other compounds labelled with short-lived radionuclides*. Pergamon, New York, 29–49
94. Wolf AP, Fowler JS (1979) Organic radiopharmaceuticals: recent advances. *Radiopharmaceuticals II*. New York Society of Nuclear Medicine, 73–93
95. Wolf AP, Christman DR, Fowler JS et al. (1973) Synthesis of radiopharmaceuticals and labelled compounds using short-lived isotopes. *Radiopharmaceuticals and labelled compounds*, vol I. Vienna IAEA, 345–381
96. Wu JHC, Harper PV, Lathrop KA (1981) Separation of racemic tryptophan. *J Nucl Med* 22: 74

Biochemical Parameters in Radiopharmaceutical Design

By J. R. Barrio

Introduction

The advent of positron computed tomography (PCT) has permitted for the first time the non-invasive application of tracer kinetic principles to man for the assessment of local physiologic processes (Phelps 1981). This approach, referred to as physiologic tomography (Phelps et al. 1977), relies upon the selection of appropriate radiolabeled compounds to monitor specific biochemical processes and the development of tracer kinetic models for the measurement of these processes.

Cyclotron-produced positron-emitting radioisotopes of carbon, nitrogen, and oxygen allow labeling of a wide variety of compounds without disrupting their biochemical properties. Thus, substitution of ^{11}C (20.38 min, half-life), ^{13}N (9.96 min), or ^{15}O (2.03 min) for the natural isotopes of carbon, nitrogen, or oxygen, respectively, would render agents physiologically indistinguishable from their natural counterparts. In addition substrate analogs that trace a biochemical process in a predictable manner, e.g. $[1\text{-}^{11}\text{C}]2\text{-deoxy-D-glucose}$ (MacGregor et al. 1981; Padgett et al. 1982) and $[1^8\text{F}]2\text{-deoxy-2-fluoro-D-glucose}$ ($[1^8\text{F}]2\text{-FDG}$) (Ido et al. 1978; Barrio et al. 1981; Fowler et al. 1981) are also available. ^{18}F (109.72 min) is used as a substitute for hydrogen due to the strong carbon-fluorine bond (109 kcal mol^{-1}) and to the similar Van der Waals radius of fluorine ($r_{\text{F}} = 1.35\text{ \AA}$) when compared with a hydrogen atom ($r_{\text{H}} = 1.10\text{ \AA}$) (Schlosser 1978), which minimizes steric hindrance.

Studies utilizing $[1^8\text{F}]2\text{-FDG}$ have yielded remarkable results (Phelps 1981; Phelps et al. 1981). Its utilization for the measurement of the cerebral metabolic rate of glucose (CMRGlc) is based upon the application of the basic principles of the technique developed by Sokoloff et al. (1977) for the autoradiographic measurement of local organ glucose metabolism in animals using $[1\text{-}^{14}\text{C}]2\text{-deoxy-D-glucose}$. Using the

Sokoloff model (Sokoloff et al. 1977) the glucose analog 2-deoxy-D-glucose (2-DG) competes with glucose for the facilitated membrane transport (Lund-Anderson 1979) and for its phosphorylation to 2-DG-6-phosphate with hexokinase. 2-DG-6-phosphate is not a substrate for further metabolism (Sokoloff et al. 1977) and is essentially trapped in the tissue due to its low membrane permeability. Like 2-DG, 2-FDG has been shown by Bessell et al. (1972), Reivich et al. (1979), and Gallagher et al. (1977, 1978) to be a good substrate for hexokinase, and its utilization in man was validated by Phelps et al. (1979) and Huang et al. (1980). Although ^{11}C -labeled glucose was synthesized and a model developed for the measurement of CMRGlc (Raichle et al. 1975), $[1^8\text{F}]2\text{-FDG}$ is certainly the best example available today of the application of biochemical principles for studying metabolic processes with PCT. With this information at hand, the present paper will specifically discuss current successful applications of basic biochemical concepts to the design and synthesis of radiopharmaceuticals for physiologic studies in the brain.

Application of Biochemical Principles in Radiopharmaceutical Synthesis

When properly treated, enzymes are excellent performers in the chemistry laboratory. Enzymes display great structural and geometric specificity in binding and are particularly useful for the synthesis of radiopharmaceuticals with chiral centers. The potential utility of enzymes in synthesis was early observed by different investigators, and the concept applied to the preparation of ^{13}N - and ^{11}C -labeled L-amino acids. Today, among the numerous methods available for synthesizing amino acids labeled with ^{13}N or ^{11}C , enzymatic procedures are the most effective for regio- and stereospecifically

attaching the ^{13}N or ^{11}C label (e.g., with precursors like ^{13}N ammonia or ^{11}C CO₂) onto a functionalized carbon unit.

^{13}N Ammonia was initially used in the enzymatic synthesis of several amino acids using the batch method, but a major concern was the presence of potentially pyrogenic enzymes in the final preparation (Straatmann and Welch 1973; Cohen et al. 1973). This problem has been overcome using the concept of enzyme immobilization to bind enzymes to solid-state supports, including DEAE Sephadex and polyacrylamide gel lattice (Tosa et al. 1973; Pollak et al. 1980), silica beds (Cohen et al. 1974), and cyanogen bromide activated Sepharose (Havekes et al. 1974; Gelbard et al. 1979; Baumgartner et al. 1981). A large variety of L- ^{13}N amino acids have been synthesized using the concept of enzyme immobilization and successfully used for imaging osteogenic sarcoma, brain tumors, and heart in animals and humans (Gelbard et al. 1979, 1980; Baumgartner et al. 1981; Gelbard 1981; Henze et al. 1982).

The covalent immobilization of phosphoenolpyruvate carboxylase and glutamic/oxaloacetic acid transaminase to Sepharose by the cyanogen bromide activation method has recently permitted the enzymatic synthesis of L-[4- ^{11}C]aspartic acid, the first ^{11}C -labeled amino acid synthesized using immobilized enzymes (Barrio et al. 1982a). For the preparation of other ^{11}C -labeled amino acids, however, enzymatic procedures may not be possible for multiple reasons, inter alia, enzyme availability. By far the most efficient chemical synthesis of 1- ^{11}C -labeled amino acids has been reported by Washburn et al. (1979), using the high-pressure modification of the Bucherer-Strecker reaction.

A common problem in the chemical synthesis of the amino acids is that the products are enantiomeric mixtures. Therefore, a rapid and effective isolation of the L-amino acid is mandatory for physiologic studies in man using PCT. The enantiomers (D- and L-amino acids) can be separated by high-pressure liquid chromatography using a chiral mobile phase and a reversed-phase column (Gil-Av et al. 1980; Washburn et al. 1982). Resolution is excellent and the system resolves basic and acidic amino acids in addition to the neutral amino acids. Alternatively, isolation of L-amino acids can be

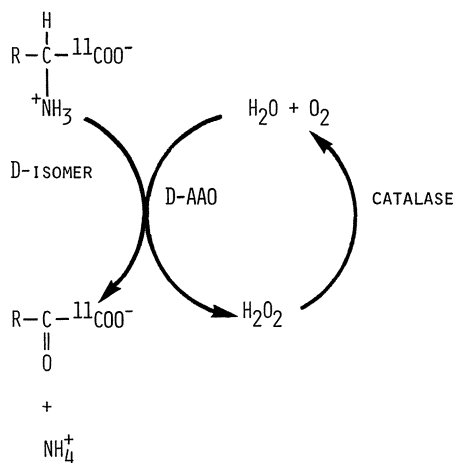


Fig. 1. Schematic representation of D-amino acid oxidation catalyzed by FAD-dependent D-amino acid oxidase (D-AAO) immobilized on a sepharose support. Oxygen is a specific proton acceptor that is produced in solution with immobilized catalase. The unmodified radiolabeled L-amino acid and the resulting radiolabeled α -ketoacid are subsequently separated by ion exchange chromatography

achieved by treating the enantiomeric mixture with D-amino acid oxidase, an enzyme that deaminates D-amino acids without affecting L-amino acids (Bright and Porter 1975; Casey et al. 1981; Barrio et al. 1982b). D-Amino acid oxidase is most active with D-isomers of neutral amino acids and least active with acidic or basic amino acids (Dixon and Kleppe 1965a). The D-isomers of alanine, methionine, and proline are among the best substrates for D-amino acid oxidase, whereas those of leucine, isoleucine, valine, phenylalanine, serine, and tryptophan are oxidized somewhat less rapidly. Arginine, lysine, histidine, glutamic acid, and aspartic acid are oxidized at a very low rate.

Barrio et al. (1982b) recently utilized immobilized D-amino acid oxidase (co-immobilized with catalase) to prepare L-[1- ^{11}C]leucine for quantitative determination of cerebral protein synthesis rates in man using PCT (Phelps et al. 1982). D-Amino acid oxidase is a light-sensitive flavoprotein oxidase that requires O_2 as a hydrogen acceptor (Bright and Porter 1975; Dixon and Kleppe 1965b) which, we found, could be efficiently supplied by immobilized catalase (Fig. 1). Covalent enzyme immobilization to cyanogen bromide activated Sepharose

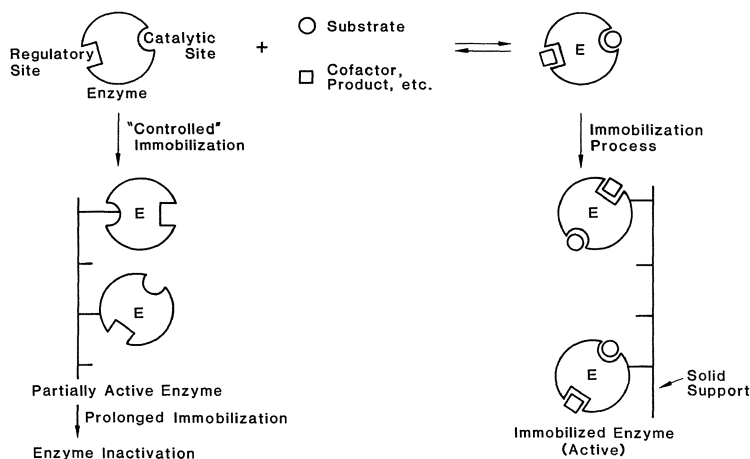


Fig. 2. Graphic representation of enzyme immobilization using activated solid supports, such as cyanogen bromide activated Sepharose. The presence of cofactors, products, and/or substrates protect the enzyme (*E*) active sites from covalent attachment to the solid support through nucleophiles present at these sites.

The immobilized enzyme will be, therefore, active. Prolonged immobilization in the absence of substrates or cofactors would produce inactivation of the enzyme. Enzyme activity can be partially preserved by short immobilization reactions

was performed in the presence of 1×10^{-6} M FAD. This procedure was preferred in this case over the "controlled" immobilization technique in the absence of substrates and cofactors (Fig. 2), because FAD is known to be necessary to prevent dissociation of the active holoenzyme (Dixon and Kleppe, 1965 a, b). Therefore immobilization was carried out when the protein was in its active holoenzyme form, in the absence of light < 550 nm, and in the presence of catalase, substrates, and other necessary cofactors to keep intact enzymatic activity of the coupled enzyme by protecting active sites from covalent linkage to the Sepharose support (Fig. 2).

We have extensively used the immobilization method described here and have found it to be very convenient and to give good yields of radiolabeled L-amino acid. The immobilized enzymes may be reused weeks after immobilization without appreciable loss in product yields. In addition protein loss after immobilization was essentially nonexistent as verified spectrophotometrically, which ensured apyrogenic preparations of radiolabeled amino acids.

An added advantage is that the enzyme-sepharose gels can be used directly in columns, without including supporting materials (Pollak et al. 1980). Columns having good mechanical

properties and flow characteristics were thus assembled. The enzyme columns were preserved by storage at 4°C in 2M KCl in order to inhibit bacterial growth, and they were thoroughly washed with the appropriate buffer prior to use.

Protein Biosynthesis and PCT: Selection of the Radiolabeled Amino Acid

As stated earlier analytical studies of physiological processes with PCT require labeled compounds that can be accurately modeled using the principles of tracer kinetics. L-Amino acids are the building blocks of proteins and, therefore, natural radiolabeled L-amino acids are the obvious candidates for quantitative evaluation of protein synthesis.

In the first stage of protein synthesis highly specific cytosolic aminoacyl-transfer RNA synthetases catalyze the esterification of the L-amino acids to their corresponding transfer-RNAs (tRNA) to form the aminoacyl tRNA. This energy-dependent process of activation of the amino acid is necessary for the formation of the polypeptide chain in the ribosome in a process

by which the aminoacyl tRNA is recognized by the messenger RNA coding system. It is obvious then that the specificity of the aminoacyl tRNA synthetases contributes to the specificity of the biosynthetic process and the chance of error under cellular conditions is less than 1:10000; these chances, however, seem to increase with age (Lehninger 1975). Under some conditions some amino acids analogs could be accepted by the specific aminoacyl tRNA synthetases instead of their normal substrates, e.g., p-halogenophenylalanine, ethionine, and norleucine. Para-chlorophenylalanine, for example, served as a substrate for phenylalanine tRNA synthetase of rat brain (Gäl 1974). It has been observed that p-fluorophenylalanine, an a priori attractive amino acid for PCT studies due to the feasibility of its labeling with ^{18}F (Goulding and Palmer 1972), is activated as well as phenylalanine with *E. coli* tRNA synthetase (Conway et al. 1962). In rabbit reticulocytes, however, the active synthetase showed less affinity for p-fluorophenylalanine than for phenylalanine (Arnstein and Richmond 1964). Later, p-fluorophenylalanine was considered unsuitable for quantitative studies in the brain, since 50% of the administered product was hydroxylated to tyrosine both in vivo and in vitro (Kaufman 1961; Gäl 1974).

At present the limited information possessed on membrane transport, protein synthesis incorporation, competitive enzyme inhibition, and metabolic behavior of these amino acid analogs does not make them *primary* targets for determination of cerebral protein synthesis rates using PCT. This is not to say, however, that some of these analogs labeled with short-lived positron emitters, e.g. [^{18}F]p-fluorophenylalanine, or [^{11}C]norleucine, could not be used in the future when a better understanding of their biochemistry is developed.

If we resort to the use of natural radiolabeled L-amino acids for in vivo quantitative determination of cerebral protein synthesis rates it will be important to select them very cautiously. Of paramount importance is the definition of the metabolic pathways that usually compete with protein synthesis. Ideally these metabolic pathways should be traceable and the metabolites eliminated from the tissue, allowing for the calculation of protein synthesis rates from the residual tissue activity. L-[1- ^{11}C]leucine is a good

candidate for investigation and validation of its use for the measurement of local cerebral protein synthesis rates in man and animals with PCT (Smith et al. 1980). Two distinctive biochemical features make L-[1- ^{11}C]leucine attractive for these studies: (a) L-leucine is transported across the blood-brain barrier (Oldendorf and Szabo 1976) [brain uptake index (BUI) = 54 ± 2], with L-phenylalanine (BUI = 55 ± 5) being the only other amino acid with a higher brain uptake in rats (Oldendorf 1971); (b). In addition to its incorporation into proteins the L-leucine metabolic pathway in brain, liver, and muscle proceeds via reversible transamination with α -ketoglutarate to form α -ketoisocaproate, which is then oxidatively decarboxylated in mitochondria (Chaplin et al. 1976; Williamson et al. 1979). This metabolic pathway, which competes with protein synthesis, results in the removal of the ^{11}C label as CO_2 (Fig. 3), and the residual activity in the tissue allows for the calculation of the protein synthesis rate. Phelps et al. (1982) have described initial kinetic PCT studies in man after intravenous injections of L-[1- ^{11}C]leucine, plasma sampling, and the application of a tracer kinetic model, originally developed by Smith et al. (1980), which allows for the calculation of transport, pool turnover rates, ratio metabolism/cerebral protein synthesis rate, rate constants for each process, and cerebral protein synthesis rates in monkeys and man.

By inspection of Fig. 3 one could expect that with L-leucine not only the position of the label but the type of labeling, e.g., ^{11}C or ^{13}N , would produce a different in vivo distribution pattern of positron activity. Figure 4 shows cerebral clearance curves for L-[1- ^{11}C]leucine and L-[^{13}N]leucine from carotid bolus injections in a monkey. The extraction fraction is about 28% and, as expected, equal for both radiolabeled amino acids. The longer time plot of the clearance curves clearly shows the difference between L-leucine labeled with ^{11}C in the carboxylic acid group and ^{13}N -labeled L-leucine.

The ^{13}N label is retained in the tissue by two different mechanisms: (a) incorporation of L-leucine into proteins and (b) transference of the ^{13}N label to a large pool of L-glutamate (Quastel 1979) in a transamination reaction catalyzed by the pyridoxal phosphate-dependent L-leucine transaminase. The ^{13}N -labeled

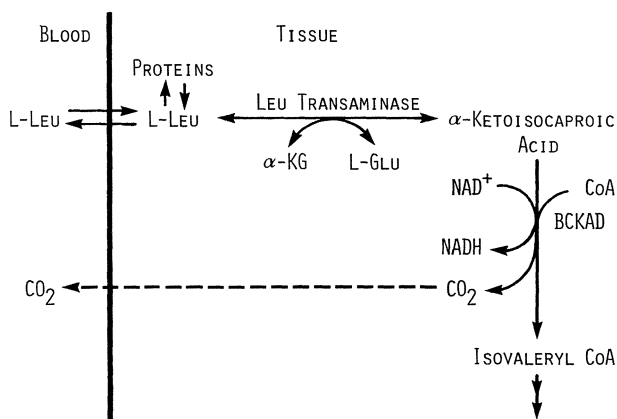


Fig. 3. Schematic illustration of L-leucine metabolic pathways for protein synthesis and metabolism. Abbreviations used are: *Leu*, leucine; α -KG, alpha-ketoglutaric acid; *Glu*, glutamic acid; NAD^+ , nicotinamide adenine dinucleotide; $NADH$, reduced form of NAD^+ ; *CoA*, coenzyme A; *BCKAD*, branched-chain ketoacid dehydrogenase

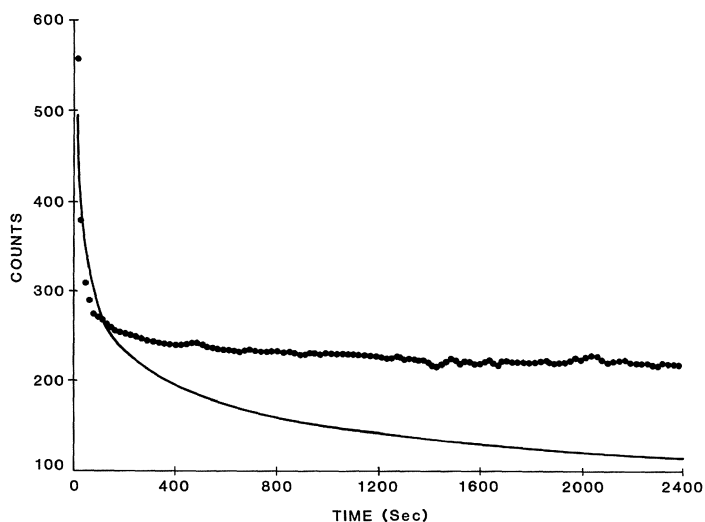


Fig. 4. Typical time course of activity in monkey brain subsequent to an intracarotid bolus injection of 0.2 ml of L-[1- ^{11}C]leucine (—) and L-[^{13}N]leucine (•••••). The activity peaks were normalized to 10^3 counts in both cases

L-glutamate formed is highly compartmentalized in the brain and retained because of its poor permeability of the blood-brain barrier, which is reflected by the long half-time (≈ 900 min) of the slow phase of the clearance curve.

The clearance of ^{11}C results from the transamination of L-leucine and subsequent irreversible decarboxylation of the α -ketoacid formed (Fig. 3) to yield ^{11}C CO_2 which diffuses out of the tissue. L-[1- ^{11}C]leucine incorporated into proteins is, as indicated earlier, retained in the tissue.

The results described by Phelps et al. (1982) on the use of L-[1- ^{11}C]leucine for the noninvasive determination of cerebral protein synthesis rates in man with PCT demonstrate the feasi-

bility of the approach shown here. On the other hand, the results of the first-pass extraction experiment indicate the difficulty of using L-[^{13}N]leucine, and most probably any L-[^{13}N] amino acids, for the *quantitative* evaluation of local protein synthesis rates using PCT.

Dopamine Neurotransmission and PCT

The nature of the interaction between receptor binding sites and effector hormones, the molecular structure of the receptors themselves, and their associated effector mechanisms are limiting factors in our understanding of brain receptor physiology. Central dopamine neurotrans-

mission plays a critical role in several diseases such as Huntington's chorea, Parkinson's disease, and possibly schizophrenia. These diseases have been poorly studied due to the difficulty in measuring localized biochemical events in the human brain. Postmortem tissue examinations, measurements of neurotransmitters and metabolites in body fluids, and pharmacological interventions have been the accessible methodology to study receptor properties and significant alterations resulting from these diseases.

The development of PCT has now made possible the potential mapping of neuroreceptors. The realization of such a goal, however, requires a careful selection of the radiotracers which should possess ideally a high affinity for the receptor ($K_A \geq 10^{10} M^{-1}$), high specificity (high tissue target/nontarget ratios), and adequate blood-brain barrier permeability. Further, the short-lived radionuclide-labeled radiotracer should have high specific activity, and a detailed knowledge of its kinetics and metabolism must be acquired for in vivo quantification of ligand-receptor binding. Finally, an understanding of the functional significance of changes in ligand binding is necessary for meaningful interpretation of human tomographic scans. Frost (1982) has recently made an elegant analysis of the necessary brain activity, specific activity requirements, and optimum imaging time required for in vivo, non-invasive quantification of regional dopamine receptor concentrations in man using PCT.

Biochemistry of Dopamine Receptors

A rapid examination of molecular models of dopamine indicates that it could exist as trans and gauche conformers (Fig. 5). Two extreme forms, I and II, which differ in the solid angle between the plane of the aromatic ring and a plane through the $CH_2CH_2NH_2$ group, are possible for the trans conformer. Experimental evidence indicates that I is the preferred conformer in both the crystal form (Bergin and Carlström 1968) and in solution (Giessner-Prettre and Pullman 1975). However there is no a priori reason to assume that a preferred conformation in the crystal or solution should be

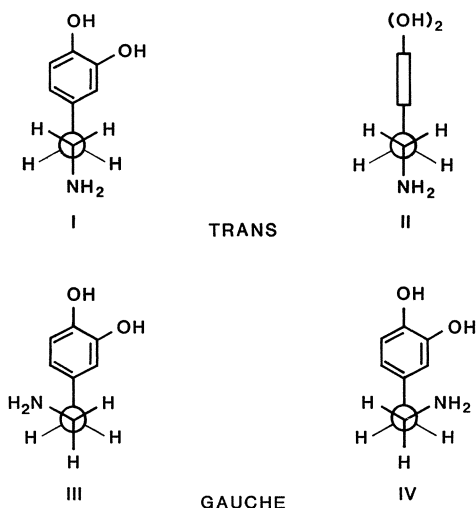


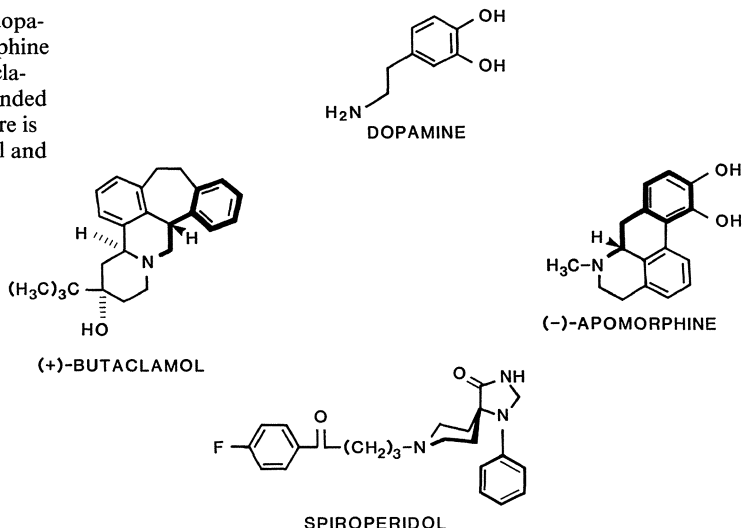
Fig. 5. Newman projections of dopamine in its trans and gauche conformations

that in which a ligand binds to a biological receptor. Pullman et al. (1972) have in fact noted that energy barriers separating anti and gauche conformers in phenethylamines are low, from which it could be concluded that dopamine can adapt its conformation to bind in vivo receptors (Cannon et al. 1979). Extensive pharmacological evidence from structure-activity relationship studies with semirigid analogs of trans conformers (Burn et al. 1982) seem to support conformer II as the pharmacologically active species.

For dopamine-like effects the critical interatomic distance between the nitrogen and the p-hydroxy group seems to be 7.8 Å (Rekker et al. 1972). Other synthetic agonists stereochemically related to trans conformers of dopamine, e.g., 5,6-dihydroxydimethylaminotetraline, also meet this criterion (Cannon 1975).

Recently, different semirigid molecular structures with unique chiralities and dopamine-antagonist effects have been synthesized. The most remarkable member of this series is (+)-butaclamol. (+)-Butaclamol contains within its polycyclic framework a phenethylamine moiety in the conformation in which endogenous dopamine interacts with dopamine receptors (Fig. 6). Conformational analysis based on the crystal structure of (+)-butaclamol revealed a striking similarity in distances between the piperidine nitrogen and the phenyl ring plane

Fig. 6. Chemical structures of dopamine; the agonist (–)-apomorphine and the neuroleptics (+)-butaclamol and spiroperidol. The extended phenethylamine pharmacophore is present in both (+)-butaclamol and (–)-apomorphine



of this semirigid ligand and the trans conformer of dopamine (Humber et al. 1975).

The extended phenethylamine moiety of (+)-butaclamol was assumed to be the pharmacophore responsible for the neuroleptic activity of this molecule. The observation that the dopamine receptor, which has genetically evolved to bind endogenous dopamine, also binds (+)-butaclamol with high affinity (Burt et al. 1975; Seeman et al. 1975) was a clear experimental indication of the complexity of the dopamine receptor biochemistry.

The initial conceptualization of the dopamine site comprised of a nitrogen location, and the aromatic binding site (Humber et al. 1975) had to be redefined (Humber et al. 1979a; Phillip et al. 1979). Since (+)-butaclamol is a much larger molecule than dopamine, it is obvious that other receptor features contribute to the observed high-affinity constant of (+)-butaclamol with central dopamine receptors. To explain this anomaly Humber et al. (1979a) proposed the existence of a highly lipophilic accessory binding site at the dopamine receptor (Fig. 7). From a careful and detailed study with analogs of (+)-butaclamol it was possible to define the probable dimensions of the accessory binding site on the central dopamine receptor (Humber et al. 1979a) and to specify its locus with respect to the primary binding sites.

It was the presence of an extended phenethyl-

DOPAMINE RECEPTOR TOPOGRAPHY

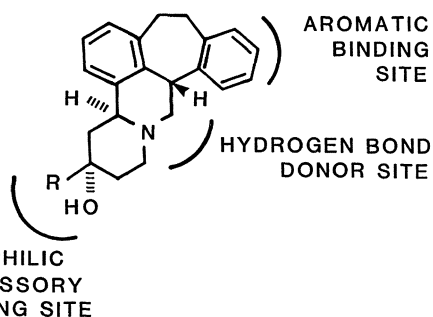


Fig. 7. Schematic representation of the central dopamine receptor binding sites derived from structural modifications in the neuroleptic (+)-butaclamol (Humber et al. 1979a; Phillip et al. 1979)

amine in (+)-butaclamol, also present in the agonist (–)-apomorphine (Fig. 6), which led to the study of the topographical features of the dopamine receptor. The unexpected observation that (+)-isobutaclamol, lacking the extended arylethylamine moiety, presents neuroleptic properties almost identical to that of (+)-butaclamol (Humber et al. 1979b) was rationalized in terms of the optimum distances between the critical centers required for recognition of the dopamine receptor.

The neuroleptic butyrophenones are chemically unrelated to dopamine or any of the agonists or antagonists discussed previously (Fig. 6). In

fact these drugs are a characteristic example of serendipity in medicinal chemistry, since they were developed from a group of meperidine analogs originally synthesized as analgesics. All potent neuroleptics in this series contain a p-substituted aromatic dialkylaminoalkyl ketone, with the tertiary basic nitrogen incorporated into a p-substituted piperidine, piperazine, or tetrahydropyridine ring. A piperidino derivative of this type, spiroperidol, is at present one of the most potent neuroleptics known.

Unfortunately our present knowledge does not permit a rationalization (but only speculation) of the molecular interaction between neuroleptic butyrophenones; e.g., spiroperidol, haloperidol, and the dopamine receptor. Unlike enzymes drug receptors are difficult to purify, which precludes systematic *in vitro* studies aimed at understanding the relationship between the solution conformation and the receptor-bound conformation of drugs. Therefore, dopamine receptors are at present mainly characterized on the basis of physiological and pharmacological criteria (Creese et al. 1975, 1976; Kebabian and Calne 1979). The demonstration of a dopamine-sensitive adenylyl cyclase, an enzyme that converts ATP into cyclic AMP, provided the basis for some of the studies on structure-activity relationship (dopamine agonists and antagonists) discussed above. It is clear now that not all physiological responses to dopamine involve the mediation of an adenylyl cyclase (Kebabian and Calne 1979), which illustrates the complexities of the various dopaminergic mechanisms.

Pharmacological characterization of the receptors is equally difficult. Due to the complexity of the pharmacological response (Creese et al. 1975; Niemegeers and Janssen 1979), accumulating evidence now favors the existence of several subclasses of endogenous dopamine binding sites, *inter alia*: (a) binding sites with high affinity for dopamine agonists and antagonist ligands; (b) binding sites with high affinity for dopamine agonists and low affinity for antagonist ligands; and (c) binding sites with low affinity for dopamine agonist ligands and high affinity for antagonist ligands. It has been recently argued, however, that a single macromolecule complex subjected to conformational changes in the presence or absence of

cooperatively linked serotonin or endogenous noradrenergic transmitters, could play the functional role in the central dopamine receptor physiological action (Cools 1981). In any event this attractive concept remains to be unequivocally proved.

Selection of the Ligand for Quantitative Determination of Central Dopamine Receptors with PCT

The choice of the radiotracer ligand for *in vivo* imaging of dopamine receptors is critical, as evidenced by an early attempt with ^{11}C -labeled chlorpromazine (Comar et al. 1979). In that study the brain radioactivity following intravenous administration of no carrier added ^{11}C (methyl) labeled chlorpromazine (specific activity $\approx 500 \text{ Ci/mmol}$) (Berger et al. 1979) was homogeneous in gray matter in 22 schizophrenic patients, preventing observation of specific receptor binding.

This experiment exemplifies the fact that many problems must be solved before quantitative localization of dopamine receptors can be accomplished with PCT. Much valuable information may be obtained toward solving some of these problems using *in vivo* autoradiography in animals (Kuhar et al. 1978). Animal studies suggest, for example, that spiroperidol is superior to both haloperidol and pimozide for *in vivo* imaging (Laduron et al. 1978a, 1978b).

A concentrated effort to label spiroperidol with positron-emitting radionuclides is presently taking place in several laboratories, after initial attempts on the synthesis of [^{18}F]haloperidol (Tewson et al. 1980). Most neuroleptic butyrophenones, e.g., spiroperidol, have a fluorine substituent at the p-position of the phenyl ring attached to the keto group and could, *a priori*, be radiolabeled with either ^{11}C or ^{18}F . Since the [^3H]spiroperidol-binding striatum: cerebellum ratio in rat at 60 min is about 6 and slowly increases with time (Kuhar et al. 1978), the ^{18}F label should be preferred to ^{11}C for PCT imaging at times when high target: nontarget binding ratios are observed – particularly since the equivalent times in man will be significantly longer due to the lower rate of blood flow dependent clearance of ligand associated with nonspecific sites.

An important consideration is that spiroperidol does not specifically bind to dopamine receptors. Palacios et al. (1981) have shown that [^3H]spiroperidol also binds serotonin receptors and α -adrenergic receptors. Murrin and Kuhar (1979) reduced the serotonergic component in the in vivo binding of [^3H]spiroperidol to frontal cortex by administration of pipamperone, a drug with high affinity for serotonergic components, 1 h after administration of [^3H]spiroperidol. This treatment eliminated the autoradiographically demonstrated ligand concentration in frontal cortex areas associated with serotonergic receptors when [^3H]spiroperidol was administered alone.

[^3H]Pimozide, a neuroleptic butyrophenone with high affinity for dopamine receptors (similar to [^3H]spiroperidol), is more selective than [^3H]spiroperidol, but due to the higher lipophilicity of pimozide is not a convenient selection for in vivo studies. Pimozide has higher nonspecific binding than spiroperidol following intravenous administration as evidenced by higher retention of [^3H]pimozide than [^3H]spiroperidol in cerebellum, an area essentially free of dopamine receptors (Laduron and Leyson 1977).

In spite of its binding to multiple receptors, [^{18}F]spiroperidol seems to be the best candidate for in vivo mapping of dopamine receptors in the brain using PCT due to its high affinity for the neuroleptic receptor, its low nonspecific binding, and its superiority to other available ligands for in vivo autoradiography in animals.

Conclusions

Analytical studies of physiological processes with PCT require labeled compounds that can be accurately modeled using tracer kinetic principles. Such tracer kinetic models could be used to estimate, inter alia, the rates of membrane transport, metabolism, and synthesis products. Moreover, the foundation of a tracer kinetic measurement lies upon the selection of appropriate radiotracers that behave in a predictable manner. With the increased complexity of the processes being studied using PCT, e.g., glucose metabolism, protein synthesis, and receptor binding, the understanding of the

cellular biochemical processes in which the radiotracers are involved has become necessary. In this chapter we have shown specific examples of the application of current biochemical concepts to areas of research that provide encouraging perspectives for their applications in man using tracer kinetic techniques and PCT, and how these concepts can be used in the design and synthesis of short-lived positron-emitting labeled radiopharmaceuticals.

Acknowledgments. The author is grateful to the many faculty colleagues, postdoctoral fellows, students, and research associates affiliated with our laboratories who contributed time and data, especially Drs. M.E. Phelps, N.S. MacDonald, S.-C. Huang, N. Satyamurthy, H.C. Padgett and Mssrs. R.E. Keen, C. Selin, D. Chugani, and J. Miller. This work was supported in part by DOE contract DE-AM03-76-SF00012 and National Institutes of Health grant 7R01-GM 24839.

References

1. Arnstein HRV, Richmond MH (1964) Utilization of p-fluorophenylalanine for protein synthesis by the phenylalanine-incorporation system from rabbit reticulocytes. *Biochem J* 91: 340–346
2. Barrio JR, MacDonald NS, Robinson GD et al. (1981) Remote, semiautomated production of ^{18}F -labeled 2-deoxy-2-fluoro-D-glucose. *J Nucl Med* 22: 372–375
3. Barrio JR, Egbert JE, Henze E et al. (1982a) L-[4- ^{11}C]aspartic acid: enzymatic synthesis, myocardial uptake and metabolism. *J Med Chem* 25: 93–96
4. Barrio JR, Phelps ME, Huang S-C et al. (1982b) Positron-emitting labeled L-amino acids for measurement of protein synthesis. *Trans Am Nucl Soc* 41: 17–18
5. Baumgartner FJ, Barrio JR, Henze E et al. (1981) ^{13}N labeled L-amino acids for in vivo assessment of local myocardial metabolism. *J Med Chem* 24: 764–766
6. Berger G, Maziere M, Knipper R et al. (1979) Automated synthesis of ^{11}C -labeled radiopharmaceuticals: imipramine, chlorpromazine, nicotine and methionine. *Int J Appl Radiat Isot* 30: 393–399
7. Bergin R, Carlström D (1968) Structure of the pyrocatecholamines. II. crystal structure of dopamine hydrochloride. *Acta Crystallogr Sect B* 24: 1506–1510

8. Bessell EM, Foster AB, Westwood JH (1972) The use of deoxyfluoro-D-glucopyranoses and related compounds in a study of yeast hexokinase specificity. *Biochem J* 128: 199–204
9. Bright HJ, Porter DJT (1975) Flavoprotein oxidases. D-Amino acid oxidase. In: Boyer PD (ed) *The enzymes*, 3rd ed, vol 12, Academic, New York, pp 445–456
10. Burn P, Crooks PA, Heatley F et al. (1982) Synthesis and dopaminergic properties of some exo- and endo-2-aminobenzonorbornenes designed as rigid analogs of dopamine. *J Med Chem* 25: 363–368
11. Burt DR, Enna S, Creese I et al. (1975) Dopamine receptor binding in the corpus striatum of mammalian brain. *Proc Natl Acad Sci USA* 72: 4655–4659
12. Cannon JG (1975) Chemistry of dopaminergic agonists. In: Calne DB, Chase TN, Barbeau A (eds) *Advances in neurology*. Raven, New York, pp 177–183
13. Cannon JG, Perez Z, Long JP et al. (1979) *N*-alkyl derivatives of ($\pm$)- α -methyl-dopamine. *J Med Chem* 22: 901–907
14. Casey DL, Digenis GA, Wesner DA et al. (1981) Preparation and preliminary tissue studies of optically active ^{11}C D- and L-phenylalanine. *Int J Appl Radiat Isot* 32: 325–330
15. Chaplin ER, Goldberg AL, Diamond I (1976) Leucine oxidation in brain slices and nerve endings. *J Neurochem* 26: 701–707
16. Cohen MB, Spolter L, MacDonald NS et al. (1973) Production of ^{13}N -labeled amino acids by enzymatic synthesis. In: *New developments in radiopharmaceuticals and labeled compounds*. International Atomic Energy Agency, Vienna, vol 1, pp 269–290
17. Cohen MB, Spolter L, Chang CC et al. (1974) Immobilized enzymes in the production of radiopharmaceutically pure amino acids labeled with ^{13}N . *J Nucl Med* 15: 1192–1195
18. Comar D, Zarifian E, Verhas M et al. (1979) Brain distribution and kinetics of ^{11}C -chlorpromazine in schizophrenics: positron emission tomography studies. *Psychiatry Res* 1: 23–29
19. Conway TW, Lansford EM, Shive W (1962) Purification and substrate specificity of a phenylalanine-activating enzyme from *Escherichia coli* 9723. *J Biol Chem* 237: 2850–2854
20. Cools AR (1981) The puzzling cascade of multiple receptors for dopamine. *Trends Pharmacol Sci* 2: 178–183
21. Creese I, Burt DR, Snyder SH (1975) Dopamine receptor binding: differentiation of agonist and antagonist states with [^3H]dopamine and [^3H]haloperidol. *Life Sci* 17: 993–1002
22. Creese I, Burt DR, Snyder SH (1976) Dopamine receptor binding predicts clinical and pharmacological potencies of antischizophrenic drugs. *Science* 192: 481–483
23. Dixon M, Kleppe K (1965a) D-amino acid oxidase. I. dissociation and recombination of the holoenzyme. *Biochim Biophys Acta* 96: 357–367
24. Dixon M, Kleppe K (1965b) D-amino acid oxidase. II. specificity, competitive inhibition and reaction sequence. *Biochim Biophys Acta* 96: 368–382
25. Fowler JS, MacGregor RR, Wolf AP et al. (1981) A shielded synthesis system for production of 2-deoxy-2-[^{18}F]fluoro-D-glucose. *J Nucl Med* 22: 376–380
26. Frost JJ (1982) Pharmacokinetic aspects of the in vivo, noninvasive study of neuroreceptors in man. In: Eckelman WC (ed) *Receptor binding radiotracers*. CRC Boca Raton, FL, pp. 25–39
27. Gál EM (1974) Synthetic p-halogenophenylalanines and protein synthesis in the brain. *Ciba Found Symp* 22: 343–359
28. Gallagher BM, Ansari A, Atkins H et al. (1977) Radiopharmaceuticals XXIII. ^{18}F -labeled glucose metabolism in vivo: tissue distribution and imaging studies in animals. *J Nucl Med* 18: 990–996
29. Gallagher BM, Fowler JS, Gutterson NI et al. (1978) Metabolic trapping as a principle of radiopharmaceutical design. The factors responsible for the biodistribution of ^{18}F -2-deoxy-2-fluoro-D-glucose (^{18}F FDG). *J Nucl Med* 19: 1154–1161
30. Gelbard AS (1981) Biosynthetic methods for incorporating positron-emitting radionuclides into compounds of biomedical interest. *J Labelled Compd Radiopharm* 18: 933–945
31. Gelbard AS, Benua RS, Laughlin JS et al. (1979) Quantitative scanning of osteogenic sarcoma with nitrogen-13-labeled L-glutamate. *J Nucl Med* 20: 782–784
32. Gelbard AS, Benua RS, Reiman RE et al. (1980) Imaging of the human heart after administration of L-(^{13}N)glutamate. *J Nucl Med* 21: 988–991
33. Gil-Av E, Tishbee A, Hare PE (1980) Resolution of underivatized amino acids by reversed-phase chromatography. *J Am Chem Soc* 102: 5115–5117
34. Giessner-Prettre C, Pullman B (1975) Molecular-orbital study of the orthobenzylic long-range proton-proton coupling constant $^4J_{\text{HH}}$ in biological phenethylamines. *J Magn Reson* 18: 564–568
35. Goulding RW, Palmer AJ (1972) The preparation of fluorine-18-labeled p-fluorophenylalanine for clinical use. *Int J Appl Radiat Isot* 23: 133–137
36. Havekes L, Buckmann F, Visser J (1974) Immobilized glutamate dehydrogenase: some catalytic and structural aspects. *Biochim Biophys Acta* 334: 272–286
37. Henze E, Schelbert HR, Barrio JR et al. (1982) Evaluation of myocardial metabolism with ^{13}N and ^{11}C -labeled amino acids and positron computed tomography. *J Nucl Med* 23: 671–681
38. Huang SC, Phelps ME, Hoffman EJ et al. (1980) Noninvasive determination of local cerebral met-

- abolic rate of glucose in man. *Am J Physiol* 238: E69–E82
39. Humber LG, Bruderlein FT, Voith K (1975) Neuroleptic agents of the benzocycloheptapyridoisoquinoline series. A hypothesis on their mode of interaction with the central dopamine receptor. *Mol Pharmacol* 11: 833–840
40. Humber LG, Bruderlein FT, Phillip AH et al. (1979a) Mapping the dopamine receptor. I. features derived from modifications in ring E of the neuroleptic butaclamol. *J Med Chem* 22: 761–767
41. Humber LG, Phillip AH, Voith K et al. (1979b) (+) Isobutaclamol: a crystallographic, pharmacological and biochemical study. *J Med Chem* 22: 899–901
42. Ido T, Wan CN, Casella V et al. (1978) Labeled 2-deoxy-D-glucose analogs. ^{18}F -labeled 2-deoxy-2-fluoro-D-glucose, 2-deoxy-2-fluoro-D-mannose and ^{14}C -2-deoxy-2-fluoro-D-glucose. *J Labelled Compd Radiopharm* 14: 175–183
43. Kaufman S (1961) Enzyme conversion of 4-fluorophenylalanine to tyrosine. *Biochim Biophys Acta* 51: 619–621
44. Keabian JW, Calne DB (1979) Multiple receptors for dopamine. *Nature* 277: 93–96
45. Kuhar MJ, Murrin LC, Malouf AT et al. (1978) Dopamine receptor binding in vivo: the feasibility of autoradiographic studies. *Life Sci* 22: 203–210
46. Laduron P, Leyson J (1977) Specific in vivo binding of neuroleptic drugs in rat brain. *Biochem Pharmacol* 26: 1003–1007
47. Laduron PM, Janssen FM, Leyson JE (1978a) Spiperone: a ligand of choice for neuroleptic receptors. 1. kinetics and characteristics of in vitro binding. *Biochem Pharmacol* 27: 307–316
48. Laduron PM, Janssen FM, Leyson JE (1978b) Spiperone: a ligand of choice for neuroleptic receptors 2. regional distribution and in vivo displacement of neuroleptic drugs. *Biochem Pharmacol* 27: 317–321
49. Lehninger AL (1975) *Biochemistry* (2nd ed). Worth, New York
50. Lund-Anderson H (1979) Transport of glucose from blood to brain. *Physiol Rev* 59: 305–352
51. MacGregor RR, Fowler JS, Wolf AP et al. (1981) A synthesis of 2-deoxy-D-[^{11}C]glucose for regional metabolic studies: concise communication. *J Nucl Med* 22: 800–803
52. Murrin LC, Kuhar MJ (1979) Dopamine receptors in the rat frontal cortex: an autoradiographic study. *Brain Res* 177: 279–285
53. Niemegeers CJE, Janssen PAJ (1979) A systematic study of the pharmacological activities of dopamine antagonists. *Life Sci* 24: 2201–2216
54. Oldendorf WH (1971) Brain uptake of radiolabeled amino acids, amines and hexoses after arterial injection. *Am J Physiol* 221: 1629–1639
55. Oldendorf WH, Szabo J (1976) Amino acid assignment to one of three blood-brain barrier amino acid carriers. *Am J Physiol* 230: 94–98
56. Padgett HC, Barrio JR, MacDonald NS et al. (1982) The unit operations approach applied to the synthesis of [^{11}C]2-deoxy-D-glucose for routine clinical applications. *J Nucl Med* 23: 739–744
57. Palacios JM, Niehoff DL, Kuhar MJ (1981) ^3H -Spiperone binding sites in brain: Autoradiographic localization of multiple receptors. *Brain Res* 213: 277–289
58. Phelps ME (1981) Positron computed tomography studies of cerebral glucose metabolism in man: theory and applications in nuclear medicine. *Sem in Nucl Med* 11: 32–49
59. Phelps ME, Hoffman EJ, Kuhl DE (1977) Physiologic tomography: a new approach to in vivo measure of metabolism and physiologic function. In: *Medical radionuclide imaging*. International Atomic Energy Agency, Vienna, vol 1, pp 233–253
60. Phelps ME, Huang SC, Hoffmann EJ et al. (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with ^{18}F 2-fluoro-2-deoxy-D-glucose: validation of method. *Ann Neurol* 6: 371–388
61. Phelps ME, Kuhl DE, Mazziotta JD (1981) Metabolic mapping of the brain's response to visual stimulation: Studies in humans. *Science* 211: 1445–1448
62. Phelps ME, Barrio JR, Huang S-C et al. (1982) The measurement of local cerebral protein synthesis in man with positron computed tomography (PCT) and [^{14}C]L-leucine. *J Nucl Med* 23: 6
63. Phillip AH, Humber LG, Voith K (1979) Mapping the dopamine receptor. 2. features derived from modifications in the rings A/B region of the neuroleptic butaclamol. *J Med Chem* 22: 768–773
64. Pollak A, Blumenfeld H, Wax M et al. (1980) Enzyme immobilization by condensation copolymerization into cross-linked polyacrylamide gels. *J Am Chem Soc* 102: 6324–6336
65. Pullman B, Coubeils JL, Courriere P et al. (1972) Quantum mechanical study of the conformational properties of phenethylamines of biochemical and medicinal interest. *J Med Chem* 15: 17–23
66. Quastel JH (1979) The role of amino acids in the brain. *Essays Med Biochem* 4: 1–48
67. Raichle ME, Larson KB, Phelps ME et al. (1975) In vivo measurement of brain glucose transport and metabolism employing [^{11}C] glucose. *Am J Physiol* 228: 1936–1948
68. Reivich M, Kuhl D, Wolf A et al. (1979) The [^{18}F]fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
69. Rekker RF, Engel DJC, Nys GG (1972) Apomor-

- phine and its dopamine-like action. *J Pharm Pharmacol* 24: 589–591
70. Schlosser M (1978) Introduction of fluorine into organic molecules: why and how. *Tetrahedron* 34: 3–17
71. Seeman P, Chau-Wong M, Tedesco J et al. (1975) Brain receptors for antipsychotic drugs and dopamine. Direct binding assays. *Proc Natl Acad Sci USA* 72: 4376–4380
72. Smith CB, Davidsen L, Deibler G et al. (1980) A method for the determination of local rates of protein synthesis in brain. *Trans Am Soc Neurochem* 11: 94
73. Sokoloff L, Reivich M, Kennedy C et al. (1977) The [C-14]deoxy-glucose method for the measurement of local cerebral glucose utilization: theory, procedure and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
74. Straatmann MG, Welch MJ (1973) Enzymatic synthesis of ^{13}N -labeled amino acids. *Radiat Res* 56: 48–56
75. Tewson TJ, Raichle ME, Welch MJ (1980) Preliminary studies with [^{18}F]haloperidol: A radioligand for in vivo studies of the dopamine receptors. *Brain Res* 192: 291–295
76. Tosa T, Sato T, Mori T, et al. (1973) Continuous production of L-aspartic by immobilized aspartase. *Biotechnol Bioeng* 15: 69–84
77. Washburn LC, Sun TT, Byrd BL et al. (1979) High-level production of ^{11}C carboxyl-labeled amino acids. In: Sorenson JA (ed) *Radiopharmaceuticals II*. Society of Nuclear Medicine, New York, pp 767–777
78. Washburn LC, Sun TT, Byrd BL et al. (1982) Production of L-[1- ^{11}C]valine by HPLC resolution. *J Nucl Med* 23: 29–33
79. Williamson JR, Walajtys-Rode E, Coll KE (1979) Effects of branched chain alpha-ketoacids on the metabolism of isolated rat liver cells. *J Biol Chem* 254: 11511–11520

Cerebral Blood Flow

(Chairman: M. E. Raichle)

Regional Cerebral Blood Flow Measurement and Dynamic Positron Emission Tomography

By Y. L. Yamamoto, E. Meyer, D. Menon, P. Roland, and M. Diksic

Introduction

Over the past few years several positron emission tomographic techniques (PET) have been introduced for quantitative measurement of regional cerebral blood flow (rCBF). Quantitative rCBF measurement by PET can be subdivided into two categories, namely dynamic imaging and equilibrium imaging. However, our

experience with both methodologies indicates that these techniques have several practical limitations, including uncertain rCBF measurement as shown in Tables 1 and 2. We have therefore, proposed a new technique of dynamic plus equilibrium imaging to overcome these problems.

Dynamic Imaging

A major advantage of this methodology is its high temporal resolution requiring only a few minutes of examination time; it is thus suitable for measuring a rapidly changing rCBF as occurs in ictal and postictal states of epilepsy, and certain physiological activations. However, major drawbacks are the time limitation for sampling and the necessity of operating at a lower resolution mode to maximize counting efficiency with minimum statistical error. Dynamic imaging can be further subdivided into two groups, namely recirculating diffusible tracer and inert gas clearance methods. For the inert gas clearance method, we have developed a ^{77}Kr -bolus inhalation technique (Yamamoto et al. 1977, 1982).

Table 1. rCBF measurement by PET

● Dynamic imaging	● Equilibrium imaging
Diffusible tracer	Continuous inhalation of C^{15}O_2 (E-1)
• H_2^{15}O i. v. injection (D-1)	Continuous inhalation of N_2^{15}O (E-2)
• 2 min inhalation of C^{15}O_2 (D-2)	
Inert gas clearance	● Dynamic plus equilibrium imaging
• ^{77}Kr bolus inhalation (D-3)	Continuous inhalation of C^{15}O_2 (A-1)
^{13}N -labeled $^{13}\text{N}_2\text{O}$ (D-4)	Continuous inhalation of N_2^{15}O (A-2)

Table 2. rCBF methodologies by PET

	Subdivision	Arterial sampling	Examination time (min)	Spatial resolution	Administered dose (mCi)	Radiation dose (W.B.) (mrem)
Dynamic imaging	D-1	Yes	< 5	Low	100	> 100
	D-2	No	< 5	Low	4	< 7
	D-3	No	< 8	Low	10	< 8
	D-4	No	< 8	Low	10	< 10
Equilibrium imaging	E-1	Yes	> 20	High	20	36
	E-2	No	> 20	High	20	36
Dynamic plus equilibrium imaging	A-1	No	> 30	High	20	36
	A-2	No	> 30	High	20	36

⁷⁷Kr-Bolus Inhalation Method

Before our medical cyclotron became available in September, 1981, we produced 10–15 mCi ⁷⁷Kr by ⁷⁹Br (p,3n) reaction following bombardment of 35 MeV protons in the McGill synchrocyclotron. A highly concentrated ⁷⁷Kr gas was administered by a short inhalation technique, which gave the highest counting efficiency and minimum recirculation problems. A major advantage of the inert gas technique is that quantitative information can be obtained with a nontraumatic procedure. The arterial concentration of ⁷⁷Kr can be estimated from the radioactivity in end-tidal expired air (Veall and Mallett 1965), and the arterial pCO₂ level can be estimated from the CO₂ monitor of the expired air. Major drawbacks of the ⁷⁷Kr technique are its lower spatial resolution mode and its need for patient cooperation in bolus inhalation.

We used a Positome III device, consisting of two rings of 64 BGO crystals. It produces three slice cerebral hemodynamic studies simultaneously with a high counting efficiency of 84000 true coincidence counts/s in the outer slice and 130000 true coincidence counts/s in the center slice for a phantom 20 cm in diameter containing a concentration of 1 μCi/ml ⁶⁸Ga-EDTA (Thompson et al. 1979; Yamamoto et al. 1982).

Following bolus inhalation of ⁷⁷Kr, measurement of radioactivity in the brain was done at a 20-s sampling time over a 4- to 8-min period. A gamma-11 computer system with 64-K memory is used to acquire and reconstruct the concentration of ⁷⁷Kr in each pixel of a cross-section of the head by the modified convolution back projection method and attenuation correction (Thompson et al. 1979).

The size of each pixel is defined as 3.6 × 3.6 mm. The mean concentration in each pixel is calculated from the nine adjacent pixels, since the spatial resolution in Positome III in the dynamic mode is about 15 mm. The concentration of ⁷⁷Kr in the pixel at the time *t*, after the initial administration of ⁷⁷Kr, can be expressed in Kety's convolution integral equation derived from the Fick principle (Kety 1951). However, we cannot apply this equation directly to our study since our sampling time covers a period of 20–60 s. We have therefore adapted

the double integral method developed by Kanno and Lassen (1979). Use of this double integral method has the advantage of accommodating a long sampling time, giving an adequate counting efficiency of over 300000 counts/image.

The following double integral equation will be used to determine rCBF in each pixel:

$$(C_i)\dot{n} = 100 \cdot \frac{\int_{T(n-1)}^{T_n} Ca(t) \cdot e^{-K_i t} dt}{\int_0^{T_n} Ca(t) \cdot e^{-K_i t} dt}$$

The ratio $(C_i)'n$ of accumulated concentration of ⁷⁷Kr in the *i*th compartment from $T(n-1)$ to T_n , and accumulated concentration of ⁷⁷Kr in the *i*th compartment from 0 to the total examination time are determined over the head by PET. Also the ratio of accumulated radioactivity of ⁷⁷Kr in the arterial blood from $T(n-1)$ to T_n and accumulated radioactivity of ⁷⁷Kr in the arterial blood from 0 to the total examination time can be determined through continuous monitoring of radioactivity in the end-tidal expired air. The suitable K_i value is therefore established from the above data. Kanno and Lassen (1979) proposed a table look-up method to solve the equation for K_i , which is obtained as the weighted sum of N K_i values originally determined from each individual frame (N is the total number of integrated frames). Due to a certain ambiguity inherent in this method, we chose a different approach to obtain the proper K_i value. While still maintaining the table look-up idea, the best K_i was calculated by a least squares procedure (Yamamoto et al. 1982). The local brain-blood partition coefficient is also defined as the ratio of the accumulated radioactivity in the *i*th compartment from 0 to the total examination (A_i) time and accumulated radioactivity of the entire cross section of the brain (A), and multiplied by the mean brain-blood partition coefficient of krypton (Lassen and Munck 1955), which is 1.06 with normal hemoglobin. This is shown in the following equation:

$$\lambda_i = \frac{\int_0^\infty C_i(t)dt}{\int_0^\infty Ca(t)dt} = \lambda \frac{A_i}{A}$$

Following calculation of K_i and λ_i values, rCBV values of each pixel (F_i) can be determined.

Results

Normal Subjects

We studied eleven normal volunteers ranging in age from 21 to 36. The following rCBF values for various structures of the human brain were obtained.

Frontal gyrus	76 ± 9 ml/100 g/min
Superior mesial frontal	82 ± 11 ml/100 g/min
Supplemental motor area	70 ± 9 ml/100 g/min
Precentral gyrus	82 ± 8 ml/100 g/min
Parietal cortex	79 ± 10 ml/100 g/min
Caudate nucleus	69 ± 11 ml/100 g/min
Putamen	86 ± 13 ml/100 g/min
Globus pallidus	69 ± 11 ml/100 g/min

A recent study with normal volunteers indicated that a sequence of ballistic movements of the individual fingers of one hand induced a significant focally increased rCBF over 25% in the contralateral sensorimotor hand area, globus pallidus, and the bilateral supplementary motor areas (Roland et al. 1982).

Clinical Results

^{77}Kr PET studies using the bolus inhalation technique have been carried out in over 240 patients with cerebrovascular disorders. The degree and area of focal cerebral ischemia was established initially in all cases. Improvement of rCBF in the focal ischemic areas as well as topographical pattern changes of rCBF was then examined by repeated ^{77}Kr PET study following 5% CO_2 inhalation, which increased the arterial pCO_2 level to 4–6 torr through a vasodilatory effect on the cerebral microcirculation. Details of these clinical findings are reported elsewhere (Yamamoto et al. 1977, 1981a, 1981b, 1982).

Radiation Dose

Calculations for absorbed radiation dose resulting from ^{77}Kr PET study are based on equations and data provided by the MIRD Committee (1978).

Whole body	8 mrad/10 mCi
Lung	280 mrad/10 mCi
Brain	35 mrad/10 mCi
Liver	57 mrad/10 mCi
Kidneys	10 mrad/10 mCi
Ovaries	60 mrad/10 mCi

Recently introduced ^{13}N -labeled $^{13}\text{N}_2\text{O}$ and ^{18}F -labeled fluoromethane methods both have the advantage of a minimum radiation dose to patients and no need for arterial blood sampling (Holden et al. 1981; Madsen et al. 1981). For the recirculating diffusible tracer method, the H_2^{15}O intravenous injection technique was introduced by Raichle et al. (1981). This technique has the advantage of offering the shortest examination time, less than 60 s. However, it requires traumatic arterial blood samplings and administration of 100 mCi H_2^{15}O , which gives about ten times the radiation of the other dynamic methods.

Equilibrium Imaging

The great advantage of equilibrium imaging is that there is no specific limitation for sampling time; it is therefore possible to accumulate over 10 million coincidence counts/image, obtaining less than 10% statistical error with the highest resolution mode (Phelps et al. 1979). However, the technique has a poor temporal resolution of over 20 min. The technique of continuous inhalation of C^{15}O_2 was developed by the Hammersmith and Boston groups (Frackowiak et al. 1980; Jones et al. 1976; Subramanyam et al. 1978) and the continuous inhalation of ^{15}O -labeled nitrous oxide by Diksic et al. (1982). Using short-lived ^{15}O -labeled carbon dioxide, which transfers rapidly to H_2^{15}O in vivo, the C^{15}O_2 continuous inhalation technique has several practical advantages to the patient – simple procedure, high counting imaging, and minimum radiation dose of 1.8 mrad/mCi per minute for the whole body.

Radiation Dose

Calculations of the absorbed radiation dose resulting from C^{15}O_2 and N_2^{15}O PET studies are based on equations and data provided by the

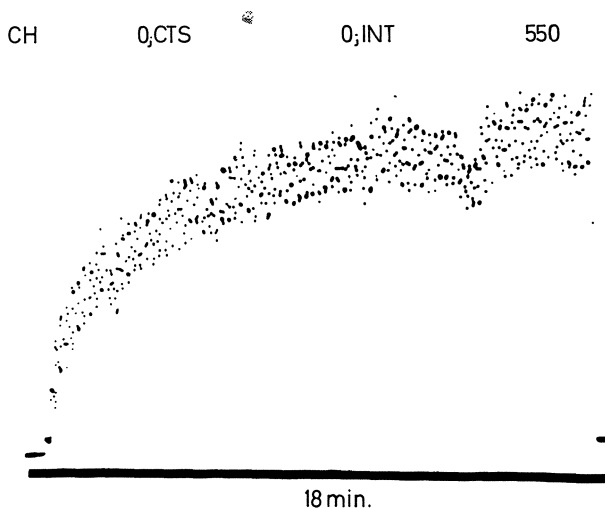


Fig. 1. Graphic display of continuous monitoring of the radioactivity in the femoral arteriovenous loop in dog during continuous and constant inhalation of $C^{15}O_2$, indicating over 13% fluctuation of radioactivity with respiratory cycle and sudden changes of unknown origin

MIRD committee. Administration dose and mode = 1 mCi/min with continuous inhalation mode for 20 min.

	$C^{15}O_2$	$N_2^{15}O$
Whole body	36 mrads/20 mCi	36 mrads/20 mCi
Gonads	138 mrads/20 mCi	54 mrads/20 mCi
Kidney	144 mrads/20 mCi	56 mrads/20 mCi
Liver	127 mrads/20 mCi	53 mrads/20 mCi
Brain	94 mrads/20 mCi	25 mrads/20 mCi
Lung	300 mrads/20 mCi	273 mrads/20 mCi

Although simple and practical in its methodology, equilibrium imaging has several practical shortcomings and limitations, some of which have already been discussed by several authors (Frackowiak et al. 1980; Huang et al. 1979; Lammertsma et al. 1981). These include inherent nonlinearity, variation in the brain-blood partition coefficient (Ter-Pogossian et al. 1969), cross calibration between the PET study and the gamma counter, partial volume effect, and counting statistics of radioactivity in each pixel of the brain. A major requirement for the technique is the constancy of arterial ^{15}O -labeled water activity (Ca) over the course of the scanning period in the steady state. However, our observation of Ca in the steady state following continuous inhalation of $C^{15}O_2$ in humans, and by multiarterial samplings during the scanning period and continuous measurement of the arterial blood radioactivity of a femoral arteriovenous loop in dogs, indicates that Ca varies considerably during the scanning process due

to variation of the respiratory mode and complexity of water distribution and extraction patterns in the in vivo state, as shown in Fig. 1. In the rCBF value range of 50 ml/100 g per minute Menon et al. (1982) have shown that a 10% variation in the arterial blood activity results in over 30% error in rCBF value.

These findings indicate that a reliable rCBF value is difficult to obtain by measurement of activity in randomly sampled arterial blood. We have therefore proposed a new method for quantitation of rCBF following continuous inhalation of $C^{15}O_2$ or $N_2^{15}O$ in order to correct the time variable input function (*I*) (Meyer et al. 1982).

Dynamic Plus Equilibrium Imaging

Our new proposal is that the time variable input function (*I*) be calculated from the dynamic mode operation from the beginning of $C^{15}O_2$ or $N_2^{15}O$ inhalation to the equilibrium state. Each pixel rCBF is then determined from the data obtained during equilibrium imaging with an adequate counting rate of over 6 million coincidence counts per slice in the high-resolution mode. In order to measure the time variable arterial radioactivity (*A*), the continuous monitoring of *A* is required. We have used the continuous monitoring of end-tidal expired air and lung radioactivity for the ^{77}Kr bolus inhalation technique (Yamamoto et al. 1982).

In dogs we correlated the radioactivity through continuous monitoring of the arterial blood and both end-tidal expired air and the mid portion of the right lung during continuous inhalation of $C^{15}O_2$ and $N_2^{15}O$ after an external arteriovenous loop between the femoral artery and femoral vein had been made. These animal experiments indicate excellent correlation between the arteriovenous loop radioactivity curves and the lung or end-tidal expired air radioactivity curve during continuous $C^{15}O_2$ and $N_2^{15}O$ inhalation. Therefore, the continuous monitoring of either lung or end-tidal expired air radioactivity might be considered as a non-invasive means for continuous measurement of A during $C^{15}O_2$ and $N_2^{15}O$ inhalation. During the build-up phase from the beginning of $C^{15}O_2$ and $N_2^{15}O$ inhalation to the equilibrium state, the radioactivity in the brain is measured every 40 s sampling time for 10–12 min by Positome III. The camera gives three slices of information simultaneously with parallel continuous monitoring of both end-tidal expired air and lung radioactivities for every 0.5 s of sampling time. Using the modified double integral meth-

od described previously in ^{77}Kr PET methodology, mean rCBF (F) in the selected cortical area is calculated from the data obtained during the dynamic study.

Following the dynamic scan, the equilibrium high-resolution scan is performed to accumulate 6–10 million coincidence counts per slice. Five to 8 min of scanning time are required to minimize the statistical error of radioactivity in the pixel.

Following determination of average tissue concentration in the selected cortical area (B) in the equilibrium imaging, the time variable input function I is calculated from F determined from dynamic imaging, and B determined from the equilibrium imaging using the following equation developed by Jones et al. (1976), and Subramanyam et al. (1978).

$$\bar{F} = \frac{\lambda}{I/\bar{B}-1} \rightarrow I = \bar{B} \left(1 + \frac{\lambda}{\bar{F}} \right)$$

Using the calculated input function I , each pixel rCBF (F_i) is calculated from the following equation:

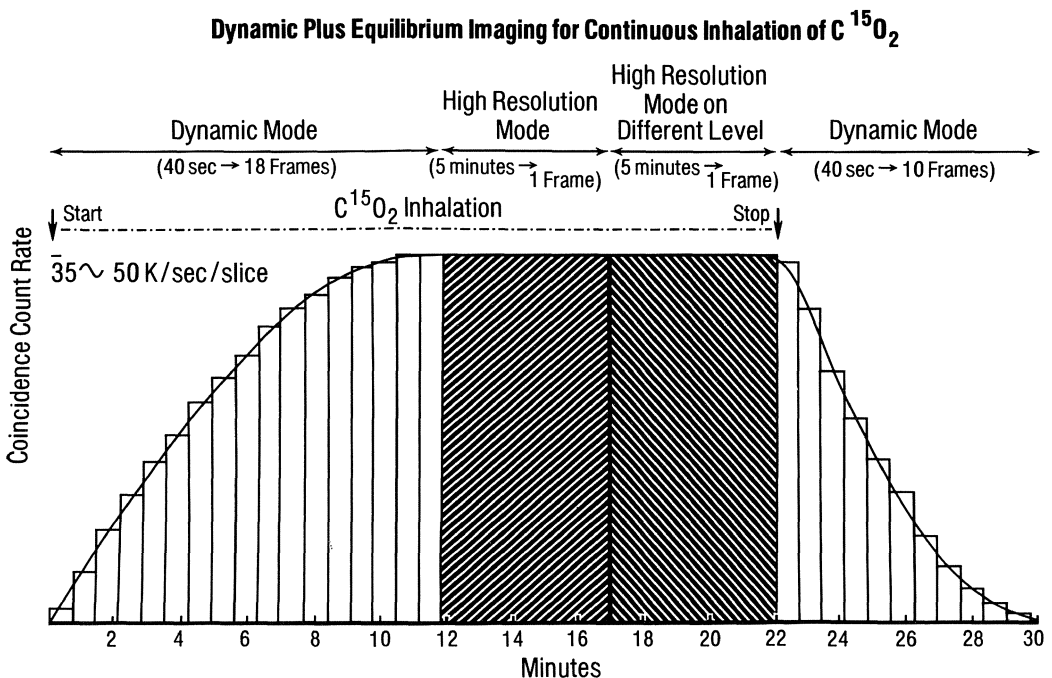


Fig. 2. Diagrammatic display of scanning mode of "dynamic plus equilibrium imaging" for six tomographic mapping of rCBF with Positome III

$$F_I = \frac{\lambda}{I/B_I - 1}$$

This technique of dynamic plus equilibrium imaging combined with continuous monitoring of lung radioactivity corrects the time variable input function to give more reliable information for quantitation of rCBF by PET through noninvasive means. Since Positome III can give three slices of information simultaneously, the combined build-up and the initial equilibrium images are given rCBF values in all three slices. The second equilibrium imaging at the various levels, combined with dynamic study in the clearance phase, provides additional rCBF measurements in three other levels as shown in Fig. 2. The combined build-up phase plus equilibrium imaging and the second equilibrium plus clearance phase at the different levels by PET with parallel continuous monitoring of either lung or end-tidal expired air radioactivity provides six tomographic mappings of rCBF. Furthermore, this additional information is collected with a high-resolution mode and the same radiation dose of 36 mrad for the whole body as the equilibrium imaging method.

Discussion

We have reviewed some practical aspects of currently available PET methodology for quantitative measurement of rCBF. The dynamic imaging methods discussed here have the advantage of a short scanning time with minimum radiation dose and noninvasive techniques. (The exception is the H₂O intravenous injection method, which requires ten times the normal radiation dose, and multiple arterial blood samplings). The major disadvantage of all types of dynamic imaging is the lower spatial resolution mode, which is necessary to minimize statistical error.

The advantages of equilibrium imaging are its excellent imaging quality, its high-resolution mode, and its reasonable counting rate per image. However, arterial radioactivity during scanning is found to fluctuate considerably (over 10%) because of the change of respiratory and possibly complex distribution patterns of radioactive water in the in vivo state.

We have therefore recently proposed the dynamic plus equilibrium imaging method. It overcomes these problems and provides noninvasive continuous monitoring of lung or end-tidal expired air radioactivity rather than arterial samplings.

Furthermore, the recently introduced ¹⁵O-labeled N₂¹⁵O has several advantages. It is a simple physiological model employing a metabolically inert tracer and, unlike the C¹⁵O₂ method, is not affected by the brain-blood partition coefficient difference between gray and white matter (Kety et al. 1948).

References

1. Diksic M, Yamamoto YL, Feindel W (1982) Recent development of tracers for BCNU pharmacokinetics and regional cerebral blood flow measurements. In: Filipow L, Menon D (eds) Maria Workshop V: Positron Emission Tomography, 20–23 April, 1982. University of Alberta Press, Edmonton
2. Frackowiak RS, Lenzi GL, Jones T, Heather JD (1980) Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man using ¹⁵O and positron emission tomography: theory, procedure and normal values. *J Comp Ass Tomogr* 4: 727–736
3. Holden JE, Gatley SJ, Hichwa RD et al. (1981) Cerebral blood flow using PET measurements of fluoromethane kinetics. *J Nucl Med* 22: 1084–1088
4. Huang SC, Phelps ME, Hoffman EJ (1979) A theoretical study of quantitative flow measurements with constant infusion of short-lived isotopes. *Phys. Med Biol* 24: 1151–1161
5. Jones T, Chesler DA, Ter-Pogossian MM (1976) The continuous inhalation of oxygen-15 for assessing regional extraction in the brain of man. *Br J Radiol* 49: 339–343
6. Kanno I, Lassen NA (1979) Two methods for calculating regional cerebral blood flow from emission computed tomography of inert gas concentration. *J Comput Assist Tomogr* 3: 71–76
7. Kety SS (1951) The theory and applications of the exchange of inert gas at the lungs and tissue. *Pharmacol Rev* 3: 1–41
8. Kety SS, Harmell MH, Broomell HJ, Rhode CB (1948) The solubility of nitrous oxide in blood and brain. *J Biol Chem* 173: 487–496
9. Lammertsma AH, Jones T, Frackowiak RS, Lenzi GL (1981) A theoretical study of the steady-state model for measuring regional cerebral blood flow and oxygen utilisation using oxygen-15. *J Comput Assist Tomogr* 5: 544–550
10. Lassen NA, Munck O (1955) The cerebral blood

- flow in man determined by the use of radioactive krypton. *Acta Physiol Scand* 33: 30
11. Madsen MT, Hichwa RD, Nickles RJ (1981) An investigation of ^{11}C -methane, ^{13}N -nitrous oxide and ^{11}C -acetylene as regional cerebral blood flow agents. *Phys Med Biol* 26: 875–882
 12. Menon D, Meyer E, Yamamoto YL (1982) Limitations of the steady-state oxygen-15 technique for quantitation of regional cerebral blood flow using positron emission tomography. In: Filipow L, Menon D (eds) *Maria Workshop V: Positron Emission Tomography*, 20–23 April 1982. University of Alberta Press, Edmonton
 13. Meyer E, Yamamoto YL, Menon D (1982) A dynamic plus equilibrium PET imaging technique for quantitation of rCBF using inhalation of radioactive gases. In: Filipow L, Menon D (eds) *Maria Workshop V: Positron Emission Tomography*, 20–23 April 1982. University of Alberta Press, Edmonton
 14. MIRD (1978) Pamphlet No. 5, Society of Nuclear Medicine, New York
 15. Phelps ME, Hoffman EJ, Huang SC, Kuhl DE (1979) Design considerations in positron computed tomography (PCT). *IEEE: Trans Nucl Sci* NS-26: 2746–2751
 16. Raichle ME, Markham J, Larson K et al. (1981) Measurement of local cerebral blood flow in man with positron emission tomography. *J Cerebr Blood Flow Metabol* 1 [suppl 1]: 19
 17. Roland PE, Meyer E, Shibasaki T et al. (1982) Regional cerebral blood flow changes in cortex and basal ganglia during voluntary movements in normal human volunteers. *J Neurophysiol* 48: 467–480
 18. Subramanyam R, Alpert NM, Hoop B et al. (1978) A model for regional cerebral oxygen distribution during continuous inhalation of $^{15}\text{O}_2$, C^{15}O , and C^{15}O_2 . *J Nucl Med* 19: 48–53
 19. Ter-Pogossian MM, Eichling JC, Davis DO et al. (1969) The determination of regional cerebral blood flow by means of water labeled with radioactive oxygen-15. *Radiology* 93: 31–40
 20. Thompson CJ, Yamamoto YL, Meyer E (1979) Positome II: a high efficiency positron imaging device for dynamic brain studies. *IEEE Trans Nucl Sci* NS-26: 583–589
 21. Veall N, Mallett BC (1965) Two-compartment model using ^{133}Xe inhalation and external recording. *Clin Sci* 29: 179
 22. Yamamoto YL, Thompson CJ, Meyer E et al. (1977) Dynamic positron emission tomography for study of cerebral hemodynamics in a cross-section of the head using positron emitting ^{18}Ga -EDTA and ^{77}Kr . *J Comput Assist Tomogr* 1: 43–55
 23. Yamamoto YL, Matsunaga M, Meyer E et al. (1981a) Evaluation and management of cerebral ischemia by positron emission tomography. In: Barnett H, Paoletti P, Flamm E, Brambilla G (eds) *Cerebrovascular disease: new trends in surgical and medical aspects*. Elsevier/North-Holland Biomed, Amsterdam, pp 123–134
 24. Yamamoto YL, Thompson C, Meyer E, Feindel W (1981b) Positron emission tomography for measurement of regional cerebral blood flow. *Adv Neurology* 30: 41–53
 25. Yamamoto YL, Meyer E, Thompson CJ, Feindel W (1982) ^{77}Kr clearance technique for measurement of regional cerebral blood flow by positron emission tomography. In: Kuhl DE (ed) *Positron and single photon emission tomography*. G.T. Management, New York

The Measurement of Regional Cerebral Blood Flow in Man Using an in Vivo Autoradiographic Technique and Positron Emission Tomography

By A. H. Lockwood, M. D. Ginsberg, B. E. Howard, P. Freed, and W. Hassel

Introduction

Since the rate at which blood is delivered to discrete regions of the brain is an essential component of a variety of metabolic, physiological, and pharmacological processes, it is highly desirable to be able to measure regional cerebral blood flow (rCBF) in the brains of living human subjects with a minimal degree of invasiveness. The development of positron emission tomography (PET), and the construction of sensitive instruments with good resolution that are specifically optimized for studies of cerebral function has made this goal attainable. In previous reports from this laboratory, we have described a modified in vivo autoradiographic strategy for measuring rCBF in man. The method is based on the familiar in vitro autoradiographic strategy developed from the Fick principle (Kety 1951; Freygang and Sokoloff 1959; Sakurada et al. 1978). Preliminary validation studies, conducted in animals, showed excellent agreement between the in vivo and in vitro autoradiographic methods. This report reviews the theoretical basis of the method, and describes some practical and theoretical considerations necessary for its successful application. Preliminary results in normal humans are presented.

Theoretical Considerations

The following symbols are used: F , cerebral blood flow ($\text{ml} \cdot \text{min}^{-1}$); A , concentration of tracer in artery ($\mu\text{Ci} \cdot \text{ml}^{-1}$); C_V , concentration of tracer in cerebral vein ($\mu\text{Ci} \cdot \text{ml}^{-1}$); B , concentration of tracer in brain ($\mu\text{Ci} \cdot \text{g}^{-1}$); W , weight of brain tissue in pixel (g); Q_B , amount of tracer in pixel (μCi); λ , brain: blood partition coefficient for tracer ($\text{ml} \cdot \text{g}^{-1}$); α , decay constant for tracer (min^{-1}); f , cerebral blood

flow per unit weight of tissue (F/W); k , $f/\lambda + \alpha$.

From the Fick principle, modified to account for physical decay of a tracer during the course of an investigation:

$$dQ_B/dt = F(A - C_V) - \alpha Q_B \quad (1)$$

Assuming instantaneous equilibration of the tracer between brain and blood (i.e., $C_V = B/\lambda$), $Q_B = B/W$, and substituting into Eq. (1) and integrating from the beginning to the end of a study (time 0 to t) yields:

$$B(f, t) = f \int_0^t A(t-\tau) e^{-k\tau} d\tau, B(f, 0) = 0 \quad (2)$$

This form of the equation is similar to that developed for measuring rCBF from autoradiographs obtained during the study of experimental animals (Freygang and Sokoloff 1959; Reivich et al. 1969, Sakurada et al. 1978). The equation states that the instantaneously measured concentration of the tracer in a homogeneous region of the brain at time T , is a function of f , and the specific activity curve for the tracer in arterial blood during the time of the study. Since even the best of modern PET scanners cannot record enough events in a short enough period to produce an "instantaneous" measurement of brain tracer content, the method cannot be employed without further modification. In practice, PET scan data are integrated measurements of brain activity during the period that the scanning equipment is collecting data. Restated:

$$C = \int_0^T B(f, t) dt \quad (3)$$

The cumulative content of tracer within a homogeneous region C , in the time interval 0 to T , can be expressed as a function of flow, f , the ar-

terial specific activity curve, as before [Eq. (2)], if the equation is integrated a second time over the interval 0 to T :

$$C(f, T) = f \int_0^T \int_0^t A(t-\tau) e^{-k\tau} d\tau dt \quad (4)$$

This equation is the operational equation that forms the basis for computing rCBF from an intravenously injected tracer and rapid PET scanning. When the method is implemented, a strategy for administering the tracer and scanning the subject needs to be developed that maximizes the computed flow difference for a given measured difference in tracer content between discrete brain regions, and, at the same time, avoids potential situations where multiple values for blood flow can be computed from a single isotope content measurement in a given brain region.

In initial validation studies, done in rats, Ginsberg et al. (1982) found excellent agreement between the classical in vitro autoradiographic technique, and the modified in vivo autoradiographic technique when ^{15}O -labeled water was employed as the tracer, and the partial pressure of carbon dioxide in arterial blood was varied over a wide range (Ginsberg et al. 1982). In these studies, a ramp-shaped tracer infusion schedule was chosen that permitted the arterial specific activity curve to be expressed as a low-order polynomial. This permitted computational simplifications to be made that obviated the need to employ a convolution technique to calculate flow (for details see Ginsberg et al. 1982). Although the method was attractive in its simplicity, when ramp infusions were used in human studies, too much of the isotope decayed in the syringe and in the subject before it reached the brain. This caused the PET images to be of inadequate quality, because insufficient amounts of the tracer were delivered to the brain. This prompted a reevaluation of the experimental protocol.

To avoid multiple solutions for the operational equation, given a single value of cumulative isotope content in a homogeneous brain region f , expressed as a function of C , must always increase, or, restated, the partial derivative of C with respect to flow, f , must always be positive:

$$\frac{\delta C}{\delta f} = \int_0^T \int_0^t \left(1 - \frac{f\tau}{\lambda}\right) A(t-\tau) e^{-k\tau} d\tau dt \quad (5)$$

This expression can be evaluated explicitly by integrating by parts. This produces the following equation:

$$\frac{\delta C}{\delta f} = \left(\frac{1}{f} - \frac{T}{\lambda}\right) C + \frac{2}{\lambda} \int_0^T C(f, t) dt \quad (6)$$

Since the integrated value of C is always positive, the value of the entire expression will be positive if $(1/f - T/\lambda)$ is also positive. Restated, if T is less than λ/f , unique solutions are assured. This analysis indicates that there may be an upper time limit on the duration of a PET scan, which, if exceeded, could result in multiple solutions for the operational equation. The clear indication is that to minimize this possibility, the scan should be of short duration. A further analysis of the operational equation forms the basis for a number of uniqueness theorems that are being developed.

This method is invasive to the degree that arterial blood sampling is required. This is conveniently and safely done by placing a 20-gauge catheter in the radial artery at the wrist. In the operational equation, all time values are assumed to be in the same frame of reference. However, since the length of time required for the bolus of activity to travel from the heart to the brain is less than the time required for the bolus to travel from the heart to the wrist and into a sampling device, this difference in time must be known so that all of the data can be placed in the same time frame. Thus, the scanner must be capable of performing short-duration scans so that the arrival of the bolus in the brain can be measured and correlated with the arrival of the bolus at the arterial sampling site. Figure 1 shows the effect of a 0.06-min time offset on the configuration of the C versus blood flow plot. The plot indicates that there is about a 5% error in the flow calculation due to this amount of uncertainty, and points out the need for accurate time measurements. The measurement is best made when both the brain and arterial activity curves rise sharply. Again, the ramp infusion appears to be unsatisfactory. Furthermore, and perhaps more importantly, the requirement for timing the arrival of the

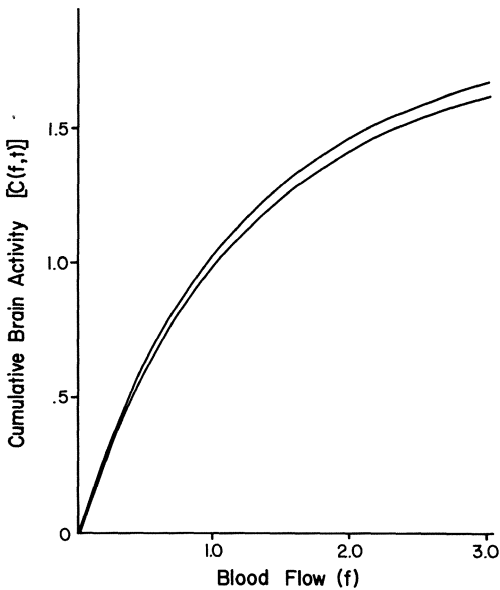


Fig. 1. The relationship between values of blood flow, f , obtained from the operational equation [Eq. (4)], is shown as a function of measured values for the cumulative tracer content in a brain region $C(f, T)$. The data used were from an actual patient study that employed an intravenous bolus injection of ^{15}O -water. The difference between the curves represents the effect of a 0.06-min time shift in the time base that relates blood and brain activity. This produces an error of about 5% in the physiological range for blood flow

tracer in the brain points out the need to be able to modify PET scan data collection routines to fit individual applications.

Although the theoretical discussion assumes that the tracer to be used is not diffusion limited, in practice this is rarely, if ever, the case. A derivation of the operational equation that is more correct would involve redefining k , so that $k = mf/\lambda + \alpha$, where m is a positive, monotonically decreasing function of f , with $m(0) = 1$, that describes the degree to which the tracer is completely equilibrated with the brain during a single transit of blood through the cerebral capillary bed. In the *in vitro* autoradiographic method, the effect of diffusion limitation on calculated values for blood flow can be reduced by using a short study time, as discussed by Eklöf et al. (1974). A similar argument can be made for the present method. In the operational equation, number 4, m appears in two places, in f , and in k . The effect of f is minimized if $(1-m)$ is very small and if

$(1-m)fT/\lambda$, in the exponent, is also very small. Since the time of the study, T , is the only one of these variables that can be controlled, the effect of diffusion limitation on calculated values of C , and hence f , can be made smaller by employing shorter scan times. With a scanning strategy that allows the length of the total time of data acquisition to be broken down into small increments, it is theoretically possible to calculate the value of m for any given homogeneous region of the brain.

These practical considerations lead to the conclusion that the tracer must be administered rapidly. This causes the amount of tracer in the brain to be at high levels, since it arrives before substantial radioactive decay occurs. This, in turn, leads to high brain tracer concentrations and images that contain a sufficient number of events to be of good statistical quality, while avoiding long scan times that could lead to ambiguous solutions of the operational equation. Rapid injections of the tracer also facilitate accuracy in placing the arterial and brain data in the same time frame. We have concluded that an intravenous bolus injection is preferred.

Preliminary Data

To date, we have conducted two studies in one individual, using the ramp infusion technique. As was stated, the resulting PET images were statistically unsatisfactory, even though the syringe contained 35 mCi of ^{15}O -water at the beginning of the infusion. Calculable differences in flow did, however, show the expected effect of hypocapnia on rCBF.

Four additional subjects have undergone a total of five studies, using the bolus injection technique. Two tomographic slices illustrating the transformation of integrated brain activity into functional images showing rCBF are shown in Fig. 2. The flow versus cumulative tracer in the brain relationship is shown in Fig. 1. In subsequent studies we have performed 1-s scans every other second for 30 scans, in order to time the arrival of the bolus of isotope in the brain, followed by a single 90-s scan that was used as the basis for the solution of the operational equation. Thus far, this strategy has allowed us to distinguish the dif-

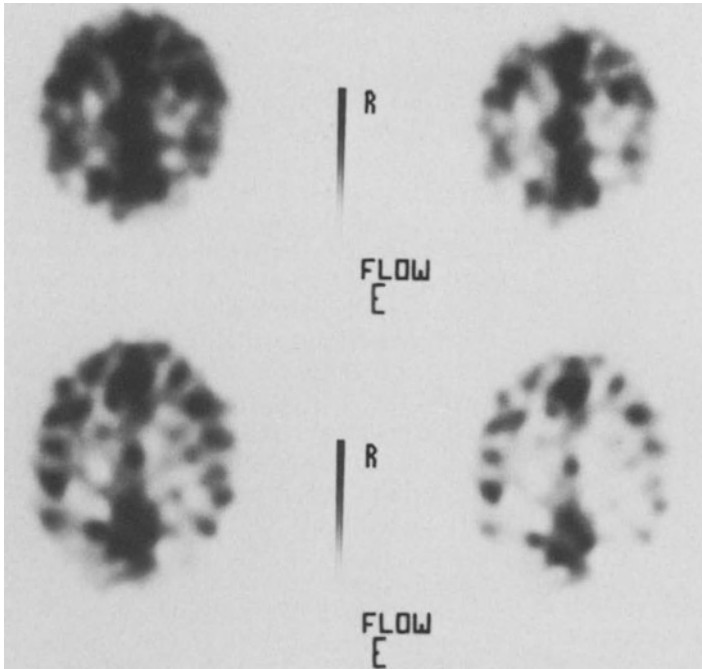


Fig. 2. The images on the *left side* of the figure are the raw data from a 1.5-min PET scan of the brain after an i.v. bolus of ^{15}O -water. The bolus required about 0.3 min to reach the brain, thus limiting total useful imaging time to about 1.2 min. The images on the right have been transformed from isotope content to flow images, using a "look-up" table containing the data shown in Fig. 1

ference between flow values in the physiological range (see Fig. 1), and, at the same time, to obtain data of adequate statistical quality.

Discussion

The modified, *in vivo* autoradiographic method for measuring rCBF in man is a relatively simple technique that is best applied in centers where multislice PET imaging systems are in use or planned. The entire procedure takes only a few minutes once the appropriate catheters are inserted. Since the radionuclide is injected as a bolus, rather than inhaled or infused until an equilibrium distribution is achieved, the PET camera can be placed at some distance from the cyclotron. Furthermore, the need for precise control of the cyclotron beam and isotope delivery system is eliminated, since the method is independent of the requirement for achieving a true steady-state isotope distribution. Although we have chosen to use ^{15}O -water as a tracer, any suitable tracer can be used. Similarly, although we have chosen to use an intravenous bolus injection technique to administer

the tracer, it would be possible to administer the tracer by inhalation. Institutions that do not have multislice imaging capabilities will not find this a satisfactory method, since the injection and blood sampling would have to be repeated for each tomographic slice that is desired.

Our choice of ^{15}O -water as a blood flow tracer is based on several considerations. This laboratory has reported the successful synthesis of ^{11}C -iodoantipyrine, and the validation of its use as a CBF tracer in experimental animals (Campbell et al. 1981; Ginsberg et al. 1981). Because of the low yield, the complexity of the synthesis, and the need to perform preinjection quality control on the radiopharmaceutical, we sought an alternate tracer. Water has many attractive features. It is metabolically inert, and easily synthesized in large amounts with a high yield by passing ^{15}O -oxygen and hydrogen gas over a platinum catalyst. Although the movement of water across the blood-brain barrier is limited by diffusion, and thus it does not totally conform to this requirement for a flow tracer, diffusion limitation only becomes problematic in regions where blood flow is high (Raichle et al. 1974). Since the ischemia is a major interest

in this laboratory, we are interested in measuring low flow values. Water is well suited to this application. In regions of the brain where blood flow is high, it is possible to apply a correction to the calculated flow value using m values from the literature, or, possibly by calculating m values from the data. Thus, we believe that when an injection strategy is used that is designed to make the study as short as possible, for the theoretical reasons already discussed, water is a tracer that is well suited to most needs.

The preliminary studies that we have conducted have convinced us that this method is very workable in a research laboratory environment. Typical studies require one-half a day and several technicians. The requirement for technical assistance can be reduced by using an automated blood sampling and counting device. Large amounts of time and effort are expended during the collection and measurement of arterial blood samples, since great attention must be paid to the timing of sample collection and counting, as well as weighing or measuring the volume of individual samples. Since an arterial catheter is required, the usual problems associated with measuring arterial blood gas values are circumvented. We believe that the method is advantageous, and can be used in most PET centers.

Acknowledgments. We thank our many colleagues that helped make this project possible. This work was supported in part by Positron-Emission Tomography Center Grant NS 15639, and USPHS grant NS 14996.

References

1. Campbell JA, Finn RD, Boothe TE et al. (1981) Synthesis of ^{11}C -iodoantipyrine for positron-emission tomography. *J Nucl Med* 22: 538–541
2. Eklöf B, Lassen NA, Nilsson L et al. (1974) Regional cerebral blood flow in the rat measured by the tissue sampling technique; a critical evaluation using four indicators. ^{14}C -antipyrine, ^{14}C -ethanol, ^3H -water and ^{133}Xe on. *Acta Physiol Scand* 91: 1–10
3. Freygang WH Jr, Sokoloff L (1959) Quantitative measurement of regional circulation in the nervous system by the use of inert radioactive gas. *Adv Biol Med Phys* 6: 263–279
4. Ginsberg MD, Lockwood AH, Busto R et al. (1981) ^{11}C -iodoantipyrine for the measurement of regional cerebral blood flow by positron emission tomography. *Stroke* 12: 745–750
5. Ginsberg MD, Lockwood AH, Busto R et al. (1982) A simplified in vivo autoradiographic strategy for the determination of regional cerebral blood flow by positron emission tomography: theoretical considerations and validation studies in the rat. *J Cereb Blood Flow Metabol* 2: 89–98
6. Kety SS (1951) The theory and applications of the exchange of inert gas at the lung and tissues. *Pharmacol Rev* 3: 1–41
7. Raichle ME, Eichling JO, Grubb RL Jr (1974) Brain permeability of water. *Arch Neurol* 30: 319–321
8. Reivich M, Jehle J, Sokoloff L et al. (1969) Measurement of regional cerebral blood flow with antipyrine- ^{14}C in awake cats. *J Appl Physiol* 27: 296–300
9. Sakurada O, Kennedy C, Jehle J et al. (1978) Measurement of local cerebral blood flow with iodo (^{14}C) antipyrine. *Am J Physiol* 234: H59–H66

Regional Cerebral Blood Flow Measurement with Fluoromethane and Positron Tomography

By J. E. Holden, S. J. Gatley, R. J. Nickles, R. A. Koeppe, G. G. Celesia, and R. E. Polcyn

Tomographic methods of CBF determination offer the obvious advantages of anatomical specificity and access to deep-lying cerebral structures. In addition, the uniform, quantitative scaling of local indicator concentration measurements allows approaches to data analysis not appropriate for probe-based measurements, in which each detector has a different, unspecified sensitivity to the radioactivity in its field of view. However, the relatively low sensitivity of tomographic systems requires peak cerebral tissue radioactivity concentrations on the order of several $\mu\text{Ci}/\text{ml}$. This has been the motivation for our continuing search for inert, gaseous indicators with blood Ostwald solubility coefficients that are high enough to provide the required cerebral tissue radioactivity levels, and yet low enough to preserve good discrimination between neighboring flow values. Fluoromethane (methyl fluoride, CH_3F) labeled with ^{18}F has proven to be an excellent prospect. Its Ostwald solubility coefficient was measured to be 1.07 in whole blood at body temperature (Gatley et al. 1981). Its chemical inertness in vi-

vo has been demonstrated repeatedly. Figure 1 shows its total extraction in brain tissue. The figure compares residue curves from bolus injections of similar quantities of fluoromethane and 3-fluoro-3-deoxy-D-glucose (3FDG) and in an isolated dog brain preparation (Gilboe et al 1973). The incomplete extraction of the 3FDG (41% in this case) is clearly evident. No trace of an unextracted fraction of fluoromethane was evident either in the residue curves or in the venous outflow for average whole-brain blood flows ranging up to 70 ml/min per 100 g.

Further validation of fluoromethane as a flow indicator was carried out in human subjects using a high-sensitivity coincidence counting system. Response to elevated PaCO_2 was measured by two compartment analysis of brain residue curves acquired following venous injection of small quantities of fluoromethane. The observed elevation of gray matter flow corresponded well with that expected (Lennox et al. 1975).

In a previous report of our basic approach to CBF measurements using fluoromethane (Holden et al. 1981), we introduced the possibility of deriving both the local blood flow and tissue/blood partition coefficient simultaneously from the same data set, even if neither saturation nor desaturation are measured to completion. In this paper we report our further investigation of this question. In addition, we discuss the effects of the large Ostwald solubility on the arterial fluoromethane concentration, and the effects of the more slowly clearing arterial concentration on the CBF determinations. Finally, a number of actual clinical results serve as examples of applications of the fluoromethane method being investigated in our laboratory.

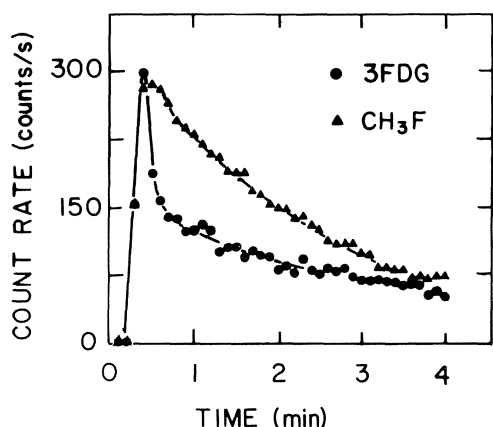


Fig. 1. Brain tissue residue curves from bolus injections of similar quantities of fluoromethane and 3-fluoro-3-deoxyglucose in an isolated dog brain model. The curves demonstrate the 100% extraction of fluoromethane

Methods

Fluoromethane was synthesized from ^{18}F -tetraethylammonium fluoride dihydrate by nucleophilic displacement of iodide from methyl iodide (Gatley et al. 1981). The CBF studies were carried out using the ORTEC ECAT-II scanner (Phelps et al. 1978) at the Middleton VA Hospital. The subject was fitted with a respirator mask with one-way valves for the control of expired air, and positioned appropriately. The subject inhaled 10–50 mCi fluoromethane in a single breath and held his breath for 30 s. At the instant of inhalation, a sequence of 1-min scans was initiated and continued for at least 8 min. Throughout the procedure, count rate over the right lung, and expired air radioactivity and CO_2 concentration were measured at a frequency of at least 60 channels/min. Typical prethoracic and expired air curves are shown in Fig. 2. The smooth expired air curve represents an interpolation between the end-tidal peaks in the actual data. The time course of the arterial indicator concentration $C_a(t)$ is assumed to be proportional to the expired air curve during washout. The relative vertical scales of the thoracic and expired air count rates are estimated in a simple compartmental model, which also provides an estimation of the unobserved arterial time course during breath-hold.

The analysis of the scan data is based on the assumptions of linearity and indicator conservation:

$$C_i(t) = \int_0^t C_a(t') G_i(t-t') dt' \quad (1)$$

where $C_i(t)$ is the instantaneous indicator concentration in the i^{th} pixel, and $G_i(t)$ is the response of the pixel tissues to a normalized impulse input of arterial concentration. It is this function that contains the desired physiological information; it can be shown that its zero intercept and area are equal to the blood flow per unit volume and the total indicator distribution volume per unit volume in the pixel, respectively. The measured pixel values are the instantaneous concentrations integrated over the scan duration:

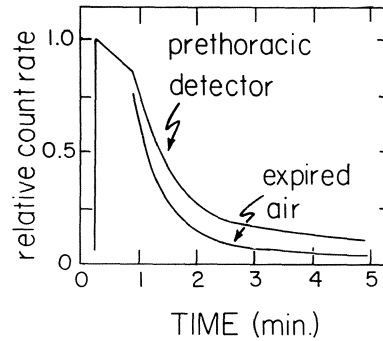


Fig. 2. Prethoracic and expired air curve rates measured during a 45-s breath-hold administration. The expired air curve is the smooth interpolation between the end-tidal peaks in the original data. The relative vertical scales are estimated in a simple compartmental model which also provides the shape of the arterial input functions used in fitting the scan data

$$C_i(I) = \int_{(I-1)T}^{IT} C_i(t) dt \quad (2)$$

where I is the scan number in the dynamic sequence and T is the scan duration. In our method, a single exponential function is used as an estimator for $G_i(t)$:

$$G_i(t) = A f_i \exp(-f_i t / \lambda_i) = A \lambda_i k_i \exp(-k_i t) \quad (3)$$

where A is the ratio of absolute radioactivity calibrations of the pixel value and arterial concentration measurements, f_i is the blood flow per unit volume in the pixel, λ_i is the total distribution volume per unit volume in the pixel (essentially equal to the Kety tissue/blood partition coefficient), and k_i is f_i / λ_i , the flow per unit distribution volume in the pixel. Trial $C_i(I)$ values calculated using equations 1, 2, and 3 are compared with the actual scan pixel values; the parameters $A \lambda_i$ and k_i are adjusted in a nonlinear least squares optimization scheme until the mean square discrepancy between model and data is a minimum. Typically, the first eight scan values are fitted simultaneously. Despite our use of single exponential impulse response function, our approach to the data is stochastic, rather than compartmental, in nature. Our goal is to estimate most accurately the average flow and distribution volume in each pixel; isolation of tissue types or of anatomical features must thus be provided to-

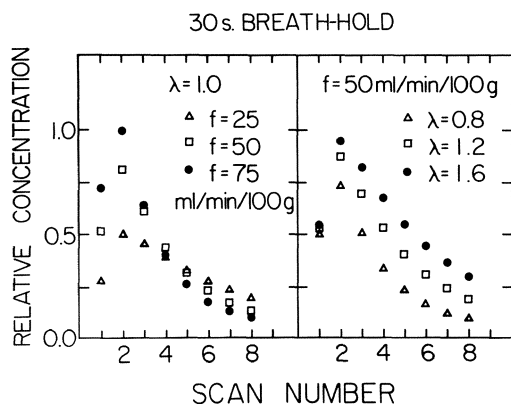


Fig. 3. Predicted scan pixel values $C_i(I)$ (arbitrary units), for a 30-s breath-hold period and 1-min scan durations, showing the f dependence at constant λ and the λ dependence at constant f . The pixel values in the first minutes are nearly proportional to f and independent of λ . At 4.5 min, the values are nearly proportional to λ and independent of f .

mographically rather than in the fitting procedure.

The process of determining the optimal $A\lambda_i$ and k_i values is repeated for each pixel in the image sequence. Image maps of $A\lambda_i$, k_i and their product Af_i are stored in a display file for subsequent viewing and region of interest analysis. To date, we have not used direct arterial sampling for the estimation of A and subsequent conversion of the relative distribution volume and flow maps to absolute values. Our usual procedure is to set the average λ value in a large region of interest, typically at least an entire hemisphere exclusive of ventricular regions, to 1.0. This procedure produces rescaled maps of λ_i with normal pixel values ranging from 0.9 to 1.1, and of f_i with a pixel value range of 20–80 ml/min per 100 g. The flow value range is extended to more than 100 ml/min per 100 g with the inhalation of 5% CO_2 . The λ_i maps are essentially featureless, consistent with the near identity of the aqueous and lipid solubilities of fluoromethane (Gatley et al. 1981).

Because of this method of absolute scaling, flow values are compared quantitatively only for multiple studies of the same slice. This has proven adequate for observing changes in flow values induced by clinical or experimental interventions. Estimation of A from the absolute airway radioactivity concentration, measured using a dry spirometer, is being investigated, as

a compromise between the obvious need for an absolute flow scale for intersubject comparison, and our desire to preserve the noninvasive nature of the method.

Figure 3 shows values of $C_i(I)$ calculated using the fitting model. The figure demonstrates that the effects of variations of f and of λ on the pixel values are readily distinguishable from each other. In addition, the advantage of an agent with nearly uniform solubility throughout the brain is demonstrated. The response of a pixel with a flow of 50 ml/min per 100 g is seen to be very nearly equal to that of a pixel containing equal masses of tissue types with flows of 75 and 25 ml/min per 100 g. Thus the goal of accurate assessment of average pixel properties is met, even in regions such as the cortical ribbon, in which gray and white matter cannot be separated.

Effects of the Ostwald Solubility Coefficient

We have prepared a digital compartmental model of the whole body transport kinetics of inert gaseous indicators. Arterial and mixed venous indicator concentrations, and indicator quantity in the lung, gray matter, white matter, and four major body compartments can be calculated for a great variety of administration methods, including arterial and venous injection, and inhalation using both breath-hold and rebreathing. The model assumes instantaneous equilibration of the indicator in lung airway, tissue, and blood at all times. Compartmental flows and masses, and arterial and venous delays were derived from the physiology literature. The model can be used to assess the effects of the high Ostwald solubility coefficient on the clearance of the arterial concentration, and its effects in turn on the CBF determinations. Figure 4 shows model calculations of indicator concentrations in $\mu\text{Ci/ml}$ in arterial and mixed venous blood, and in gray matter for flows of 60 and 70 ml/min per 100 g for identical 30-s breath-hold administrations of 50 mCi Xe and fluoromethane. The Ostwald solubility coefficients were assumed to be 0.15 and 1.0, respectively, and the gray matter partition coefficient to be 0.8 for Xe and 1.0 for

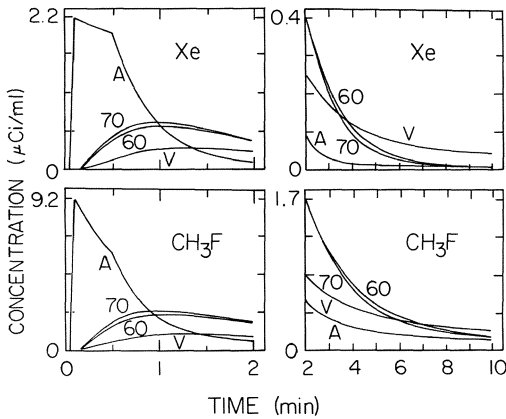


Fig. 4. Modeled indicator concentrations in $\mu\text{Ci/ml}$ in arterial and venous blood, and in gray matter for flows of 60 and 70 $\text{ml/mm per } 100 \text{ g}$, for identical breath-hold administrations of Xe and FM. The similarities and differences between the agents are described in the text

fluoromethane. The figures on the left show the time courses over the first 2 min. The most striking feature is the more than fourfold increase in concentration in the fluoromethane case. Aside from this, the two situations are very similar; the discrimination of neighboring flow values is nearly identical for the two agents. The time period from 2–10 min is shown with expanded vertical scales in the figures on the right. Interestingly, the venous concentration at 10 min is nearly the same fraction of the maximum arterial concentration for the two agents. However, the arterial concentration is a much larger fraction of the venous concentration in the fluoromethane case, slowing the whole body clearance rate at 10 min by nearly a factor of two. In the steady state, the arterial and mixed venous concentrations are expected to have the relationship (Riggs 1963):

$$C_v = C_a \left(1 + \frac{\dot{V}}{\lambda_0 F} \right) \quad (4)$$

where λ_0 is the blood Ostwald solubility coefficient and $\dot{V}$ and F are the mean ventilation rate and the cardiac output, respectively. The model ratios C_v/C_a at 10 min are very nearly equal to the values of about 8 for Xe and 2 for fluoromethane predicted by Eq. (4). The elevated arterial concentration in the fluoromethane case

tends to compress the tissue response curves vertically, with a consequent loss of discrimination between flow values. However, it also improves the statistical precision of the tissue response measurements, which tends to compensate for this loss of discrimination. In summary, while the effects of the sevenfold increase in blood Ostwald solubility relative to Xe are readily evident, the utility of fluoromethane as a blood flow indicator is not compromised.

Clinical Applications

Figure 5 shows representative images from clinical studies. Figures 5A and B are studies of a 51-year-old male with a tumor metastasized from lung carcinoma. Figure 5A shows the breakdown of the blood-brain barrier demonstrated by Ga-68 EDTA. Figure 5B shows the calculated f image in the same slice, demonstrating a corresponding 60% loss of flow relative to the contralateral area. The lesion was not detectable on the calculated solubility map. Figures 5C and D are from studies of a 57-year-old male with infarction involving most of the territory of the right middle cerebral artery. The patient had left flaccid hemiplegia, left hemisensory loss, and left hemianopsia. However, the entire bilateral occipital lobe was normal in X-ray CT. Figure 5C shows the flow map measured with the patient at rest with eyes open in a darkened room. Figure 5D shows the same slice studied using bilateral flashing light visual stimulation. Failure to stimulate the right visual cortex confirmed its total deafferentation.

The remaining figures are from the study of a 60-year-old male with a history of episodic aphasia associated with right upper extremity weakness. Figures 5E and F are flow maps demonstrating the response to increased Pa-CO_2 . Interestingly, the reactivity of the gray matter is not uniform throughout the slice, but is significantly *greater* in a focal region of the left temporal lobe that correlates well with the patient's specific clinical symptoms. Images G and H are the corresponding solubility maps from the same studies. The stability of the solubility maps while the f maps undergo significant changes in both overall scale and relative

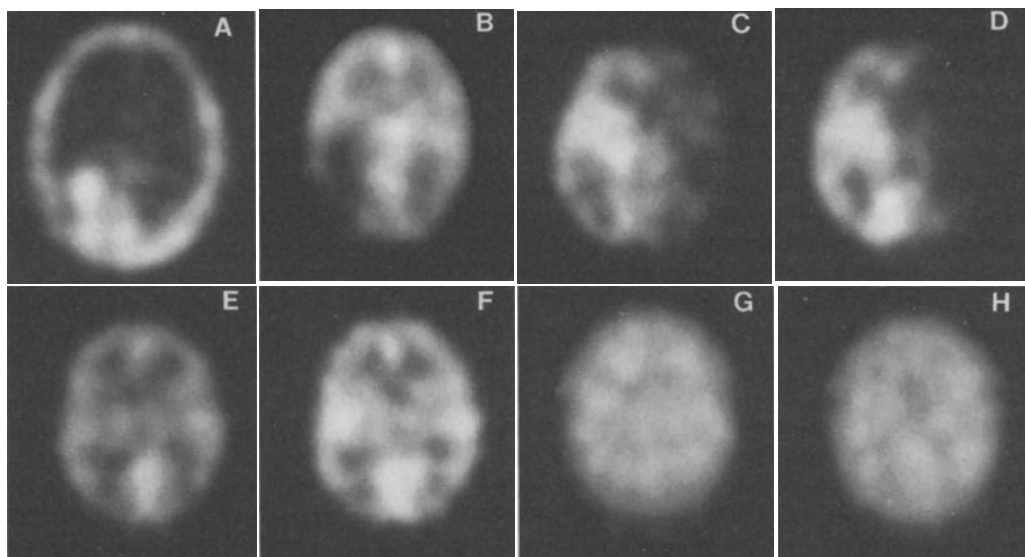


Fig. 5 A–H. Images from clinical studies. **A, B** Ga-68 EDTA local blood volume and corresponding calculated f map in a case of metastatic carcinoma. **C, D** Calculated f maps in a patient with infarction of the territory of the right middle cerebral artery. The images demonstrate the changes in occipital cortical flow with flashing light stimulus. **E, F** f maps. **G, H**

local solubility maps from studies of a patient with episodic aphasia and right extremity paresis. Images **E** and **G** correspond to normal PaCO_2 and **F** and **H** to PaCO_2 elevated by inhalation of 5% CO_2 . The flow maps demonstrate a focal area of abnormal reactivity in the left temporal lobe

spatial distribution provides encouragement that these two parameters are effectively distinguished from each other by the method.

Conclusions

The high blood Ostwald solubility coefficient of fluoromethane allows cerebral concentrations sufficient for tomographic blood flow determinations to be attained from inhalation administration. The near uniformity of its solubility in all brain tissue types allows average pixel blood flow and indicator solubility to be accurately derived from a dynamic sequence of 1-min scans following a brief breath-hold administration. Clinical applications to date have primarily involved the study of cerebral ischemia and rehabilitation after stroke. Basic development continues, with emphasis on the development of noninvasive methods of absolute scale determination.

References

1. Gatley SJ, Hichwa RD, Shaughnessy WJ et al. (1981) ^{18}F -labelled lower fluoroalkanes: reactor-produced gaseous physiological tracers. *Int J Appl Radiat Isot* 32: 211–214
2. Gilboe DD, Betz AL, Langebartel DA (1973) A guide for the isolation of the canine brain. *J Appl Physiol* 34: 534–537
3. Holden JE, Gatley SJ, Hichwa RD et al. (1981) Cerebral blood flow using PET measurements of fluoromethane kinetics. *J Nucl Med* 22: 1084–1088
4. Lennox GA, Wyper DJ, Jawad K (1975) A Xe-133 inhalation technique for rCBF measurement: normal and abnormal flow patterns. In: Harper AM, Jennett WB, Miller JD, Kowar JO (eds) *Blood flow and metabolism in the brain*. Churchill Livingstone, Edinburgh pp 8.12–8.16
5. Phelps ME, Hoffman EJ, Huang SC et al. (1978) ECAT: a new computerized tomographic imaging system for positron-emitting radiopharmaceuticals. *J Nucl Med* 19: 635–647
6. Riggs DS (1963) *The mathematical approach to physiological problems*. MIT, Cambridge pp 328

A Method for Local Evaluation of the Volume of Rapidly Exchangeable Water in the Human Brain

By J. C. Depresseux

Introduction

The quest for optimal accuracy, precision, sensitivity, and spatial and temporal resolution of the measurements, combined with considerations of feasibility and noninvasiveness has led physiologists and biomathematicians to design many different methodological approaches to the determination of regional cerebral blood flow (rCBF) in man by means of externally detected radioactive inert indicators. Those methods are all based upon variants of the model initially designed by Kety and Schmidt (1948) to describe the kinetics of nitrous oxide in the whole brain.

One of the most cumbersome features of that group of techniques is the unavoidable introduction into the equations of a parameter – tissue-to-blood partition coefficient or distribution volume – characterizing tissular distribution of the indicator. Uncertainty about the regional value to be attributed to that parameter limits the accuracy of the results, especially when pathological cases are concerned.

The possibility of quantitative evaluation of tissular radioactive concentrations of indicators by positron emission tomography (PET) (Eichling et al. 1977; Hoffman et al. 1979; Huang et al. 1979a) enables the determination of the distribution volume of the tracers in CBF measurements (Yamamoto et al. 1979; Huang et al. 1981; 1982).

Among the numerous inert positron-emitting indicators used for evaluating rCBF (Ter-Pogossian et al. 1969; Jones et al. 1976; Subramanyam et al. 1978; Frackowiak et al. 1980; Yamatoto et al. 1979; Holden et al. 1981; Ginsberg et al. 1981), ^{15}O -labeled water has the advantages of diffusing rapidly across the blood-brain barrier (Raichle et al. 1976) and of being an important biological component whose distribution and kinetics are of utmost interest in themselves.

The purpose of the present investigation was the design, validation, and application of an original numerical method for the simultaneous determination of rCBF and the corresponding distribution volume of radiowater within the brain by bolus inhalation of C^{15}O_2 and sequential PET, with special attention paid to the possibility of evaluating the rapidly exchangeable water volume in the brain.

Material and Methods

The data for numerical analysis and for retro-simulation studies were obtained from normal subjects. Two modes of administration of H_2^{15}O were sequentially used in five individuals: first, bolus inhalation from a bag of air mixed with 80 mCi C^{15}O_2 , and second, after a delay of 20 min, continuous inhalation of 0.5 mCi/min C^{15}O_2 for 10 min.

Sequential PET detections of 53 s were performed from time zero of inhalation for 8 min with a whole body positron emission tomograph (ECAT II, EG & G ORTEC) (Phelps et al. 1978; Soussaline et al. 1979; Williams et al. 1979). Subjects remained in stable basal psychophysiological condition. Arterial blood gases were monitored. The tomographic plane was adjusted parallel to the orbitomeatal reference plane of the subject at a distance of 5 cm above it. Data were collected and reconstructed in the medium resolution mode (FWHM = 13.2 mm). The time course of radioactive concentration within the arterial blood was measured by catheterization of one humeral artery and by sequential counting of blood samples. Well counter and PET measurements were calibrated by imaging and counting radioactive concentrations in a pie phantom 20 cm in diameter.

Results and Discussion

General Equation of the Model

Whatever the method and time course of administration of the indicator, its concentration in each element of organ may be differentially described by the general equation:

$$dC_{bi}(t) = F_i C_a(t) dt - F_i C_{vi}(t) dt - \lambda C_{bi}(t) dt \quad (1)$$

with subscript i identifying the organ element under concern; t , the time; C_b , the radioactivity of radiowater within the organ element per unit mass of organ; C_a , the radioactivity of radiowater per unit volume of arterial blood; C_v , the radioactivity of radiowater per unit volume of venous blood; F , the blood flow perfusing the organ element in unit volume of blood per unit mass of organ and per unit time; λ , the radioactive decay constant of ^{15}O .

The variable $C_{vi}(t)$ is not accessible to measurement but may be expressed using the central volume theorem:

$$C_{vi}(t) = C_{bi}(t) / V_i \quad (2)$$

with V_i , the distribution volume of radiowater within the organ element i , expressed in unit volume per unit mass of organ (tissue + blood). Eqs. (1) and (2) classically give a general expression of the kinetics of the indicator through the element i of organ:

$$dC_{bi}(t) = F_i C_a(t) dt - (k_i + \lambda) C_{bi}(t) dt \quad (3)$$

with $k_i = F_i / V_i$

Solution from Data at Equilibrium

Most published research using C^{15}O_2 and H_2^{15}O for the evaluation of rCBF utilizes continuous inhalation of C^{15}O_2 and PET detection performed at equilibrium of radioactive concentration of the indicator within blood and tissues (Ackerman et al. 1981; Lenzi et al. 1981; Bousser et al. 1980; Baron et al. 1978, 1981; Frackowiak et al. 1981).

The left member of equation (3) goes to zero at equilibrium, whereas $C_{bi}(t)$, $C_a(t)$ and $V_i(t)$

reach constant values. The CBF is then computed as:

$$F_i = \frac{\lambda}{C_a(\text{equ}) / C_{bi}(\text{equ}) - 1 / V_i(\text{equ})} \quad (4)$$

The accuracy, sensitivity and precision of this method were examined in recent papers (Jones et al. 1979, 1982; Huang et al. 1979b; Lam-mertsma et al. 1982). One of the limitations lies in the fact that the amount of data gathered at equilibrium is not sufficient for the evaluation of $V_i(\text{equ})$, the value of which is presently derived from the normal total water content of brain tissue as measured by wet and dry weight differences. Any systematic error in $V_i(\text{equ})$ is unfortunately propagated by Eq.(4) (Jones et al. 1982).

Solutions from Integrated Data from $t=0$

The only way to gather sufficient information to evaluate F_i and V_i simultaneously is to collect data by rapid sequential PET after a bolus inhalation of C^{15}O_2 (Raichle et al. 1981; Huang et al. 1981, 1982) or during the equilibration phase occurring when the indicator is continuously inhaled.

Integration of Eq.(3) may be performed in two different ways.

The first possible solution uses *mathematical integration* (Huang et al. 1981) in the form of a convolution integral:

$$C_{bi}(t) = F_i \int_0^t e^{-(k_i + \lambda)(t - \tau)} \cdot C_a(\tau) d\tau \quad (5)$$

The numerical analysis giving F_i and V_i from observed data may be differently developed, depending on the expression which is given to the input function $C_a(t)$. For example, if $C_a(t)$ can be expressed as

$$C_a(t) = C_a(0) e^{-\mu t} \quad (6)$$

the formulae for computing F_i and V_i are as follows:

$$F_i = r_1 (k_i + \lambda) \quad (7)$$

$$V_i = F_i / k_i \quad (8)$$

with

$$k_i = \frac{\lambda + \mu - \lambda \mu r_2}{\mu r_2 - 1} \quad (9)$$

$$r_1 = \frac{\int_0^{\infty} C_{bi}(t) dt}{\int_0^{\infty} C_a(t) dt} \quad (10)$$

$$r_2 = \frac{\int_0^{\infty} t C_{bi}(t) dt}{\int_0^{\infty} C_{bi}(t) dt} \quad (11)$$

This approach suffers from the difficulty of necessarily (a) extrapolating of observed data up to $t = \infty$ and (b) fitting the observed input function with a suitable equation.

The second approach avoids convolution and uses *numerical integration* by integrating both members of Eq.(3):

$$C_{bi}(t) = F_i \int_0^t C_a(t) dt - (k_i + \lambda) \int_0^t C_{bi}(t) dt \quad (12)$$

Developments of Eq.(12) for data processing were recently published by Huang et al. (1982)

$$F_i = \frac{\lambda \int_0^T C_{bi}^*(t) dt - C_{bi}(T)}{\int_0^T C_a(t) dt \int_0^T C_{bi}^*(t) dt - \int_0^T C_{bi}(t) dt - \int_0^T C_a^*(t) dt} \quad (13)$$

$$V_i = \frac{\lambda \int_0^T C_{bi}^*(t) dt - C_{bi}(T)}{\lambda \int_0^T C_a^*(t) dt - C_{bi}(T) \int_0^T C_a(t) dt / \int_0^T C_{bi}(t) dt} \quad (14)$$

with superscript * denoting radioactivities being time-corrected to time zero.

Contrary to the preceding method, this has the advantages of (a) using time integration of data from $t=0$ to $t=T$ without necessitating any extrapolation and (b) integrating the input function by numerical procedure without mathematical fitting of data.

The model in use is exactly the same in both approaches but the latter fits actual data processing imperatives more easily.

Whatever procedure is chosen, it remains a

prerequisite that the parameter V_i be submitted to very critical examination, taking into consideration (a) that it could be variable with time during the period of invasion of the organ by the indicator and (b) that it could not be equal to the tissue-blood partition coefficient of water as computed from brain and blood total content of water.

One of the objectives of the present investigation was to confront those two hypotheses with arguments derived from observed and simulated kinetics of radiowater within the brain tissue.

Stationariness of Volume V_i

The stationariness of the investigated volume V_i after bolus inhalation of $C^{15}O_2$ was tested by solving Eq.(3) in the form of the convolution integral (16): if the input function is generally described as

$$C_a(t) = C_a(0) e^{-\mu t} + A(t) \quad (15)$$

Eq.(3) may be solved:

$$C_{bi}(t) = \frac{C_a(0)F_i}{k_i + \lambda - \mu} (e^{-\mu t} - e^{-(k_i + \lambda)t}) + F_i e^{-(k_i + \lambda)t} * A(t) + \varepsilon_i(t) \quad (16)$$

with initial condition $C_{bi}(0) = 0$.

The parameter k_i in Eq.(16) is constant with time, with a resulting time-dependent error denoted $\varepsilon_i(t)$ if V_i is variable with time.

In all studied cases the arterial blood concentration measurements fitted the first term of Eq. (15) in the time interval ($1 \text{ min} \leq t < 10 \text{ min}$) with $r > 0.99$, μ being equal to the sum of the radioactive decay constant of ^{15}O and of the physiological decay parameter of the indicator concentration in the arterial blood. The difficult-to-fit function $A(t)$ vanishes for $t \geq 1 \text{ min}$. Equation (16) is simplified and linearized as

$$\frac{C_{bi}(t)}{C_a(0)} = \frac{F_i}{k_i + \lambda - \mu} (e^{-\mu t} - e^{-(k_i + \lambda)t} 25\%) \quad (17)$$

and

$$Y_i(t, k_i) = \frac{F_i}{k_i + \lambda - \mu} X_i(t, k_i) \quad (18)$$

The parameter k_i in Eqs.(17) and (18) is then evaluated by iteration with search for best linear fitting. Figure 1 illustrates that it is possible to determine one value of k_i satisfying the linearity hypothesis of the relationship between observed values of $C_{bi}(t)$ and the modeled expression (17), for $t \geq 1$ min.

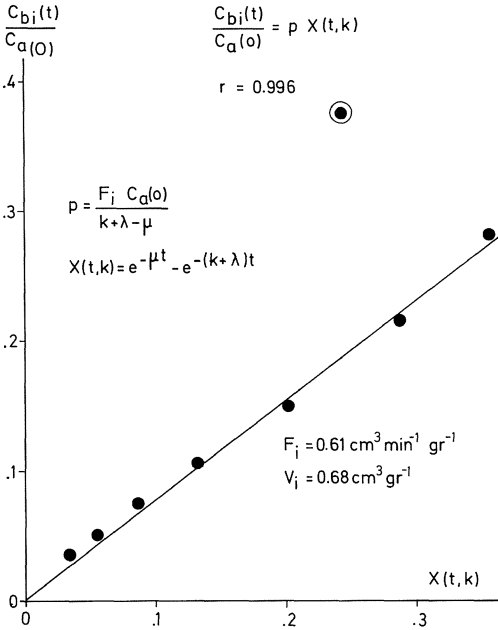


Fig. 1. Example of the linear relationship obtained by application of Eqs.(17) and (18) between observed and simulated values of $C_{bi}(t)$ in the time interval ($1 \text{ min} \leq t < 8 \text{ min}$): k_i was obtained by iteration and search for the best linear fit

Retrosimulation using Eq.(17) satisfyingly reproduced observed data within the same time interval in all cases (Fig. 2).

These convergent results validate Eq.(17) and the derived computing procedure. They allow furthermore the consideration that the second and third terms of the right member of Eq.(16) vanish for $t \geq 1$ min after a bolus inhalation of $C^{15}O_2$ and that V_i is therefore constant after that delay.

The bad mathematical prevision of $C_{bi}(t)$ by Eq.(17) for $t < 1$ min could result from the undefined input function $A(t)$ and from a possible initial variation in V_i , the incidence $\varepsilon_i(t)$ of which vanishes after 1 min.

In order to test the validity of the hypothesis of an initial variation of V_i , Eqs.(13) and (14) were successively applied to observed and simulated data.

Table 1 shows the bad convergence of computed values for V_i as obtained from observed data if increasing integration times are used in Eq.(14). As the input function is integrated by numerical procedure and thus fully determined from $t=0$, the bad convergence of results is suspected to be linked to a variation of V_i , the incidence of which is propagated in the results. The error propagation in Eqs.(13) and (14) due to an initial variation of V_i was studied by applying those equations to the analysis of simulated data obtained through Eq.(17) (Table 2). Both numerical procedures are in excellent correlation, provided that the whole transfer function $C_{bi}(t)$ is obtained from Eq.(17) from

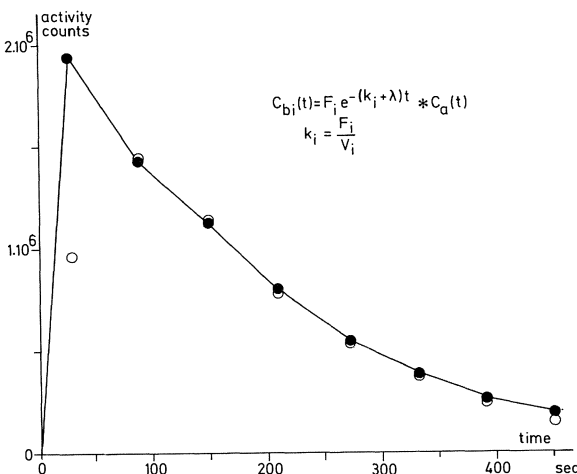


Fig. 2. Retrosimulation of $C_{bi}(t)$ using Eq.(17) and values of F_i and k_i obtained by iteration procedure. Plain points, observed data; circles, simulated values

$t=0$, i. e., that V_i is strictly constant. Slight fluctuations in the results are linked to approximations occurring within the numerical integration process.

In contrast, the introduction of a higher initial value of $C_{bi}(t)$, corresponding to a transitorily higher value of V_i , leads to an error which is propagated by Eqs. (13) and (14).

Comparison of results obtained from observed data, using both iteration and numerical integration procedures of analysis (Table 1), thus allows the inference that the discordance between observed and computed values of $C_{bi}(t)$ for $t < 1$ min results not only from the difficulty in describing the early time course of $C_a(t)$

Table 1. Values of rCBF F_i and distribution volume of water V_i typically obtained from observed data with Eqs. (13) and (14), with increasing integration intervals from $t=0$ to $t=T$

T (s)	$C_{bi}(T)$ (ips/g)	F_i (cm ³ /min per gram)	V_i (cm ³ /g)
29.5	16 145	-2.37	0.32
91.5	12 960	-0.69	0.31
155.5	9 290	-0.20	0.23
219.5	4 549	0.06	-0.27
283.5	6 251	0.15	6.70
347.5	2 892	0.26	1.00
411.5	2 179	0.26	1.03
475.5	1 319	0.32	0.88

Table 2. Values of F_i and V_i obtained with Eqs. (13) and (14), with increasing integration intervals from $t=0$ to $t=T$. Data were obtained by simulation using Eq. (17). A disturbance introduced in the first value of $C_{bi}(t)$ is propagated by Eqs. (13) and (14)

T (s)	V_i constant in Eq. (17)			V_i initially variable [Eq. (16)]		
	$C_{bi}(T)$ (ips/g)	F_i (cm ³ /min per gram)	V_i (cm ³ /g)	$C_{bi}(T)$ (ips/g)	F_i (cm ³ /min per gram)	V_i (cm ³ /g)
26.5	15 998	-5.04	0.20	32 138	-10.12	0.39
86.5	22 244	-2.78	0.26	22 244	-6.47	0.42
146.5	17 895	-0.97	0.30	17 895	-2.11	0.42
205.5	12 598	-0.18	0.22	12 598	-0.04	0.09
267.5	8 261	0.19	1.48	8 261	0.68	0.66
327.5	5 377	0.37	0.69	5 377	0.95	0.62
386.5	3 493	0.46	0.64	3 493	1.05	0.61
447.5	2 226	0.51	0.61	2 226	1.11	0.61

Table 3. Values obtained from observed data for F_i and V_i , using different numerical approaches: mathematical integration [Eqs. (7) to (11)], numerical integration [Eqs. (13) and (14)], and iteration procedure [Eqs. (17) and (18)]

Region	F_i (cm ³ /min per gram)			V_i (cm ³ /g)		
	Eq. (17)	Eq. (13)	Eq. (7)	Eq. (18)	Eq. (14)	Eq. (8)
1	0.57	0.57	1.09	0.59	0.68	0.69
2	0.68	0.70	1.90	0.67	0.79	0.76
3	0.70	0.98	3.47	0.63	0.71	0.70
4	0.70	0.79	2.47	0.69	0.78	0.74
5	0.69	0.84	1.61	0.56	0.62	0.67
6	0.84	0.91	3.13	0.52	0.57	0.58
7	0.60	0.57	1.20	0.59	0.69	0.69
8	0.56	0.62	1.58	0.72	0.81	0.76
9	0.69	0.90	2.25	0.64	0.70	0.74
10	0.48	0.55	1.36	0.70	0.76	0.71
11	0.71	0.79	2.11	0.57	0.64	0.64
12	0.68	0.66	1.68	0.52	0.59	0.59

mathematically, but also from an *initial variation of the distribution volume of radiowater*.

That variation of V_i during the period of invasion of brain tissue by radiowater could be considered as the result of an initial predominantly unidirectional extraction of the indicator, as was described in Crone's single-transit extraction model (1963).

The present investigation therefore invalidates classical solutions (5) and (12) of the differential Eq.(3), because the parameter V_i is time-dependant during the initial part of the integration interval.

The error resulting from that variation opportunely vanishes with time after the invasion period, with the possibility of a different approach to the data processing.

Results respectively obtained by mathematical integration [Eqs. (7) to (11)], by numerical integration [Eqs.(13) and (14)], and by iteration procedure are comparatively reported in Table 3.

Values of V_i with Different Input Functions

The stationariness of V_i for $t \geq 1$ min after bolus inhalation of $C^{15}O_2$ could lead to attributing the computed volume to a physically stable exchangeable water compartment. Additional arguments in favor of this hypothesis were derived by using a differently shaped input function of the indicator and by adapting the above iteration method.

Homologous values of F_i and V_i , obtained respectively from bolus (index B) and continuous (index C) inhalation of $C^{15}O_2$ are in good correlation, with a small systematical difference probably linked to the well-known difficulty of realizing a strictly constant rate of administration of the tracer with the continuous inhalation technique:

$$F_C = (0.101 + 0.787 F_B) \text{ cm}^3/\text{min g}$$

$$r = 0.942, n = 12$$

$$V_C = (-0.127 + 1.160 V_B) \text{ cm}^3/\text{g}$$

$$r = 0.948, n = 12$$

Figure 3 exemplifies values of F_i and V_i obtained in a normal subject in 5.3-cm² regions. The mean values of F_i and V_i in normal predominantly gray (index G) and white (index W) regions were (± 1 SD):

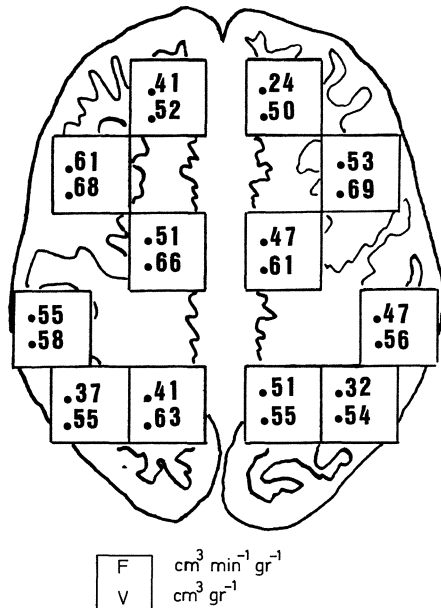


Fig. 3. Values of F_i and V_i obtained in a normal subject (plane 5 cm above the orbitomeatal line)

$$F_G = 72.7 \pm 8.0 \text{ cm}^3/\text{min } 100 \text{ g}$$

$$F_W = 31.4 \pm 6.3 \text{ cm}^3/\text{min } 100 \text{ g}$$

$$V_G = 69.6 \pm 5.4 \text{ cm}^3/100 \text{ g}$$

$$V_W = 59.3 \pm 5.0 \text{ cm}^3/100 \text{ g}$$

The volumes of rapidly exchangeable water in the brain matter which are derived by the present method are less than the total water content of gray and white matter (determined as around 0.85 cm³/g in cortical gray matter and 0.71 cm³/g in white matter (Torack et. al. 1976). The lower values of rapidly exchangeable water content as compared with total water content presumably correspond to the multicompartmental distribution of free and linked molecules of water within the cerebral tissue.

Conclusions

As previously stated (Huang et al. 1981, 1982), use of $H_2^{15}O$ and PET potentially allows non-invasive and quantitative evaluation of local cerebral blood flow and local distribution volume of radiowater within the human brain.

The present investigation, using detection and simulation data obtained with differently

shaped input functions of radiowater, provides arguments which allow us to infer that the cerebral distribution volume of radiowater does vary with time during the initial invasion period of tissue by the indicator and demonstrates that the observed variation leads to systematic errors and nonconvergence in results as long as the classical mathematical or numerical integration procedures are utilized for processing the data.

A different approach to the problem shows (a) that the regional distribution volumes of radiowater within the brain remain constant for 1–8 min after the bolus inhalation of $C^{15}O_2$ and (b) that the computed distribution volumes do not depend on the shape of the input function, be it a degraded delta function after bolus inhalation or a degraded square-wave function during continuous inhalation of $C^{15}O_2$.

Results argue for the validity of the model and its numerical expression and allow the so-determined distribution volume of radiowater to be considered as equal to a physiologically meaningful volume of rapidly exchangeable water of the brain tissue.

Applications of the method to clinical and pharmacological investigations are presently planned.

Acknowledgments. The author is grateful to J. P. Cheslet, J. Hodiaumont-Demblon, N. Walch, D. Lamotte, L. Quaglia, J. P. Peters, and G. Del Fiore for their respective contributions to his research. The work was supported by Grant 1.5.278.82 from the Belgian National Foundation for Scientific Research.

References

1. Ackerman RH, Correia JA, Alpert NM, Baron JC, Gouliamos A, Grotta JC, Brownell GL, Taveras JM (1981) Positron imaging in ischemic stroke disease using compounds labeled with ^{15}O . *Arch Neurol* 38: 537–547
2. Baron JC, Comar D, Boussier MG, Soussaline F, Crouzel C, Plummer D, Kellershohn C (1978) Etude tomographique chez l'homme du débit sanguin et de la consommation d'oxygène du cerveau par inhalation continue d'oxygène- ^{15} . *Rev Neurol* 134: 545–556
3. Baron JC, Boussier MG, Comar D, Castaigne P (1981) Crossed cerebellar diaschisis in human supratentorial brain infarction. *Trans. Am Neurol Assoc* 105: 459–461
4. Boussier MG, Baron JC, Iba-Zizen MT, Comar D, Cabanis E, Castaigne P (1980) Migrainous cerebral infarction: a tomographic study of cerebral blood flow and oxygen- ^{15} inhalation technique. *Stroke* 11: 145–148
5. Crone C (1963) The permeability of capillaries in various organs as determined by use of the "indicator diffusion" method. *Acta Physiol Scand* 58: 292–305
6. Eichling JO, Higgins CS, Ter-Pogossian MM (1977) Determination of radionuclide concentrations with positron C.T. scanning (P.E.T.). *J Nucl Med* 18: 845–847
7. Frackowiack RSG, Lenzi GL, Jones T, Heather JD (1980) Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man, using ^{15}O and positron emission tomography: theory, procedure, and normal values. *J Comput Assist Tomogr* 4: 727–731
8. Frackowiak RSG, Pozzilli C, Legg NJ, Du Boulay GH, Marshall J, Lenzi GL, Jones T (1981) Regional cerebral oxygen supply and utilization in dementia. A clinical and physiological study with oxygen- ^{15} and positron tomography. *Brain* 104: 753–778
9. Ginsberg MD, Busto R, Lockwood AH, Finn RD, Campbell JA, Boothe TE (1981) ^{11}C -Iodoantipyrine for the measurement of regional cerebral blood flow by positron emission tomography: synthesis and validation studies. *J Cereb Blood Flow Metab* 1 [Suppl. 1]: 37–38
10. Hoffman EJ, Huang SC, Phelps ME (1979) Quantitation in positron emission computed tomography: 1. Effect of object size. *J Comput Assist Tomogr* 3: 299–308
11. Holden JE, Gatlen SJ, Hichwa RD, Ip WR, Shaughnessy WJ, Nickles RJ, Polcyn RE (1981) Regional cerebral blood flow using positron emission tomography measurement of fluoromethane kinetics. *J Cereb Blood Flow Metab* 1 [Suppl 1]: 35–36
12. Huang SC, Hoffman EJ, Phelps ME, Kuhl DE (1979a) Quantitation in positron emission computed tomography: 2. Effects of inaccurate attenuation correction. *J Comput Assist Tomogr* 3: 804–814
13. Huang SC, Phelps ME, Hoffman EJ, Kuhl DE (1979b) A theoretical study of quantitative flow measurements with constant infusion of short-lived isotopes. *Phys Med Biol* 24: 1151–1161
14. Huang SC, Phelps ME, Carson R, Hoffman EJ, Plummer D, MacDonald N, Kuhl D (1981) Tomographic measurements of local cerebral blood flow in man with ^{15}O water. *J Cereb Blood Flow Metab* 1 [Suppl 1]: 31–32
15. Huang SC, Carson R, Phelps ME (1982) Measurement of local blood flow and distribution

- volume with short-lived isotopes: a general input technique. *J Cereb Blood Flow Metab* 2: 99–108
16. Jones T, Chesler DA, Ter-Pogossian MM (1976) The continuous inhalation of oxygen-15 for assessing regional oxygen extraction in the brain of man. *Br J Radiol* 49: 339–343
 17. Jones SC, Reivich M, Greenberg JH (1979) Error propagation in the determination of cerebral blood flow and oxygen metabolism with inhalation of C15O₂ and 15O₂. *Acta Physiol Scand* 60 [Suppl 72]: 228–229
 18. Jones SC, Greenberg JH, Reivich M (1982) Error analysis for the determination of cerebral blood flow with the continuous inhalation of 15O-labeled carbon dioxide and positron emission tomography. *J Comput Assist Tomogr* 6: 116–124
 19. Kety SS, Schmidt CF (1948) The nitrous oxide method for the quantitative determination of cerebral blood flow in man: theory, procedure and normal values. *J Clin Invest* 27: 476–483
 20. Lammertsma AA, Heather JD, Jones T, Frackowiak RSJ, Lenzi GL (1982) A statistical study of the steady state technique for measuring regional cerebral blood flow and oxygen utilization using 15-O. *J Comp Assist Tomogr* 6: 566–573
 21. Lenzi GL, Frackowiak RS, Jones T, Heather JD, Lammertsma AA, Rhodes CG, Pozzilli C (1981) CMRO₂ and CBF by the oxygen-15 inhalation techniques: results in normal volunteers and cerebrovascular patients. *Eur Neurol* 20: 285–290
 22. Phelps ME, Hoffman EJ, Huang SC, Kuhl DE (1978) ECAT: a new computerized tomographic imaging system for positron-emitting radiopharmaceuticals. *J Nucl Med* 19: 645–647
 23. Raichle ME, Eichling JO, Straatman MG, Welch MJ, Larson KB, Ter-Pogossian M (1976) Blood-brain barrier permeability of 11C-labeled alcohols and 15O-labeled water. *Am J Physiol* 230: 543–552
 24. Raichle ME, Markham J, Larson K, Grubb RL, Welch MJ (1981) Measurement of local cerebral blood flow in man with positron emission tomography. *J Cereb Blood Flow Metab* 1 [Suppl 1]: 19–20
 25. Soussaline F, Todd-Pokropek AE, Plummer D, Comar D, Houle S, Kellershohn C (1979) The physical performances of a single slice positron tomographic system and preliminary results in a clinical environment. *J Nucl Med* 4: 237–249
 26. Subramanyam N, Alpert NM, Hoop B, Brownell GL, Taveras JM (1978) A model for regional cerebral oxygen distribution during continuous inhalation of 15 O₂, C15O and C15O₂. *J Nucl Med* 19: 48–53
 27. Ter-Pogossian MM, Eichling JO, Davis DO, Welch MJ, Metzger JM (1969) The determination of regional cerebral blood flow by means of water labeled with radioactive oxygen 15. *Radiology* 93: 31–40
 28. Torack RM, Alcalá H, Gado M (1976) Water, specific gravity and histology as determinants of diagnostic computerized Cranial Tomography (CCT). In: Pappius HM, Feindel W (eds) *Dynamics of brain edema*. Springer, Berlin Heidelberg New York, pp 129–137
 29. Williams CW, Crabtree MC, Burgiss SG (1979) Design and performance characteristics of a positron emission computed axial tomograph – ECAT II. *IEEE Trans Nucl Sc NS-26*: 619–627
 30. Yamamoto YL, Thompson C, Meyer E, Nukui H, Matsunaga M, Feindel W (1979) Three-dimensional tomographical regional cerebral blood flow in man measured with high efficiency mini-BGO two ring positron device using Krypton-77. *Acta Neurol Scand* 60 [Suppl 72]: 186–187

Oxygen Consumption

(Chairman: M. Reivich)

Measurement of Regional Cerebral Oxygen Consumption and Blood Flow in Man

By M. E. Raichle, P. Herscovitch, W. Martin, and J. Markham

Introduction

The measurement of oxygen utilization and blood flow by positron emission tomography (PET) has presented a number of interesting challenges in the design of quantitative tracer strategies that respect the spatial and temporal resolution limitations of current PET tomographic systems. Because of the limited temporal resolution of most tomographic systems currently in operation, investigators have generally employed equilibrium strategies (Frackowiak et al. 1980) utilizing the continuous inhalation of radiolabeled oxygen and carbon dioxide. Such an approach yields quantitative values for local brain oxygen utilization and blood flow but does suffer from a number of limitations. The most notable of these is the long time necessary to accomplish such a measurement (e.g., 30 min). With the advent of PET imaging systems with improved temporal and spatial resolution (Ter-Pogossian et al. 1982) we have sought to employ techniques for the measurement of local cerebral blood flow and oxygen utilization that can be accomplished in a relatively short period of time (i.e., less than 1 min/measurement). Our general approach to this is detailed below.

Cerebral Blood Flow

Recent advances in the design of PET systems now permits us to measure local cerebral blood flow in a manner analogous to the tissue autoradiographic method originally proposed by Landau and his colleagues (1955) and later refined by Reivich et al. (1969) and Sakurada et al. (1978). The method is an application of the principles of inert gas exchange between blood and tissue developed by Kety (1951). In this technique as originally applied to animals, a

freely diffusible radiolabeled tracer is infused intravenously for 1 min followed immediately by decapitation. Quantitative autoradiograms of brain slices and the tissue activity curve of arterial blood radioactivity form the data base from which local cerebral blood flow is calculated. The PET image can be substituted for the tissue autoradiogram provided that the image is obtained in a relatively short period of time (i.e., less than 1 min). We have employed this technique for the measurement of local cerebral blood flow using positron emission tomography and ^{15}O -labeled water as the diffusible indicator. In order to establish the validity of this technique we have measured local cerebral blood flow with PET and compared it directly with local cerebral blood flow measured in a single hemisphere of an adult baboon. Tracer was administered intravenously for the autoradiographic technique employed with PET and into the internal carotid artery for the measurement of water clearance using standard tracer principles. Twenty-three simultaneous measurements made in adult baboons revealed an excellent correlation between the two methods of measurement over a flow range of 10–60 ml/100 g per minute ($r = 0.96$, $p < 0.001$). Based on these experiments, which represent the first validation of this now classic technique, we have commenced studies in patients under a variety of circumstances.

Cerebral Oxygen Utilization

We are currently employing an adaptation of a technique developed earlier in our laboratory (Ter-Pogossian et al. 1970; Raichle et al. 1976) for the measurement of local oxygen utilization. As originally developed, this technique was based upon the bolus injection, into the internal carotid artery, of both ^{15}O -labeled water and ^{15}O -labeled molecular oxygen. From the

time activity curve generated by the intracarotid injection of ^{15}O -labeled molecular oxygen, it was possible to determine with considerable accuracy the local extraction of oxygen by brain tissue. From the injection of ^{15}O -labeled water we were able to compute the local blood flow and combining these two measurements the local oxygen utilization. This strategy proved quite satisfactory when employed with intracarotid injection of tracers and external detection of these tracers. Initial data in patients and subsequent direct comparison with an independent measurement of oxygen consumption (i.e., Fick Principle) clearly established its validity (Raichle et al. 1976).

A feature of this approach that was not pursued beyond an initial observation was the fact that the water of metabolism cleared from the tissue in a manner identical with radiolabeled water injected. In fact, it was possible to compute local cerebral blood flow from either the clearance of ^{15}O -labeled water of metabolism or ^{15}O -water injected directly as water (Ter-Pogossian et al. 1969). Although unimportant at the time this fact became important when we contemplated applying this approach to PET. PET, despite recent improvements which permit temporal resolution of less than a minute, does not have the capacity to describe a time activity curve with sufficient accuracy to permit implementation of this technique as initially described. Furthermore, this technique assumes that all of the administered tracers are present in the desired region of interest at the onset of the analysis (i.e., the input is a Δ function). Clearly this could be achieved, in the case of the brain, only with a direct internal carotid artery injection, an approach to tracer administration antithetical to the philosophy of PET which strives for safety of administration (i.e., intravenous or inhalation) and an image of the entire organ, not just a cerebral hemisphere of the brain. An approach to accommodate these several issues is outlined below.

The fact that tracer ^{15}O -labeled water, whether administered initially as H_2^{15}O or O^{15}O , clears from the tissue in the same manner makes it possible to compute the local fractional extraction of oxygen from the ratio of the tissue activity (minus blood activity) following oxygen administration, to the tissue activity (minus blood activity) following H_2^{15}O administration. Be-

cause the water present in the tissue from either tracer clears and recirculates to the tissue in an identical manner, the concentration of tracer within the tissue represents the fraction of tracer oxygen that actually entered the tissue. Two assumptions are implicit, of course, in this simple analysis. First, one must assume that tissue contains no unmetabolized ^{15}O -labeled oxygen. Because of the low solubility of oxygen in brain tissue water this assumption is easily met. A second important assumption is that ^{15}O -labeled water is a freely diffusible tracer. Previous studies in our laboratory have clearly indicated that at high flow rates water does not freely move from the blood to the brain; however at flow rates to be encountered clinically this should not be a problem and this tracer can be viewed as freely diffusible. Under these circumstances then, this simple modification of the technique originally described by Ter-Pogossian et al. (1970) and later validated by Raichle et al. (1976) will permit a simple determination of the local fractional extraction of oxygen following the single inhalation of a breath of air containing ^{15}O -labeled molecular oxygen and a subsequent intravenous injection of ^{15}O -labeled water. To correct the scans for the presence of tracer within the vascular compartment following the administration of each of these radiopharmaceuticals, ^{15}O -labeled carbon monoxide is subsequently administered to measure the local cerebral blood volume.

Data collection following the administration of ^{15}O -labeled water and ^{15}O -labeled molecular oxygen is 40 s in each circumstance. Measurement of cerebral blood volume is made over a period of 5 min. The total measurement time, therefore, is less than 10 min. This is lengthened somewhat in actual practice depending on the rapidity with which the tracers can be delivered to the laboratory, but under no circumstances does it take more than 20 min to complete this entire measurement.

Conclusions

As outlined above, we are currently employing nonequilibrium methods for the measurement of local cerebral oxygen utilization and blood flow using positron emission tomography and

radiopharmaceuticals labeled with ^{15}O . Both of the techniques involved have been directly validated by comparison with well-established techniques for the measurement of both parameters. Their implementation with PET appears quite satisfactory and should lead to reliable measurements of local oxygen utilization and blood flow in humans using this new imaging modality.

Acknowledgment. This research was supported by NIH Grants HL13851, NS06833, and the McDonnell Center for the Study of Higher Brain Function.

References

1. Frackowiak RSJ, Lenzi G-L, Jones T, Heather JD (1980) Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man using ^{15}O and positron emission tomography: theory, procedure, and normal values. *J Comput Assist Tomogr* 4: 727–736
2. Kety SS (1951) The theory and applications of the exchange of inert gas at the lung and tissues. *Pharmacol Rev* 3: 1–41
3. Landau WM, Freygang WH Jr, Rowland JP, Sokoloff L, Kety SS (1955) The local circulation of the living brain; values in anesthetized and unanesthetized cat. *Trans Am Neurol Assn* 80: 125–129
4. Raichle ME, Grubb RL Jr, Eichling JO, Ter-Pogossian MM (1976) Measurement of brain oxygen utilization with radiotracer oxygen-15. Experimental verification. *J Appl Physiol* 40: 638–640
5. Reivich M, Jehle J, Sokoloff L, Kety SS (1969) Measurement of regional cerebral blood flow with antipyrine- ^{14}C in awake cats. *J Appl Physiol* 27: 296–300
6. Sakurada O, Kennedy C, Jehle J, Brown JD, Carbon GL, Sokoloff L (1978) Measurement of local cerebral blood flow with iodo- ^{14}C -antipyrine. *Am J Physiol* 234: H59–H66
7. Ter-Pogossian MM, Eichling JO, Davis DO, Welch MJ, Metzger JM (1969) The determination of regional blood flow by means of water labeled with radioactive oxygen-15. *Radiology* 93: 31–40
8. Ter-Pogossian MM, Eichling JO, Davis DO, Welch MJ (1970) The measure in vivo of regional cerebral oxygen utilization by means of oxyhemoglobin labeled with radioactive oxygen-15. *J Clin Invest* 49: 381–391
9. Ter-Pogossian MM, Ficke DC, Hood JT Sr, Yamamoto M, Mullani NA (1982) A positron emission tomograph utilizing cesium fluoride scintillation detectors. *J Comput Assist Tomogr* 6: 125–133

Clinical Application of Measurement of Regional Cerebral Blood Flow and Oxygen Utilization by Positron Emission Tomography

By T. Jones, R. Frackowiak, R. Wise, and G. L. Lenzi

In the clinical application of positron emission tomography to the study of regional cerebral function the Hammersmith programme has to date rested principally on the steady-state oxygen-15 inhalation method. This experience has not only produced new clinical scientific information in certain neurological conditions, but provides a basis upon which to extend our techniques for studying regional cerebral function. This paper discusses the methodological aspects of the techniques developed. Particular emphasis is placed on the degree of detail that is necessary to achieve quantitative measurements of cerebral blood flow and oxygen utilisation. Uncertainties, arising from practical theoretical and statistical sources of error, are also outlined. The procedures discussed have been in practice for over 2 years and the results that have emerged from our laboratory are itemised.

Methodology

The tracer concepts central to the measurement of tissue blood flow and oxygen utilisation rest on creating a "steady-state" tracer concentration within the tissues by the continuous inhalation of $C^{15}O_2$ and $^{15}O_2$. In the former case there is a rapid shift of ^{15}O from the carbon dioxide to vascular water under the influence of carbonic anhydrase in the lung. This results in a constant arterial supply of $H_2^{15}O$ to the peripheral tissues. Because of the 2.1-min half-life of ^{15}O , the tissue concentration of $H_2^{15}O$ does not reach that in the arterial blood. This follows because of the finite time that is required for the arterial water to exchange with that in the tissue. This imposed time delay is, for a freely diffusible tracer like water, principally dependent on the tissue's access, with respect to time, to the arterial blood, that is its rate of perfusion. It follows that the resulting steady-state concen-

tration of $H_2^{15}O$ within the tissue relative to that in the arterial blood is dependent on the tissue's blood flow. In effect the tissue is presented with a specific challenge, namely the radioactive half-life of the ^{15}O . The result of that challenge, the tissue's equilibrium concentration of $H_2^{15}O$, provides information on its flow. When molecular ^{15}O is continuously inhaled, the pulmonary capillary red cells are labelled and the tracer transported to the tissues by blood flow. In turn the aerobically respiring tissues extract a fraction of the molecular oxygen offered to them. This extracted oxygen is used to accept hydrogen from the cytochrome system to form water of metabolism. The steady-state tissue concentration of tracer during the continuous inhalation of ^{15}O is due principally to the formation in the tissue of ^{15}O -labelled water of metabolism. It follows that this will be dependent both on the tissue's blood flow and the fraction of oxygen extracted. The $C^{15}O_2$ and $^{15}O_2$ inhalation procedures offer sufficient information to calculate the tissue's blood flow in ml/ml of tissue per minute and oxygen utilisation in milliliters of O_2 consumed/ml of tissue per minute. The theoretical concepts behind these measurements have been discussed in earlier papers by Jones et al. (1976), Subramanyam et al. (1978) and Lenzi et al. (1978). However, at the time of these publications it was not possible to develop the methodology to its logical conclusion of achieving quantitative values. The limitations imposed were due solely to the inability to measure regional tissue concentrations of tracer in the same units as those for measuring arterial blood samples. The availability of positron emission tomography and the ability to quantitate regional tissue concentrations of tracers in absolute units advanced this tracer methodology to its logical conclusions. At Hammersmith the whole body positron emission tomograph ECAT II (EG & G Ortec) has been used for this purpose. It has been shown that the machine can, with the use

of corresponding ring source transmission data, provide absolute values of regional tissue tracer concentration. This is so within the limits of the machine's spatial resolution and the associated partial volume effect (Hoffman et al. 1979). The paper by Frackowiak et al. (1980) provides detailed descriptions of the theoretical and practical aspects of the developed PET methods for quantitating regional cerebral blood flow and oxygen utilisation. This has been followed by a further paper by Lammertsma et al. (1981a), who discuss sources of error inherent in the tracer theory used. In particular the presence of the unextracted ^{15}O , which remains upon the red cells, has been shown to result in an overestimation of the oxygen extraction ratio. A first order correction for this can be achieved by introducing an additional measurement of regional cerebral blood volume using ^{11}CO -labelled red cells. Lammertsma et al. (1981b, 1982) have also reported the statistical uncertainty that is introduced from the combined emission and transmission data into the value for rCBF and rCMRO₂. Rhodes et al. (1981) and Baron et al. (1981) have both tested the sensitivity of the methods by experimentally studying the reactivity of the cerebral circulation to pCO₂ change. It is necessary to state that the methodology has been evolving with experience over the 2–3 years that it has been used. The practical aspects of the technique have continued to be perfected. In the light of this experience we feel it important to itemise the details of the technique that are necessary to obtain quantitative values of flow and oxygen utilisation.

Achieving the Steady State

A constant supply of tracer is of course essential. To ensure this, both the flow rate of the gas and its concentration must be continuously monitored. An electromechanical servo machine is needed to adjust the concentration of the gas in response to fluctuations in production from the cyclotron. A light polythene (M.C.) face mask is used to administer the gas to the patient. Side holes in this mask provide the patient with room air while only some 500 ml/min of labelled gas is actually introduced into the mask. This arrangement makes

for a comfortable, physiological means for delivering the tracer. Approximately half the labelled gas is expelled from the mask as the patient exhales and is exhausted to waste. This arrangement has been much preferred to either mouth piece breathing or the use of an anaesthetic mask where inspired/expired valves have to be worked. Such resistance to respiration can result in minute to minute variations in the patient's respiratory pattern. Care in the design and use of such a breathing system is necessary to ensure there is no build-up of inspired CO₂ which can affect the respiratory rate and tidal volume. Before administering the tracer, it is important to settle the patient in position, wearing the face mask and the exhaust-hood system. This allows the patient to relax and adopt a normal breathing pattern.

We consider ring sources transmission scans essential for correcting the emission scans for tissue attenuation. This means of correction is preferred because of the tracer model's demand for accuracy when measuring the tissue tracer concentrations. Our earlier experience with the use of the ellipse correction technique (Frackowiak et al. 1980) and the theoretical study of Huang et al. (1981) supported this practice despite the fact that it imposes an additional source of statistical noise upon the data. These transmission measurements are carried out prior to the emission scans, thus allowing a further period of 15–20 min for the patient to settle physiologically.

For arterial sampling, cannulation of the radial artery is desirable. This is done before the patient is placed in the scanner and allows frequent blood samples to be drawn safely without disturbing the patient. These serve as a check on the stability of the steady state which must be assured before a given set of measurements can be considered acceptable for quantitative analysis. An efficient system for exhausting the expired gas to waste is imperative. A hood is placed over the patient's face and exhausted to ensure that labelled gas does not build up in or around the head region.

The design of the positron emission tomograph itself is important. It needs to be efficiently designed with respect to the slit shielding used so as to ensure minimum interference from non-cranial activity. The whole body ECAT II, with its 100-cm separation between the coincident

crystals, ensures that the data collected contains minimum noise from activity outside the transaxial plane being recorded.

Finally, movement of the patient's head must be minimised. This is especially important in those studies where transmission scans and $C^{15}O_2$ and ^{15}O emission scans must be congruent for quantitation. We have designed our own head restraint system, which meets the demands required of such measurements. In addition a grid light is shone onto the patient's forehead, onto which marks have been placed. This has proved to be most effective for identifying even the slightest head movement and for precise repositioning.

Blood Sampling

Although the measurement of the arterial blood concentration is straightforward, care must be taken with correcting for the time period over which the sample is withdrawn. The syringe must be thoroughly mixed before dispensing the blood for counting and centrifuging.

In addition the whole blood counting must be carried out with the blood haemolyzed, and care when centrifuging the blood samples is necessary so as to avoid haemolysis affecting the plasma counts. As an assessment of the stability of the steady state the blood samples taken from the 60 patients investigated at Hammersmith between November 1981 and April 1982 have been analysed. On average three separate arterial blood samples are withdrawn during each steady-state period, which typically extends for 15–20 min. The blood sample itself is withdrawn over a period of about 10 s, so averaging over the respiratory cycle. Some blood data has to be rejected because of obvious instability and in some cases a whole patient study cannot be quantitated. Such failures are however, comparatively rare. For the blood data analysed from the 60 patients the percentage standard deviation from the mean was found to be 4.0%, 4.2%, 4.3% and 5.0% for the $C^{15}O_2$ whole blood, $C^{15}O_2$ plasma, $^{15}O_2$ whole blood and $^{15}O_2$ plasma concentrations respectively. Considering the sources of variation, both physiological and physical, that

can be imposed on this data, the results are good and demonstrate that the steady state can be achieved in the vast majority of patients.

Clinical Scientific Information

Initially the ^{15}O steady-state technique was used to investigate a wide range of neurological disorders. With this experience it has become increasingly clear that in order to achieve results that are of value to clinical neurology, it is necessary to concentrate on studying comparatively few patients both quantitatively and serially. Furthermore, study of the many unsolved problems in common neurological and psychiatric conditions, such as stroke, dementia and schizophrenia involves relatively uncooperative patients. Such studies place considerable strain on the investigator who is seeking absolute quantitative data. Hence a highly professional and organised approach is essential. With this experience behind us, it is now possible to identify the new information that has emerged from our laboratory. This serves as the justification for continuing this expensive means for examining the human brain.

Dementia

A prospective study of 32 demented patients, with 20 followed up 6 months later, revealed that there was no evidence for focal or global ischaemia in this disease (Frackowiak et al. 1981). In addition a negative correlation between severity of the disease and rCMRO₂ and rCBF has been found. The patterns of the focal abnormalities were different for degenerative and multi-infarct dementia. These data provide a control platform upon which it will be possible to assess the value of pharmacological agents that are claimed to be beneficial for the treatment of this disease.

Epilepsy

In the interictal stage of non-psychotic unilateral temporal lobe epileptics, a 22% fall in rCBF was found in the affected area. This was

accompanied by a 12% fall in $rCMRO_2$. A more striking finding has been the reduction of flow and oxygen utilisation in the cerebellum compared with age-matched controls. This finding may either reflect an effect of the disease itself or be the result of chronic anticonvulsant medications. (This work has been carried out in conjunction with Dr. Michael Trimble of The National Hospital for Nervous Disease, Queen Square, London.)

Schizophrenia

In conjunction with Dr. Graham Shepherd of Charing Cross Hospital, freshly diagnosed, unmedicated schizophrenics have been studied. The distribution and values of $rCBF$ and $rCMRO_2$ within the cortex were found to be superimposable on those of age-matched controls (unpublished).

Glioma

It has been possible to study a series of gliomas, prior to surgical biopsy or removal, by collaborating with Mr. David Thomas of Queen Square. The results indicate that these neoplasms appear to be relatively overperfused with respect to their utilisation of oxygen. They consistently exhibit a lower oxygen extraction ratio than that of the surrounding normal brain tissue (Lammertsma et al. 1981c; Ito et al. 1982). Such a finding has implications with respect to the understanding of neoplastic hypoxia.

Stroke

There have been several publications reporting observations on changes in $rCBF$, $rCMRO_2$ and $rOER$ in stroke (Ackerman et al. 1981; Baron et al. 1981; Lenzi et al. 1981, 1982). The mismatch between $rCBF$ and $rCMRO_2$, reflected in $rOER$ and the phenomena of crossed cerebral and cerebellar diaschisis have been described.

The ability to define ischaemia in man is creating a major advance in stroke research. Once $rCMRO_2$ is limited by oxygen supply, and not by the energy demands of the neural cells, major changes occur in the electrical work, homeostatic mechanisms and structural integrity of neurones. The degree of disruption in normal cellular processes will depend, at least in part, on the severity and duration of the reduction in $rCBF$.

Measurements of $rCMRO_2$ and $rOER$ in man after the onset of acute stroke can be used to observe the transition from ischaemia (variable depression of $rCMRO_2$ and maximal $rOER$) to impaired mitochondrial function (low $rCMRO_2$ and low $rOER$) and hence, presumably, impending infarction. Ischaemia is potentially reversible and therefore clues as to how long it may persist after cerebral artery occlusion will have a major impact on the future design of medical or surgical intervention at intervals after the ictus.

We are extending the work already reported from many centres, including our own, in an attempt to answer the following questions:

1. Does regional ischaemia normally persist for at least some hours after the onset of stroke? Effective therapeutic intervention will never be possible if infarction is normally already established before medical attendants can reach the patient. The best way to answer this question is by quantitative serial scans on individual patients and the first study must be at least within a few hours of the onset of symptoms. Our observations (Wise et al., to be published) suggest that in middle cerebral artery occlusion cortical ischaemia may persist for many hours, or even days, but subcortical grey and white matter shows evidence of impending infarction (as evidenced by a low $rCMRO_2$ and $rOER$) in all our early studies (the earliest being 90 min after the ictus).
2. How common is either chronic ischaemia or a chronically diminished perfusion reserve (where $rCMRO_2$ is only maintained by means of an elevation of $rOER$ above normal) in patients with cerebrovascular disease? This has obvious implications as regards vascular bypass procedures and carotid reconstructive surgery. We have never observed chronic ischaemia in relation to arteriosclerotic arterial disease and a chronically diminished perfusion

reserve only very rarely. This is not the experience of groups in St. Louis and Paris where persistent elevation of rOER has been observed in a number of patients.

The major problems with PET research on stroke are neither related to technology nor tracer modelling. The opportunity to scan patients early rarely presents itself and this difficulty can only be solved by the siting of acute stroke units next door to PET scanning facilities. Even if an early study is obtained, the difficulty of interpreting the quantitative changes in subsequent studies can remain a major challenge. Extension of arterial thrombus, the appearance of vasogenic oedema, the infiltration of infarcted tissue by numerous phagocytic cells, etc. can all make simple interpretations of serial changes misleading – and each area of cerebral ischaemia/infarction will be unique in respect to the balance of these factors. It is salutary to realise that measurement of oxygen consumption by ischaemic cells does not necessarily reflect the generation of high-energy phosphate bonds – it has never been established that oxidative phosphorylation is coupled to ATP production under conditions of ischaemia and there are theoretical reasons why it may not.

Therefore future advances in stroke research using PET must take two forms. The first will relate to improvements in technology and the development of tracers that can be used to quantitate reliably changes in various physiological processes such as hydrogen ion homeostasis and breakdown of the blood-brain barrier. We have begun to do consecutive studies with ^{15}O and ^{18}F FDG to study anaerobic glycolysis in acute stroke but there are problems in interpreting the results of ^{18}F FDG uptake in abnormal brain. However, advances in PET scanners and applications of tracers must be coupled to the optimum timing of studies in appropriate patients. It has become clear that single scans on cerebral infarcts, days or even weeks old, merely show the physiological processes that remain amongst the graves of dead neurones.

rCBF, rCMRO₂ and rCMRGlu

Recently, it has been possible to introduce at the Hammersmith the ^{18}F -FDG technique to study regional cerebral glucose utilisation. This provides by sequential $^{15}\text{O}_2$, C^{15}O_2 and ^{18}F FDG studies the opportunity for identifying and measuring anaerobic glycolysis. Such comparisons are, however, extremely demanding if quantitative data is to be extracted from all the studies. The combined rCBF, rCMRO₂, rCMRGlu and rCBV may take up to 3 h to complete for each patient. To date the preferential utilisation of glucose has been seen in glioma relative to oxygen utilisation despite there being excessive flow relative to oxygen usage. These data are suggestive of the tumour's preference for anaerobic glycolysis and support the early work of Waarburg in the 1920s. In stroke a preferential uptake of ^{18}F FDG has also been seen relative to ^{15}O . These preliminary results are to be published in abstract form in the Proceedings of the 3rd World Congress of Nuclear Medicine, Paris 1982 – Rhodes et al. (1982). Such comparisons raise all sorts of questions – not least about the validity of the respective tracer models in disease – lumped constants, diffusibility of tracers, shunting of tracers – but the elucidation of these remains a task for the coming years.

Summary

A great deal has been written about the potential of positron emission tomography for studying regional cerebral function. However, to date comparatively few techniques have been developed that provide quantitative values. The ^{15}O technique represents one of these and provides for a measurement of energy balance within the brain. However, extreme methodological care needs to be taken in order to achieve accurate quantitative values of rCBF and rCMRO₂. The method is sufficiently well established to be providing reports in the open literature which contain new information in disease (Frackowiak and Lenzi 1982). A platform thus exists for extending the physiological investigation of cerebral disease both to the ob-

jective assessment of treatment and to help interpret results obtained with other substrate-tracer studies of the human brain.

References

- Ackerman RH, Correia JA, Alpert NM, Baron JC, Gouliamos A, Grotta JC, Brownell GL, Tavaras JM (1981) Positron imaging in ischemic stroke disease using compounds labelled with oxygen-15. *Arch Neurol* 38: 537–543
- Baron JC, Boussier MG, Comar D, Soussaline F, Castaigne P (1981) Noninvasive tomographic study of cerebral blood flow and oxygen-metabolism in vivo: potentials, limitations and clinical applications in cerebral ischemic disorders. *Eur J Neurol* 20: 273–284
- Frackowiak RSJ, Lenzi GL (1982) Physiological measurement with positron tomography: from potential to practice. In: Ell PJ, Holman L (eds) *Computerised emission tomography*. Oxford University Press, Oxford, chap 5 pp 188–210
- Frackowiak RSJ, Lenzi GL, Jones T, Heather JD (1980) Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man using ^{15}O and positron emission tomography: theory, procedure and normal values. *J Comput Assist Tomogr* 46: 727–736
- Frackowiak RSJ, Pozzilli C, Legg NJ, Du Boulay GH, Marshall J, Lenzi GL, Jones T (1981) Regional cerebral oxygen supply and utilisation in dementia – a clinical and physiological study with oxygen-15 and positron tomography. *Brain* 104: 753–778
- Hoffman EJ, Huang SC, Phelps ME (1979) Quantitation in positron emission computed tomography: effect of object size. *J Comput Assist Tomogr* 3: 299–308
- Huang SC, Hoffman EJ, Schelbert HR, Kuhl DE (1981) A boundary method for attenuation correction in positron computed tomography. *J Nucl Med* 22: 627–637
- Ito M, Lammertsma AA, Wise RJS, Bernardi S, Frackowiak RSJ, Heather JD, McKenzie CG, Thomas DGT, Jones T (1982) Quantitation of regional cerebral blood flow and oxygen utilisation in patients with brain tumours using oxygen-15 and positron emission tomography – analytical techniques and preliminary results. *J Neuroradiol* 23: 63–74
- Jones T, Chesler DA, Ter-Pogossian MM (1976) The continuous inhalation of oxygen-15 for assessing regional oxygen extraction in the brain of man. *Br J Radiol* 49: 339–343
- Lammertsma AA, Jones T, Frackowiak RSJ, Lenzi GL (1981a) A theoretical study of the steady-state model for measuring regional cerebral blood flow and oxygen utilisation using oxygen-15. *J Comput Assist Tomogr* 5: 544–550
- Lammertsma AA, Frackowiak RSJ, Lenzi GL, Heather JD, Pozzilli C, Jones T (1981b) Accuracy of the oxygen-15 steady-state technique for measuring rCBF and rCMRO₂: tracer modelling statistics and spatial sampling. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S 3–4
- Lammertsma AA, Ito M, McKenzie CG, Jones T, Frackowiak RSJ (1981c) Quantitative tomographic measurements of regional cerebral blood flow and oxygen utilisation in patients with brain tumours using oxygen-15 and positron emission tomography. *J Cereb Blood Flow Metabol* [Suppl] 1: S 567–568
- Lammertsma AA, Heather JD, Jones T, Frackowiak RSJ, Lenzi GL (1982) A statistical study of the steady-state technique for measuring regional cerebral blood flow and oxygen utilisation using oxygen-15. *J Comput Assist Tomogr* 6: 566–573
- Lenzi GL, Jones T, McKenzie CG, Buckingham PD, Clark JC, Moss S (1978) Study of regional cerebral metabolism and blood flow relationships in man using the method of continuously inhaling oxygen-15 and oxygen-15-labelled carbon dioxide. *J Neurol Neurosurg Psychiatry* 41: 1–10
- Lenzi GL, Frackowiak RSJ, Jones T (1981) Regional cerebral blood flow (CBF) oxygen utilisation (CMRO₂) and oxygen extraction ratio (OER) in acute hemispheric stroke. *J Cereb Blood Flow Metabol* 1: [Suppl 1] S 504–S 505
- Lenzi GL, Frackowiak RSJ, Jones T (1982) Cerebral oxygen metabolism and blood flow in human cerebral ischemic infarction. *J Cereb Blood Flow Metabol* 2: 321–335
- Rhodes CG, Lenzi GL, Frackowiak RSJ, Jones T, Pozzilli C (1981) Measurement of CBF and CMRO₂ using the continuous inhalation of C^{15}O_2 and $^{15}\text{O}_2$. *J Neurol Sci* 50: 381–389
- Rhodes CG, Wise RJS, Hatazawa J, Frackowiak RSJ, Palmer AJ, Jones T (1982) Mismatching between cerebral oxygen and glucose metabolism in patients with cerebral glioma and stroke. Proceedings of the 3rd world congress of nuclear medicine, Paris, August 1982. Pergamon Press II, New York
- Subramanyam R, Alpert NM, Hoop B et al. (1978) A model for regional cerebral oxygen distribution during continuous inhalation of $^{15}\text{O}_2$, and C^{15}O and C^{15}O_2 . *J Nucl Med* 19: 48–53
- Wise RJS, Bernardi S, Frackowiak RSJ, Legg NJ, Jones T (to be published) Serial observations on the pathophysiology of acute stroke: the transition from ischaemia to infarction as reflected in regional oxygen extraction. *Brain* 106 (I)

Positron Emission Tomography of Stroke Patients

By R. H. Ackerman, N. M. Alpert, S. M. Davis, R. E. Kelley, J. A. Correia, G. L. Brownell, and J. M. Taveras

Introduction

Although the application of PET in the study of stroke disease is still in its development, it is obvious that PET can have a major impact on the diagnosis, management, and therapy of stroke disease. Ischemic and hemorrhagic stroke are regional diseases, which cause focal changes readily resolved by current PET instrumentation. Among the most critical acute pathophysiological changes occurring in stroke are those to be found in blood flow and oxygen and glucose metabolism, all of which can be imaged with existing positron techniques. Exquisite spatial resolution, time of flight measurements, and the application of highly differentiated tracer techniques, such as those for demonstrating receptor binding, are not necessary to obtain relevant pathophysiological information.

Studies with PET have been performed on patients with stroke at several centers (Kuhl et al. 1980; Ackerman 1981; Ackerman et al. 1981a, b; Baron et al. 1981a, b; Lenzi et al. 1981). We began our studies of cerebral circulation and metabolism with positron-emitting agents in 1975, using two-dimensional imaging techniques. It rapidly became obvious that changes in "final common pathway" functions such as cerebral blood flow, oxygen metabolism, and glucose metabolism could be detected within physiological ranges in normal and diseased states (Ackerman et al. 1977, 1981a) and that changes in metabolic function might be predictive of tissue viability (Ackerman et al. 1978, 1981a). When we began routine transverse section imaging in 1978, PET rapidly provided evidence of focal pathophysiological mechanisms related to the hyperemias seen following ischemic insults (Ackerman 1981) and raised questions about the possible occurrence and meaning of focal alterations of CBF and/or metabolism adjacent to or at a distance from an area of

primary tissue injury (Ackerman et al. 1982; Davis et al. 1982).

Positron-imaging studies in human subjects is limited not only by hardware, radiopharmaceutical, and modeling considerations, but by the fact that no stroke is ever the same in two individuals, so that interpreting the results on small data samples raises statistical difficulties. We have employed two strategies for helping to overcome this difficulty: (1) using the subject as his own control by monitoring sequential changes in PET data over time and (2) documenting the relationship of the physiological changes to the clinical findings and clinical course. For this exercise we have coined the term "clinicophysiological correlations," which we feel are key to understanding the importance of PET in the investigation of ischemic and hemorrhagic stroke disease. This paper demonstrates examples of clinico-physiological correlations and summarizes the major findings of our studies in ischemic stroke.

Methods

Radiopharmaceuticals

Oxygen-15 labeled CO₂ and O₂ and fluorine-18 labeled 2-deoxyglucose (FDG) were used in these studies. ¹⁸F-2-deoxyglucose was obtained from Brookhaven National Lab. The ¹⁵O-labeled compounds were produced continuously by the MGH cyclotron, piped to the study area, and administered in trace amounts mixed in room air via a standard oxygen mask. The cyclotron output was monitored continuously and maintained constant to within $\pm 3\%$. Chemical purity of the ¹⁵O gases was monitored by gas chromatography. Expired gas and/or escaping radioactivity was removed by an exhaust system surrounding the face mask.

Subject Preparation

Subjects were studied in the supine position, with the head placed in the imaging field of the positron camera such that the OM line formed an angle of about 10° with the horizontal plane. In order to monitor for movement during the study head position was documented photographically and by alignment with low-power lasers.

In the ^{15}O studies an equilibration period of 8–10 min was used to ensure that a dynamic equilibrium was achieved. In the FDG studies imaging was begun immediately to help determine transport rates, but, data collection for clinical interpretation and quantitative measurements were typically obtained 35–80 min after injection.

In an attempt to produce a standard basal state we currently study subjects in a dimly lit room. Verbal interactions with the subject are kept to a minimum, but efforts are made to prevent the patient from sleeping.

Tissue Concentration

Transverse section data were obtained with the MGH positron camera PC-II, yielding tissue concentrations of $^{15}\text{O}-\text{H}_2\text{O}$ during continuous inhalation of $^{15}\text{O}-\text{CO}_2$ or $^{15}\text{O}-\text{O}_2$. The projection data recorded by PC-II were corrected to remove the effects of random coincidences, variations in detector sensitivity and photon absorption. Transverse section reconstructions were computed using a variation of the filtered back projection algorithm. The reconstruction resolution was varied in order to obtain a useful trade-off between resolution and statistical precision. For qualitative studies with ^{15}O the in-plane resolution was 1.5 cm FWHM; for quantitative measurement this was reduced to 2.0 cm FWHM. A plane thickness of 2.8 cm was used throughout. For studies with ^{18}F -FDG an in-plane resolution of 1.0–1.5 cm FWHM and a plane thickness of 1.4 cm was used.

Measurement of CBF, CMRO_2 , and CMRGI

The mathematical models of Jones et al. (1976) and Subramanyam et al. (1978) show that the tissue concentration measured during continuous inhalation of $^{15}\text{O}-\text{CO}_2$ depends on the decay constant of ^{15}O , the blood-tissue partition coefficient, the arterial $^{15}\text{O}-\text{H}_2\text{O}$ concentration, and the CBF. The local tissue concentration provides a nonlinear representation of rCBF. Similarly, mathematical modeling predicts that the tissue concentration measured during continuous inhalation of $^{15}\text{O}-\text{O}_2$ depends on the plasma concentration of $^{15}\text{O}-\text{H}_2\text{O}$, the concentration of ^{15}O -labeled oxyhemoglobin, the CBF, and the local oxygen extraction fraction.

Therefore we see that the ^{15}O concentrations measured during the $^{15}\text{O}-\text{CO}_2$ and $^{15}\text{O}_2-\text{O}_2$ studies may be used for qualitative and quantitative assessment of CBF and oxygen metabolism. When used for qualitative evaluation the procedure is essentially noninvasive. When used for quantitative evaluation arterial sampling is required.

During quantitative measurements a catheter was placed in the femoral artery. Repeated samples were drawn during the imaging period. Aliquots of whole blood and serum were placed in weighed vials and counted in a well scintillation counter to determine the concentration of $^{15}\text{O}-\text{H}_2\text{O}$ in whole blood, red cell fraction, and plasma as required.

Arterial plasma samples were also assayed with a well scintillation counter after the IV injection of ^{18}F -FDG to determine the time history for use in computation of metabolic rates (Phelps et al. 1979).

In order to provide visual as well as quantitative representations of CBF and CMRO_2 the transverse section measurements of tissue concentration were utilized along with the measurements of arterial concentrations in the equations of Subramanyam et al. (1978) to compute CBF, OEF, and CMRO_2 on a pixel by pixel basis.

Local glucose metabolism was computed using the Sokoloff model as modified by Phelps et al. (1979).

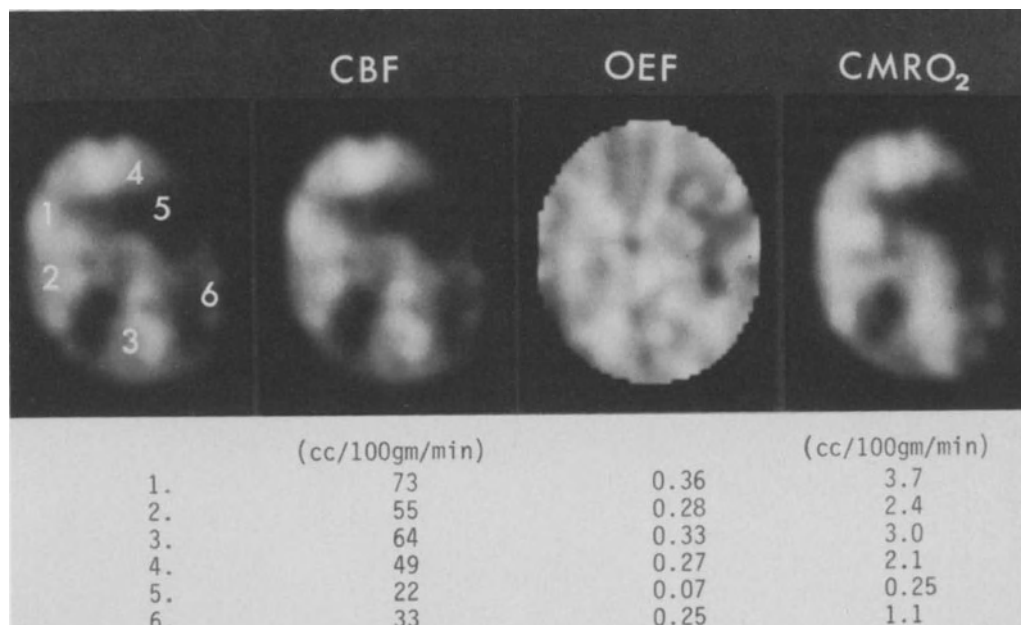


Fig. 1. Transverse section images of CBF and O₂ metabolism 24 h after RMCA artery embolus. The numerals on the *leftmost image* key the location of the

quantitative values of CBF, OEF, and CMRO₂ shown *below*

Illustrative Cases

In this section we present several cases illustrating recurring observations from our studies. The images portray the spatial distribution of physiological activity. Increasing physiological activity is indicated by increasing brightness. Images are oriented such that the slice is being viewed from above with the right on the reader's right.

Case 1. Fig. 1 shows the results obtained in a 55-year-old patient examined 24 h after suffering RMCA embolic infarction. CT scan obtained within hours of the PET study was essentially normal but a second CT scan obtained at day 32 showed a region of tissue loss which correlated exactly with the region of reduced OEF seen at 24 h. Quantitative values for CBF, OEF, and CMRO₂ are shown for selected regions as indicated by the numerals on the leftmost image. The quantitative values were more severely depressed deep in the MCA area than in cortical regions less susceptible to injury from a stem embolus when good collateral concentrations to cortical mantle is achieved.

Case 2. Serial studies of rCBF and rCMRO₂ were performed on an 87-year-old female with a left middle cerebral artery stroke on day 2, 16, and 30 following the ictus. In the images obtained during continuous inhalation of ¹⁵O—O₂ a severe depression of activity was observed in the area of infarction on all three studies. In contrast, the first ¹⁵O—CO₂ study demonstrated a depression of CBF values in the region of infarction, the second showed a patchwork of areas with both increased and decreased CBF, and the third ¹⁵O—CO₂ study again showed a more homogeneous depression of CBF. In the area of infarction CBF was more uniform on day 30 than on days 2 and 16, but higher on day 30 than on day 2.

Case 3. This 58-year-old man had two arteriographically proven emboli to the left middle cerebral artery, a week apart. The initial episode caused a mild dysphasia and mild right hemiparesis. CT scan on day 3 following the insult demonstrated low absorption values in the distribution of the penetrating branches of the left middle cerebral artery. On the 7th day following the initial insult the patient became

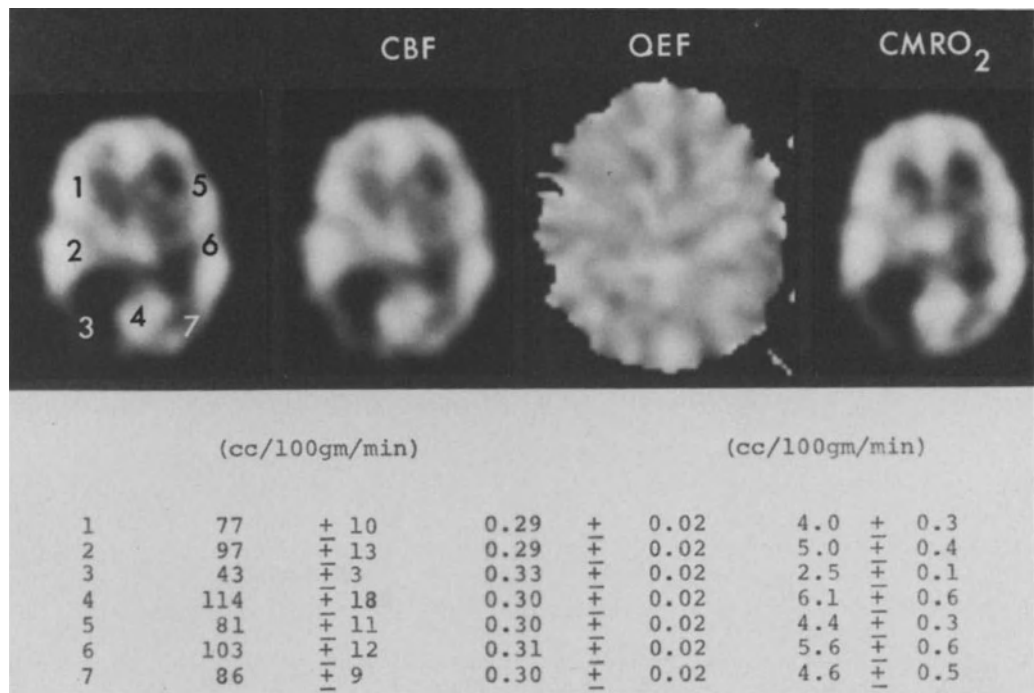


Fig. 2. Transverse section images of CBF and O₂ metabolism in a subject with a 1-year history of right homonymous hemianopsia and progressive myoclonic activity. See text for more detailed clinical

findings. The numerals on the *leftmost image* key the location of the quantitative values of CBF, OEF and CMRO₂ shown *below*

mute and developed a right hemiplegia. Clinically it was thought that the second embolus had infarcted the left cortical mantle, extending the damage done by the previous insult in sub-cortical gray structures. The positron study on the 2nd day after the initial embolic insult showed a hyperemia in the left basal ganglia region, which resolved by day 7 and recurred again at day 14. On day 7, following the second embolus the oxygen extraction fraction (OEF) was increased along the entire left cortical mantle. From our previous PET experience such an OEF finding would indicate that the cortical tissues were still viable, which would be contrary in this case to the clinical findings. A CT scan done 3 months postinfarction showed that the cortical tissue was intact.

Case 4. A 60-year-old man with right internal carotid artery occlusion was studied with ¹⁵O—CO₂ and ¹⁵O—O₂. Blood flow was depressed throughout the right anterior and middle cerebral artery territories, whereas oxygen

utilization was preserved and symmetric between the two hemispheres. As a result the oxygen extraction fraction was elevated in the distribution of the right internal carotid artery. The fixed deficit observed on initial examination cleared completely within 2–3 weeks.

Case 5. An 18-year-old boy was examined with ¹⁵O—CO₂, ¹⁵O—O₂, and ¹⁸F-FDG approximately 1 year after an acute onset of a right homonymous hemianopsia. The initial diagnosis was childhood stroke, but because of progressive myoclonic activity in his right extremities he was referred to the MGH for further evaluation. CT scan and arteriogram were negative. EEG studies showed intermittent spiking in the left parieto-occipital area. Figure 2 shows the results obtained with the ¹⁵O study. Blood flow, oxygen metabolism, and glucose metabolism were all markedly depressed in the left parieto-occipital region, but were otherwise normal. The positron study was the only study other than EEG to show definite abnormality.

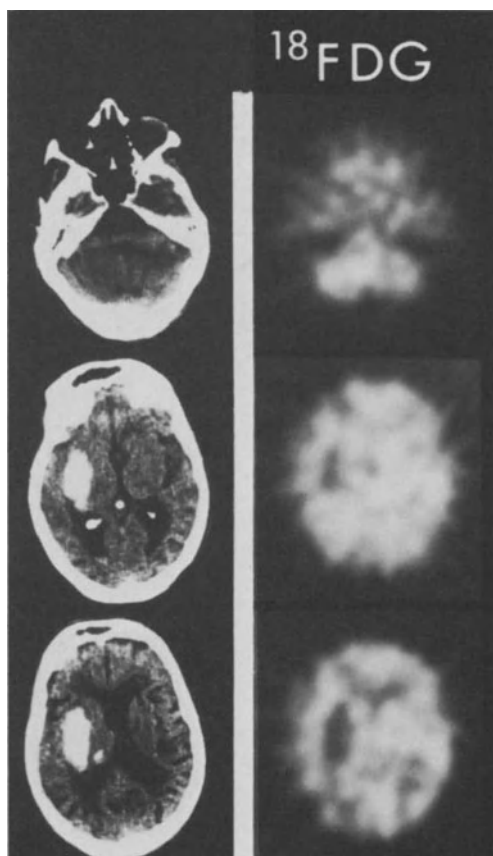


Fig. 3. X-ray CT sections (*left*) and PET sections of ^{18}F FDG concentrations in a subject with left putaminal hemorrhage. Note asymmetry of FDG concentration in cerebellar hemispheres

Case 6. Figure 3 shows the PET results obtained in a 78-year-old, right-handed woman using ^{18}F FDG. She was dysphasic, with a right hemiparesis and left gaze preference. Several months prior to admission her blood pressure was observed to be 160/100, but she was not under treatment for hypertension at the time of admission. A CT scan done at admission showed a focal left putaminal hemorrhage which did not extend into white matter structures. The ^{18}F FDG study obtained 2 weeks following the ictus demonstrated a well-defined area of depressed glucose metabolism congruent with the lesion seen on the CT scan. A lesser decrease in glucose metabolism is seen in adjacent cortical structures along with a marked asymmetry of metabolism in the cerebellar

hemispheres. The right cerebellar hemisphere had significantly lower glucose metabolism than the left.

Discussion and Conclusions

The first case demonstrates that abnormalities in blood flow and oxygen metabolism can be visualized and quantitatively measured during the acute phase of stroke injury. In our experience the finding of severely depressed oxygen extraction fraction always indicates infarcted tissue. Cases 2 and 3 demonstrate three important points: First, that in an evolving stroke blood flow is not necessarily depressed, but rather may be either increased or decreased. Serial observations in other patients with evolving stroke show that blood flow goes through a series of patchwork heterogeneous changes in areas where oxygen metabolism is uniformly depressed and tissue loss is eventually demonstrated on CT scan (Ackerman 1981). Second, our studies with ^{15}O show that hyperemia in an infarct is a biphasic phenomenon, occurring acutely and after 7–10 days. The subacute hyperemia becomes maximal at 10–20 days, which correlates temporally with the period of capillary hyperplasia identified pathologically. It would appear that the acute hyperemia represents the luxury perfusion syndrome described by Lassen. The acute hyperemia is physiologically mediated and occurs in intact tissue in which the regulating mechanisms are present but disordered. The delayed hyperemia is a different type of nonnutritional flow, occurring in tissue that is undergoing dissolution. Third, CBF findings in acute stroke are difficult to interpret without data on oxygen metabolism. Case 4 demonstrates a finding in which oxygen metabolism is relatively preserved despite diminished CBF which results in an increased OEF. In contrast to the finding of severely depressed O_2 metabolism and OEF, which indicates infarction, the demonstration in injured tissue of high oxygen metabolism relative to blood flow carries a good prognosis. In the stroke-prone patient such a situation indicates perfusion impairment. Identification of this problem might help select patients for revascularization procedures. Case 5 illustrates

how PET can be used in the differential diagnosis of the stroke syndrome. The findings of focally reduced blood flow, oxygen, and glucose metabolism with a normal CT scan 1 year after the onset of symptoms ruled out the previous diagnosis of childhood stroke. Moreover, given the clinical deficits the area of tissue injury would be unlikely to straddle two vascular territories – that supplied by the middle cerebral artery and that supplied by the posterior cerebral artery. Correlating the clinical findings and the results of the PET study we diagnosed a chronic inflammatory process or neoplasm. The finding of disturbed function in intact tissue in acute and chronic ischemic and hemorrhagic disease is one of our current areas of focus. Case 6 demonstrates that the crossed cerebellar diaschisis described by Baron et al. (1981a) in ischemic tissue can be seen subacutely in small, circumscribed hemorrhagic lesions with ^{18}F FDG.

Our studies in acute stroke have led us to suggest that indices of CBF and metabolism determined with PET can be used to stage the severity of tissue injury. Based on these studies we have identified three stages of tissue injury:

1. Hypoperfusion with normal metabolism
2. Hypoperfusion with reversibly impaired metabolism
3. Infarction.

We hypothesize that it may be possible to determine quantitative thresholds and ranges for stage 2 injury so that reliable identification of tissue which is at risk but still viable becomes possible. Once it is possible to reliably stage tissue injury it will become possible to monitor the effects of therapeutic intervention. One must realize, however, that this approach to quantitative staging of tissue injury will be difficult. High spatial resolution and precise measurement will be necessary. In addition, thresholds may be difficult to define with PET because of the size of brain structures and the varying admixtures of white and gray matter. Finally, we summarize the major conclusions arrived at by these clinicophysiological correlations as follows:

1. Positron imaging can detect physiological changes that anticipate rather than follow structural change.
2. Following a stroke, changes in cerebral blood flow evolve in a patchwork, hetero-

geneous fashion compared with the more uniform response of oxygen metabolism in the area of tissue injury.

3. CBF findings are difficult to interpret without data on oxygen metabolism.
4. Two phases of nonnutritional flow can be identified, that which occurs acutely and that which is prominent at 10–20 days. The first may best be characterized as the luxury perfusion syndrome of Lassen; the latter is related to the pathoanatomical evolution of the cerebral infarction.

References

1. Ackerman RH (1981) Positron imaging in stroke disease. In: Moossy J, Reinmuth OM (eds) Cerebrovascular diseases. 12th research (Princeton) conference. Raven, New York, pp 67–72
2. Ackerman RH, Subramanyam R, Alpert NM et al. (1977) The $\text{C}^{15}\text{O}/\text{H}_2\text{O}^{15}\text{O}$ static positron scintigram as a representation of blood flow. 2nd joint meeting on stroke and cerebral circulation, Miami, February, 1977. Abstract in Stroke 8: 10
3. Ackerman RH, Correia JA, Alpert NM et al. (1978) Positron imaging as a clinical tool for demonstrating disturbed cerebral circulation and metabolism. Symposium on positron emission tomography, Montreal, June, 1978. Abstract in J Comput Assist Tomog 2: 653
4. Ackerman RH, Subramanyam R, Correia JA et al. (1980) Positron imaging of cerebral blood flow during continuous inhalation of C^{15}O_2 . Stroke 11: 45–49
5. Ackerman RH, Correia JA, Alpert NM et al. (1981a) Positron imaging in ischemic stroke disease using compounds labeled with oxygen 15. Initial results of clinicophysiological correlations. Arch Neurol 38: 537–543
6. Ackerman RH, Alpert NM, Correia JA et al. (1981b) Importance of monitoring metabolic function in assessing the severity of a stroke insult (CBF: an epiphenomenon?). J Cereb Blood Flow Metabol 1 [Suppl 1]: 502–503
7. Ackerman RH, Alpert NM, Davis SM et al. (1982) The occurrence of hypermetabolism adjacent to subcortical artefacts and hemorrhages. Stroke 13: 118 (Abstract)
8. Baron JC, Bousser MG, Comar D et al. (1981a) Crossed cerebellar diaschisis: a remote functional depression secondary to supratentorial infarction of man. J Cereb Blood Flow Metabol 1 [Suppl 1]: 500–501
9. Baron JC, Bousser MG, Comar D et al. (1981b) Noninvasive tomographic study of cerebral blood flow and oxygen metabolism in vivo: potentials, limitations and clinical applications in

- cerebral ischemic disorders. *Eur Neurol* 20: 273–284
10. Davis SM, Ackerman RH, Correia JA et al. (1982) ^{133}Xe and positron emission tomography (PET) studies of brain physiology in chronic cerebral infarction. *Neurology* 32 (2): A195 (Abstract)
 11. Jones T, Chesler DA, Ter-Pogossian MM (1976) The continuous inhalation of oxygen-15 for assessing regional oxygen extraction in the brain of man. *Br J Radiol* 49: 339–343
 12. Kuhl DE, Phelps ME, Howell AP et al. (1980) Effects of stroke on local cerebral metabolism and perfusion: Mapping by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 47–69
 13. Lenzi GL, Frackowiak RSJ, Jones T (1981) Regional cerebral blood flow (CBF), oxygen utilisation (CMRO_2) and oxygen extraction ratio (OER) in acute hemispheric stroke. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 504–505
 14. Phelps ME, Huang C, Hoffman EJ et al. (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with ^{18}F -2-fluor-2-deoxy-D-glucose: validation of method. *Ann Neurol* 6: 371–388
 15. Subramanyam R, Alpert NM, Hoop B et al. (1978) A model for regional cerebral oxygen distribution during continuous inhalation of $^{15}\text{O}_2$, C^{15}O_2 . *J Nucl Med* 19: 48–53

Local Cerebral Blood Flow and Oxygen Consumption in Evolving Irreversible Ischemic Infarction

By J. C. Baron, D. Rougemont, P. Lebrun-Grandié, M. G. Bousser, E. Cabanis, J. Bories, D. Comar, and P. Castaigne

Positron emission tomography (PET) imaging of the local interrelationship of cerebral blood flow and metabolism in completed ischemic stroke has attracted considerable interest because of its potential to differentiate irreversibly damaged tissue from viable tissue on the basis of the CBF-metabolism patterns. Several qualitative (Ackerman et al. 1981a, b) or semi-quantitative (Baron et al. 1979, 1981a, Kuhl et al. 1980) pioneering studies provided a limited insight into this question, while the single truly quantitative study was only briefly reported (Lenzi et al. 1981a, b). We report here a detailed study of the quantitative patterns of local cerebral blood flow (ICBF) and oxygen consumption (ICMRO₂) in irreversibly infarcted brain regions.

Patients and Methods

Thirty-five quantitative studies of ICBF and ICMRO₂ were performed in 25 patients, ten of whom were studied twice. All studies were performed between the 2nd and 38th days after clinical onset of completed ischemic stroke in the internal carotid artery territory. There were 13 females and 12 males, their age ranging from 38 to 80 years (mean 63 years).

We used the ¹⁵O continuous inhalation technique (Jones et al. 1976) coupled with PET (Baron et al. 1978; Frackowiak et al. 1980). Three adjacent brain levels were scanned with the ECAT II device (slice thickness 19 mm, lateral resolution ~16 mm). At equilibrium, a femoral arterial puncture allowed measurement of arterial blood ¹⁵O concentrations, PaCO₂, PaO₂, pH, and arterial oxygen content (Ca). For each brain level studied, the C¹⁵O₂ and ratio (¹⁵O₂/C¹⁵O₂) images were transformed pixel by pixel into CBF and oxygen extraction fraction (OEF) images respectively (Subramanyam et al. 1978). A CMRO₂ image

was then generated ($CMRO_2 = CBF \cdot OEF \cdot Ca$).

The location of irreversible infarction was assessed in each case by means of repeated CT scans (24 patients) or postmortem (three patients). On the CT scans persistent hypodensity or marked contrast enhancement was taken as evidence of irreversible necrosis. Then circular regions of interest (ROI) were located in approximately the same brain areas on the CMRO₂ images and the mean ROI values for CBF, OEF, and CMRO₂, as well as the corresponding values from the contralateral homologous ROI, were automatically calculated. To allow comparison with control data, 4-cm² ROIs were used; however, in ten patients (12 studies) with CT lesions less than ~3–4 cm in diameter, 1-cm² ROIs were used to minimize the partial volume effects (Soussaline et al. 1982).

Depending on the size of the actual necrosis, from 1 to 12 ROIs were available for a given patient. If the values for neighboring ROIs on the same brain level were similar, they were averaged. Eighty-nine infarct values (up to six per patient) were finally obtained. Each infarct value was also expressed as a percentage of its contralateral homologous value. Both the absolute and the percentage values were plotted against time from onset for all patients. They were further subdivided according to the pattern of CBF-CMRO₂ relationship, as determined by the percent IOEF value, taking 12% difference as significant ($p < 0.05$ from 19 control studies) (Lebrun et al., to be published). Local "luxury-perfusion syndrome" (Lassen 1966) or "misery-perfusion syndrome" (Baron et al. 1979, 1981b), were defined if infarct IOEF was significantly low or high, respectively, and "matched" CBF-CMRO₂ otherwise. Comparison of means was performed between the above three subgroups, as well as between "early" (≤ 4 days after clinical onset, 11 studies) and "late" (≥ 5 days, 24 studies) infarcts.

Results

The individual values of ICBF, IOEF and ICMRO₂ in the areas of irreversible infarction and their evolution with time appear clearly in Figs. 1 (absolute data) and 2 (percentage data). Early ICBF was extremely variable, ranging from 5% to 125% of contralateral values, but its trend to increase during the 2nd week and to remain somewhat elevated until the 40th day was apparent and highly significant (Table 1). Early IOEF ranged from low to high values (43%–184%), but was almost universally decreased later on, again a highly significant trend (Table 1). The ICMRO₂, however, was consistently depressed (except for a few data points), showing only a barely significant (absolute and relative) increase with time (Table 1); it ranged from 7% to 84% of contralateral CMRO₂. Regarding CBF-CMRO₂ relationship, a mismatch between local demand and supply of

oxygen within the area of irreversible infarction was seen in 91% of the studies. Misery-perfusion was almost confined to the first 4 days (five of six studied), but in four early instances was associated with areas displaying “matched” or luxury-perfusion syndromes. Areas displaying luxury-perfusion (more often relative than absolute in terms of ICBF) were seen in seven of eleven early studies and in 20 of 24 late studies. Each of the seven early misery-perfusion areas restudied were seen to progress to luxury-perfusion at the 2nd week after onset (Fig. 1 and 2). The absolute and relative ICBF, IOEF, and ICMRO₂ values according to the misery-perfusion, matched, or luxury-perfusion subgroups can be seen in Figs. 1 and 2, and their means are shown in Table 2. Highly significant differences between groups were found for mean ICBF and IOEF, but not for ICMRO₂. Thus ICBF increased steadily from misery-perfusion to matched to luxury-perfusion subgroups (while IOEF decreased in

Table 1. Mean ICBF, IOEF, and ICMRO in early and late infarcts

Day	n	CBF		OEF		CMRO ₂	
		Absolute (ml/100 g/ min)	%	Absolute	%	Absolute (ml/100 g/ min)	%
≤ 4	39	14.2 ± 8.4***	56 ± 29***	0.50 ± 0.19***	94 ± 36***	1.20 ± 0.54*	45 ± 18*
≥ 5	50	23.4 ± 9.5	82 ± 24	0.35 ± 0.16	68 ± 21	1.48 ± 0.71	52 ± 16

*p < 0.05; ***p < 0.001.

Table 2. Mean ICBF, IOEF and ICMRO₂ in the three subgroups and on the contralateral side

Pattern	n	CBF		OEF		CMRO ₂	
		Absolute (ml/100 g/min)	%	Absolute	%	Absolute (ml/100 g/ min)	%
Misery-perfusion	15	7.7 ± 5.2++	34 ± 17+	0.72 ± 0.14+++	135 ± 20+++	1.01 ± 0.55	45 ± 22
Matched	8	14.3 ± 3.9×××	49 ± 15×××	0.51 ± 0.08×××	100 ± 4×××	1.30 ± 0.51	48 ± 14
Luxury-perfusion	66	22.6 ± 9.2***	81 ± 25***	0.34 ± 0.12***	64 ± 15***	1.44 ± 0.67*	50 ± 16
Control side	89	27.8 ± 9.0	100	0.52 ± 0.12	100	2.76 ± 0.86	100
Misery-perfusion vs matched		+	p < 0.05				
		++	p < 0.01				
		+++	p < 0.001				
Matched vs luxury-perfusion		×××	p < 0.001				
Misery-perfusion vs luxury-perfusion		*	p < 0.05				
		***	p < 0.001				

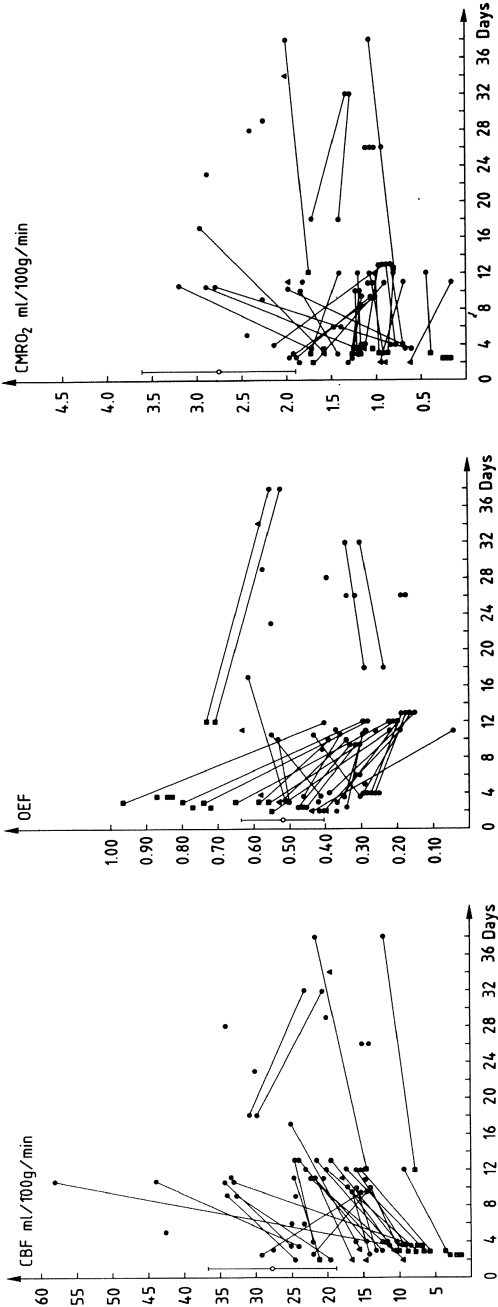


Fig.1

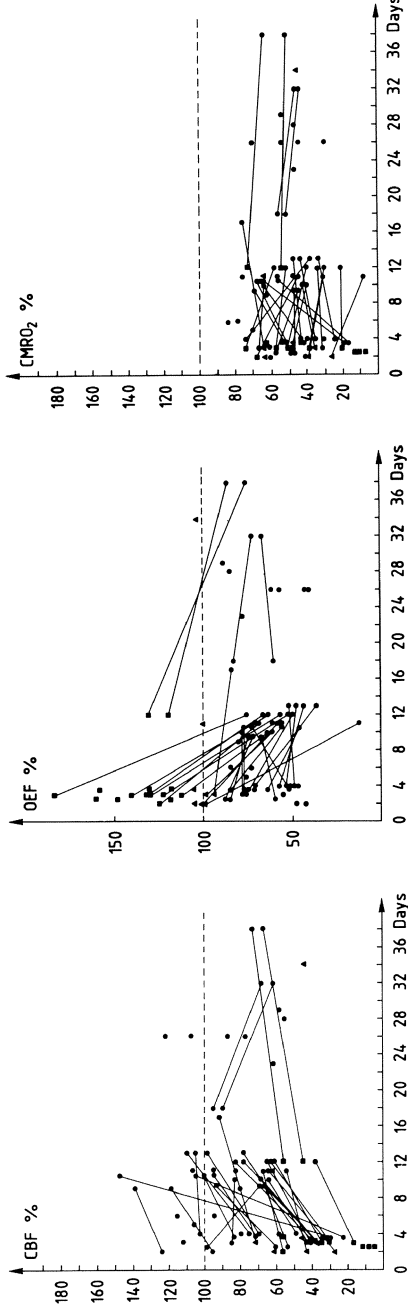


Fig.2

a mirror fashion) with a value of 14.3 ml/100 g per minute (49%) matching the local metabolic demand.

Comments

This detailed study provides estimates of ICMRO₂ and its relationship with ICBF within the area spontaneously evolving to ischemic necrosis in man.

Several potential limitations of the study deserve mention. The topographical assessment of the irreversibly infarcted area was relatively gross; although care was taken not to include the borders of infarction, ultimately preserved tissue may have been present in the ROI chosen as a result of the partial volume effect and the irregular, patchy infarction in some patients. Motion artifacts, irregular respirations, and inherent sensitivity of the C¹⁵O₂ model at high flows may have affected the measurement accuracy. In addition, the ¹⁵O model tends to underestimate CBF and overestimate OEF in normal situations (Baron et al. 1981c; Lammertsma et al. 1981), but alterations in local water content (e.g., edema) and blood volume likely to occur in stroke may have introduced an added uncertainty. Capillary dilatation, be it in low or high flow situations, is particularly apt to induce further overestimation of OEF, perhaps an explanation to some relatively high ICMRO₂ values found here. However, this factor alone could not explain most of the exceedingly high IOEF values in this study, i.e., the observation of misery-perfusion is not artifactual. Although real, the above limitations do not appear serious enough to question the overall validity of the present findings.

The most striking feature is the inconsistency of both ICBF and IOEF in early infarcts, both ranging from extremely low to extremely high values, whereas ICMRO₂ essentially remains in

low ranges. Thus neither ICBF nor IOEF separately can be taken as a good indicator of spontaneously evolving irreversible infarction when studied 2 days or more after onset. In contrast, a clearly depressed ICMRO₂ appears reliable in this respect, as suggested previously (Ackerman et al. 1981b; Baron et al. 1981a; Lenzi et al. 1981a, b). Given the above-mentioned methodological uncertainties, a tentative ICMRO₂ threshold of about 70% of contralateral value would seem to indicate evolving necrosis (Fig. 2). A theoretical CMRO₂ threshold for infarction of about 55%–65% of normal can be calculated from the experimental infarction CBF threshold (permanent arterial occlusion) of 18 ml/100 g per minute (Jones et al. 1981) and the increased OEF to about 0.70 found experimentally at similar levels of ischemia (Bruce et al, 1972). Our overall infarct ICMRO₂ was 1.35 ml/100 g per min $\pm$ 0.65 which was 49% of contralateral CMRO₂.

The spontaneous evolution was remarkable by the general trend of increasing ICBF and decreasing IOEF values, so that the vast majority of late infarcts displayed a clear-cut late luxury-perfusion, confirming earlier semiquantitative observations (Ackerman et al. 1981a; Baron et al. 1979, 1981a; Kuhl et al. 1980). This increase of ICBF which occurs in the face of an unchanged metabolic demand is a transitory phenomenon (Baron et al. 1979, 1981a; Lenzi et al. 1981a), probably directly related to the phagocytic phase of ischemic necrosis per se.

In contrast, there was no consistent pattern of CBF-CMRO₂ relationship in early infarcts. Early luxury-perfusion was frequently observed, indicating spontaneous partial reperfusion in irreversibly damaged tissue (Paulson 1971) as a result of either postembolic recanalization or efficient perfusion pressure from collateral channels. In many early instances, however, misery-perfusion was prominent, suggesting that the oxygen demand, although very depressed, was not quite matched by its supply.

Fig. 1. Absolute values of ICBF, IOEF, and ICMRO₂ plotted against time. Each point represents a single area in one patient (see text). The lines join data obtained from the same area on two separate occasions (ten patients were studied twice). *Squares, triangles,*

and dots represent areas with misery-perfusion, matched CBF-CMRO₂, and luxury-perfusion respectively. The *open dot* on the left of each graph represents the mean and standard deviation of the contralateral homologous areas

Fig. 2. Relative percentage values of ICBF, IOEF, and ICMRO₂ plotted against time (see legend to Fig. 1)

This continuing ischemia presumably results from persistent proximal occlusion with poor collaterals, or alternatively from the "no-reflow" phenomenon (Ames et al. 1968). Delayed hypoperfusion with relatively maintained energy metabolism has been reported in experimental ischemia reperfusion as a potentially damaging process (Levy et al. 1979; Snyder et al. 1975). Despite maximally increased OEF values (Bruce et al. 1972), the levels of ICBF observed in areas displaying misery-perfusion were indeed so low (Table 2) as to seem incompatible with tissue survival (Astrup et al. 1981; Jones et al. 1981). Although these ischemic areas did spontaneously evolve to luxury-perfusion and necrosis, it remains unknown whether induced recoupling could have partly salvaged the tissue. That mean ICMRO₂ was moderately (although just significantly) higher in areas where ICBF matched exactly or exceeded ICMRO₂ (Table 2) suggests that metabolism might benefit from increasing the tissue local oxygen tension when misery-perfusion is demonstrated.

The data in Table 2 suggest that the CBF threshold for relative ischemia (below which CBF becomes inadequate to meet the lowered oxygen demand of severely ischemic tissue) stands at about 14 ml/100 g per minute, perhaps not fortuitously close to the infarction threshold found acutely in experimental ischemia (Astrup et al. 1981; Jones et al. 1981). Clearly, additional information will come from more acute studies, as well as from concomitant measurement of the local glucose utilization (Baron et al. 1982). It will also be important to analyze in detail the ICBF-ICMRO₂ quantitative patterns in viable, dysfunctioning, but ultimately preserved areas surrounding the ultimately infarcted focus.

References

- Ackerman RH, Alpert NM, Correia JA, Finklestein S, Buanonno F, Davis SM, Chang JY, Brownell GL, Taveras JM (1981a) Importance of monitoring metabolic function in assessing the severity of a stroke insult (CBF: an epiphenomenon). *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 502–503
- Ackerman RH, Correia JA, Alpert NM et al. (1981b) Positron imaging in ischemic stroke disease using compounds labeled with oxygen 15. *Arch Neurol* 38: 537–543
- Ames A, III, Wright RL, Kowada M et al. (1968) Cerebral ischemia II. The no-reflow phenomenon. *Am J Pathol* 52: 437–453
- Astrup J, Siesjö BK, Symon L (1981) Thresholds in cerebral ischemia. The ischemic penumbra. *Stroke* 12: 723–725
- Baron JC, Comar D, Boussier MG, Soussaline F, Crouzel C, Plummer D, Kellershohn C, Castaigne P (1978) Etude tomographique chez l'homme, du débit sanguin et de la consommation d'oxygène du cerveau par inhalation continue d'oxygène 15. *Revue Neurol* 134: 545–556
- Baron JC, Comar D, Boussier MG, Plummer D, Loc'h C, Kellershohn C, Castaigne P (1979) Patterns of CBF and oxygen extraction fraction (EO₂) in hemispheric infarcts: a tomographic study with the ¹⁵O inhalation technique. *Acta Neurol Scand* 60 [Suppl 72]: 324–325
- Baron JC, Boussier MG, Comar D, Soussaline F, Castaigne P (1981a) Non-invasive tomographic study of cerebral blood flow and oxygen metabolism in vivo: potentials, limitations and clinical applications in cerebral ischemic disorders. *Eur Neurol* 20: 273–284
- Baron JC, Boussier MG, Rey A, Guillard A, Comar D, Castaigne P (1981b) Reversal of focal "misery-perfusion syndrome" by extra-intracranial arterial by-pass in hemodynamic cerebral ischemia: a case study with ¹⁵O positron tomography. *Stroke* 12: 454–459
- Baron JC, Steinling M, Tanaka T, Cavalheiro E, Soussaline F, Collard P (1981c) Quantitative measurement of CBF, oxygen extraction fraction (OEF) and CMRO₂ with the ¹⁵O continuous inhalation technique and positron emission tomography (PET): experimental evidence and normal values in man. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 5–6
- Baron JC, Lebrun-Grandié P, Collard P et al. (1982) Non-invasive measurement of blood flow, oxygen consumption and glucose utilization in the same brain locus in man by positron emission tomography. *J Nucl Med* 23: 391–399
- Bruce DA, Schutz H, Vapalahti M, Gunby N, Langfitt TW (1972) Interactions between cerebral blood flow and cerebral metabolism. *Neurol Surg* 23: 417–419
- Frackowiak RSJ, Lenzi GL, Jones T, Heather JD (1980) Quantitative measurement of cerebral blood flow and oxygen metabolism in man using ¹⁵O and positron emission tomography: theory, procedure and normal values. *J Comput Assist Tomog* 4: 339–343
- Jones T, Chesler DA, Ter Pogossian MM (1976) The continuous inhalation of oxygen 15 for assessing regional oxygen extraction in the brain of man. *Br J Radiol* 49: 339–343
- Jones TH, Morawetz RB, Crowell RM et al.

- (1981) Thresholds of focal cerebral ischemia in awake monkeys. *J Neurosurg* 54: 773–782
15. Kuhl DE, Phelps ME, Kowell AP, Metter EJ, Selin C, Winter J (1980) Effects of stroke on local cerebral metabolism and perfusion mapping by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 47–60
16. Lammertsma AA, Jones T, Frackowiak RSJ, Lenzi GL (1981) Theoretical study of the steady-state model for measuring regional cerebral blood flow and oxygen utilisation using oxygen 15. *J Comput Assist Tomog* 5: 544–550
17. Lassen NA (1966) The luxury perfusion syndrome and its possible relation to acute metabolic acidosis localised within the brain. *Lancet* II: 1113–1115
18. Lebrun-Grandié P, Baron JC, Soussaline F, Loc'h C, Sastre J, Bousser MG (to be published) Regional blood flow, oxygen utilization and the coupling between the two in the normal brain: a study with positron tomography and oxygen 15. *Arch Neurol*
19. Lenzi GL, Frackowiak RSJ, Jones T, Heather JD, Lammertsma AA, Rhodes CG, Pozzilli C (1981a) CMRO₂ and CBF by the oxygen 15 inhalation technique. *Eur Neurol* 20: 285–290
20. Lenzi GL, Frackowiak RSJ, Jones T (1981b) Regional cerebral blood flow, oxygen utilisation and oxygen extraction ratio in acute hemispheric stroke. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 504–505
21. Levy DE, Van Uitert RL, Pike CL (1979) Delayed postischemic hypoperfusion: a potentially damaging consequence of stroke. *Neurology* 29: 1245–1252
22. Paulson DB (1971) Cerebral apoplexy (stroke): pathogenesis, pathophysiology and therapy as illustrated by regional blood flow measurements in the brain. *Stroke* 2: 327–360
23. Snyder JV, Nemoto EM, Carroll RG, Safar P (1975) Global ischemia in dogs: intracranial pressures, brain blood flow and metabolism. *Stroke* 6: 21–27
24. Soussaline F, Baron JC, Lebrun P, Collard P, Comar D (1982) Some physical factors affecting quantitative precision in positron emission tomography brain studies. IV Congress of the World Federation of Nuclear Medicine and Biology, Paris, Aug 29–Sept 5
25. Subramanyam R, Alpert NM, Hoop B, Brownell GL, Taveras JM (1978) A model of regional cerebral oxygen distribution during continuous inhalation of $^{15}\text{O}_2$, C^{15}O_2 and C^{15}O . *J Nucl Med* 19: 48–53

Glucose Metabolism

(Chairman: A. P. Wolf and
D. E. Kuhl)

Mapping Local Cerebral Glucose Utilization in Normal Aging and in Cerebrovascular, Degenerative, and Epileptic Disorders

By D. E. Kuhl

This chapter reviews briefly what we have learned recently at UCLA in applying the ^{18}F -fluorodeoxyglucose (FDG) method to the normal aging brain and to cerebral dysfunction caused by cerebrovascular, degenerative, and epileptic disorders.

The FDG method for determining local cerebral glucose utilization (LCMR_{glc}) in humans (Reivich et al. 1979) is analogous to the ^{14}C -deoxyglucose autoradiographic method (Sokoloff et al. 1977) used in animals, but positron emission computed tomography (PECT) is substituted for the autoradiograph. After intravenous injection, FDG enters the brain, is phosphorylated by brain hexokinase, and the metabolic product, FDG-6-PO_4 , remains fixed with only slow dephosphorylation. The time course of ^{18}F activity and glucose concentration is measured in plasma, brain activity is determined by scanning, and with knowledge of predetermined rate constants, an operational equation allows calculations of LCMR_{glc} distributed as a cross-section picture (Phelps et al. 1979; Huang et al. 1980). The ECAT II tomograph (ORTEC Inc, Oak Ridge, Tennessee (Phelps et al. 1978) was used for the scans presented in Figs. 1–3, 7 and 8 (spatial resolution 16 mm) and the Neuro ECAT tomograph (ORTEC) (Williams et al. 1981) was used for Figs. 4–7 (spatial resolution 12 mm).

Aging

The FDG scan method was used to determine patterns of LCMR_{glc} in 40 normal volunteer subjects aged 18–78 Years (Kuhl et al. 1982b). Throughout the studies, every subject was quiet and motionless with eyes open and ears unplugged, exposed only to ambient room light and sound. For the entire group, the whole brain mean CMR_{glc} ($\pm\text{SD}$) was $26.1 \pm 6.1 \mu\text{mol } 100 \text{ gm}^{-1} \text{ min}^{-1}$. At age 78 years, mean

CMR_{glc} was on average 26% less than at age 18 years, a difference of the same order as the variance among subjects at any age. The gradual decline of mean CMR_{glc} with advancing age was at a rate faster than reported for mean cerebral oxygen utilization (Lammertsma et al. 1981), possibly due to increasingly altered pathways for glucose utilization or to increasing oxidation of ketone bodies or other alternative substrates (Gottstein and Held 1979).

In individuals of all ages, glucose utilization was equal in overall cortex, caudate, and thalamus. There was no selective decline in glucose utilization for any of these structures, although evidence of age-dependent selective degeneration has been reported (Smith et al. 1980). If degenerative processes during human aging cause relative differences in function or metabolism between cortical and subcortical structures, we were unable to demonstrate it by the FDG scan.

We found metabolic symmetry in the left and right hemispheres of these normal subjects, all of whom were studied in the awake but quiet state with eyes and ears open to ambient light and sound. Under other conditions of sensory stimulation or deprivation, marked asymmetries can appear in normal subjects (Mazziotta et al. 1982b, c; Phelps et al. 1981). The symmetry of the FDG scan in the normal control state was in contrast to the asymmetrical distributions of cortical and subcortical metabolism found in patients with multiple infarct dementia (e.g., in Fig. 5). Unlike the marked symmetrical hypometabolism of frontal and parietal association cortex found in FDG scans of patients with Alzheimer's disease (AD) (Benson et al. 1981), the zonal patterns of cortical metabolism of our normal subjects were more uniform and very similar in older and younger persons (Fig. 1), except for an age-dependent decline in the metabolic ratio of superior frontal cortex to superior parietal cortex (Fig. 2). This decline in metabolic hyperfrontality may

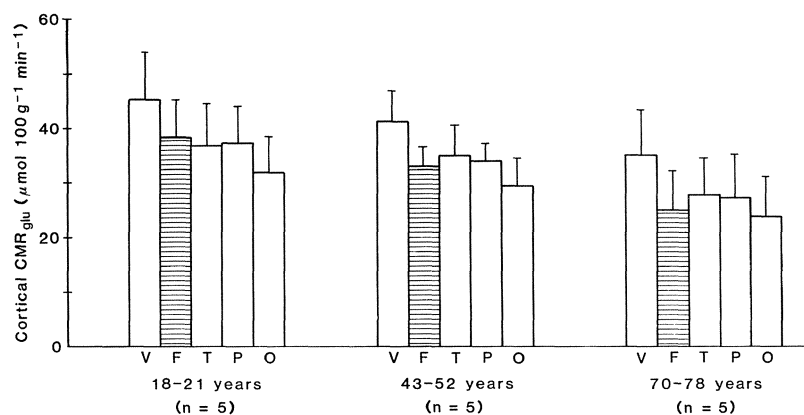
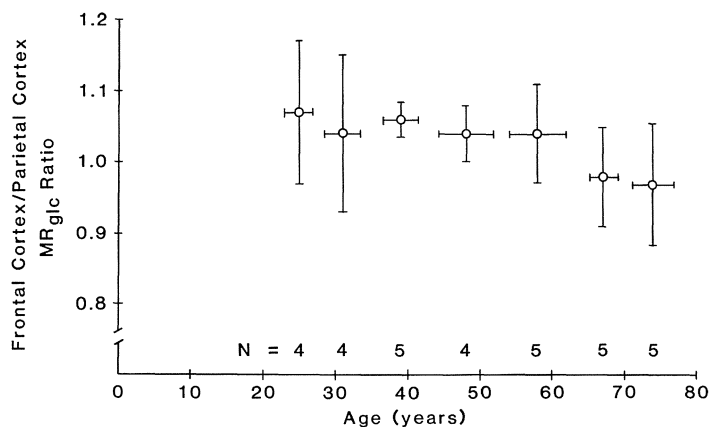


Fig. 1. When glucose utilization was averaged over large cortical zones in normal subjects, decline with age in frontal cortex metabolism (shaded) was not significantly different from decline in other cortical

zones. Cortical zones: *V*, primary visual; *F*, frontal; *T*, temporal; *P*, parietal; *O*, occipital. Error bars represent 1 SD. (From Kuhl et al. 1982b)

Fig. 2. The degree of metabolic hyperfrontality varies considerably among normal subjects, but on average declines gradually with increasing age. Each data point represents the average of measurements from *n* normal subjects, with 1-SD error bars. (From Kuhl et al. 1982b)



be associated with selective degeneration of the superior frontal cortex of normal subjects (Brody 1976), but may also only reflect a progressive change with age in the level of anxiety or cognitive response during the FDG study (Ingvar 1979). Results such as these should be useful in distinguishing age from disease effects when the FDG scan method is applied.

Stroke

Recent applications of PET in quantitative studies of cerebrovascular disease have confirmed expectations that local values of cerebral blood flow (LCBF), extraction of oxygen,

and utilization of oxygen (LCMR_{O_2}) and glucose have a varied and complex relationship in early stroke (Ackerman et al. 1981; Baron et al. 1981; Kuhl et al. 1980a; Heiss et al. 1981), in contrast to the more coupled and predictable relationships found in later stable or chronic states of cerebrovascular disease (Frackowiak et al. 1981). It is now recognized that these measures can give far more insight into local cerebral dysfunction after stroke than a structural assessment alone.

When LCMR_{glc} was determined by FDG scan and relative perfusion by $^{13}\text{NH}_3$ scan in stroke patients, we found the varied metabolism-flow relationships shown in Fig. 3 (Kuhl et al. 1980a). During the first 48 h after ictus, LCMR_{glc} is less depressed than LCBF at the in-

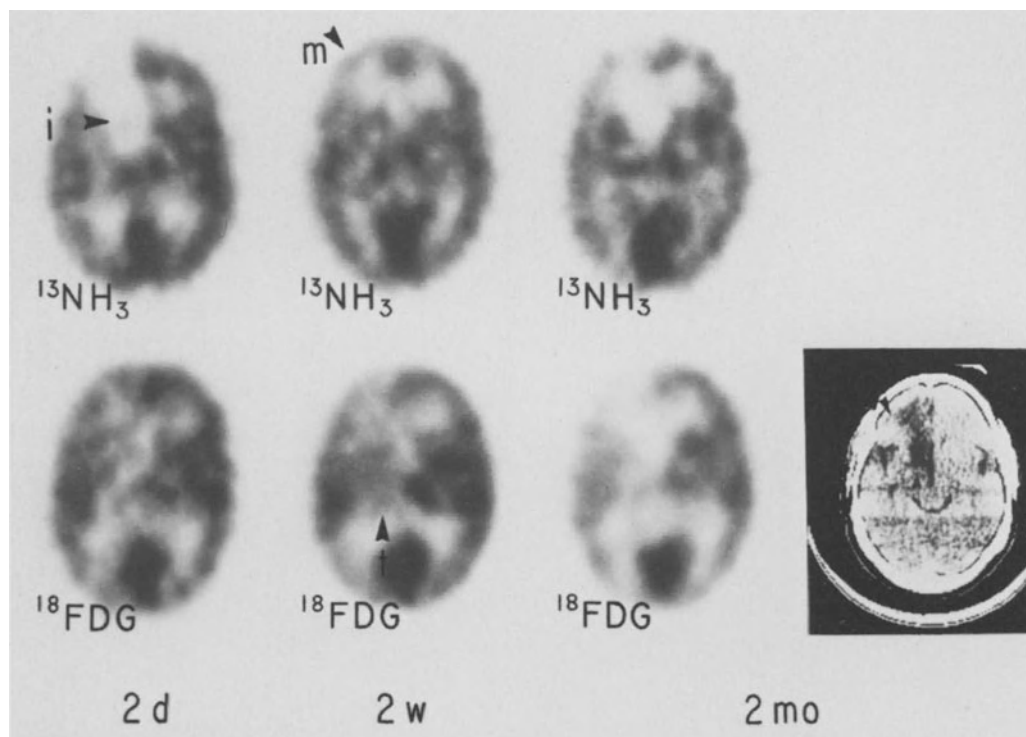


Fig. 3. Serial $^{13}\text{NH}_3$ (relative perfusion) and FDG (glucose utilization) scans of a 22-year-old man after sudden transection of the left internal carotid artery. Within the left frontal lobe infarct, perfusion was decreased more than glucose utilization (anaerobic glycolysis) in the earliest scan, but in both there was a similar decrease in later scans. Distant from the local

infarct, ipsilateral cortex and thalamus were hypometabolic. Ipsilateral primary visual cortex was hypometabolic and associated with a contralateral homonymous hemianopia. All of these distant structures appeared normal on CT scan. (From Kuhl et al., 1980a)

farct site, probably because of transient local anaerobic glycolysis. During the next 2 weeks, coexisting hyperemia and hypometabolism appear at the lesion margins. The maximum depression of LCMR_{glc} in the infarct occurs many weeks after the cerebrovascular occlusion, when depressions in LCMR_{glc} and LCBF are matched and coincide with regions of attenuation seen on the CT scan.

In projection areas distant from the infarct site, the FDG scan will demonstrate hypometabolism in deep structures and broad zones of cerebral cortex that appear normal on CT scan (Kuhl et al. 1980a; Metter et al. 1981; Phelps et al. 1981), evidence of a dysfunction that is also apparent on LCBF or LCMRO_2 scans (Baron et al. 1981; Lenzi et al. 1981). For example, decreased LCMR_{glc} or LCMRO_2 and a normal CT scan will commonly be found in the hemi-

sphere contralateral to a supratentorial infarct, in the thalamus or caudate ipsilateral to a cortical infarct (Fig. 3), in the cortex ipsilateral to a subcortical infarct (Fig. 4), in the primary visual cortex ipsilateral to an infarct involving more anterior visual pathways and associated with a homonymous hemianopia (Fig. 3), and in the cerebellar hemisphere contralateral to supratentorial infarcts (Fig. 5).

In stroke, estimating the extent of brain damage by CT scan can be misleading. Finding decreased metabolism in apparently structurally intact brain distant from, but synaptically connected to, infarcted tissue can help define the true extent of the functional impairment in stroke and explain clinical symptoms. For example, the kind of functional detail shown in the FDG scan of Fig. 4 can help localize the cortical or subcortical site of primary deficit in

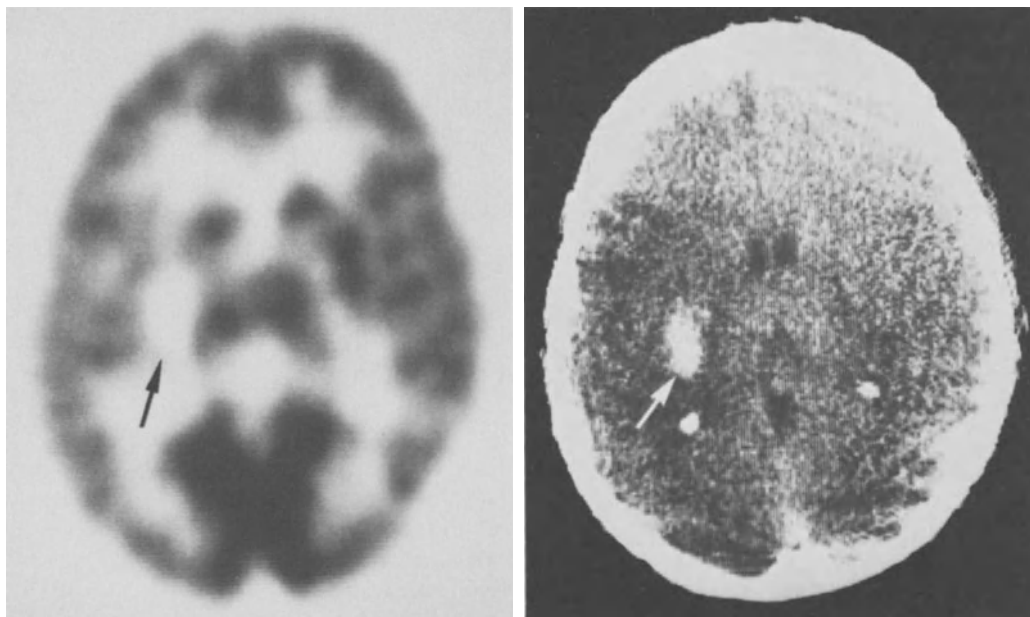


Fig. 4. FDG (*left*) and CT (*right*) scans of a 34-year-old man in whom a subcortical hemorrhage produced a small focal infarct in the left internal capsule and putamen (*arrows*) which was accompanied by transient aphasia and right hemiparesis. The CT scan was made immediately after the hemorrhage, the

FDG scan many months later. Only the FDG scan clearly shows not only the extent of local dysfunction in putamen and thalamus, but also the apparently permanent metabolic decline in left frontotemporo-parietal cortex. (From Kuhl, to be published)

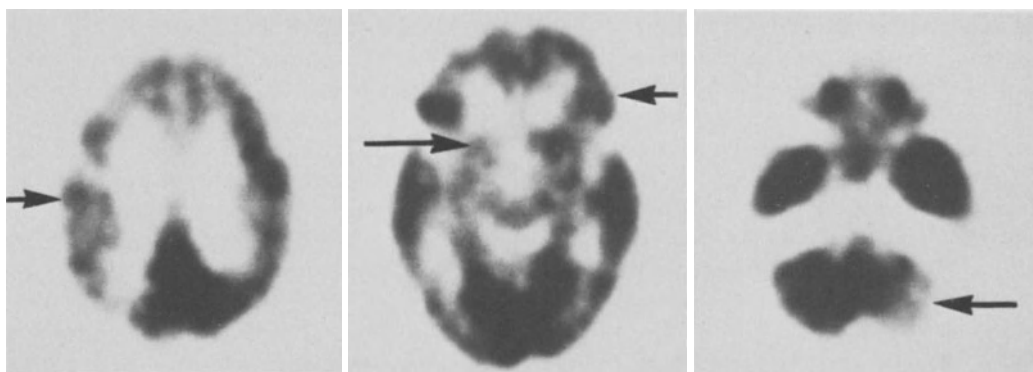


Fig. 5. FDG scans at three different brain levels in a 61-year-old man with multiple infarct dementia. *Arrows* point to asymmetric zones of hypometabolism

in left parietal cortex and caudate and in right frontal cortex and cerebellar hemisphere. (From Kuhl, to be published)

aphasic patients (Metter et al. 1981; Kuhl, to be published). The FDG scan can also be useful in the study of the elderly demented patient in whom stroke is only suspected. Although CT scans may be similar and nondiagnostic, the FDG scan in multiple infarct dementia commonly shows scattered asymmetric metabolic

defects (Fig. 5) (Kuhl, to be published), unlike the symmetrical metabolic defects which selectively involve association cortex in patients with AD (Benson et al. 1981), or the normal metabolic symmetry seen in healthy elderly patients (Kuhl et al. 1982b).

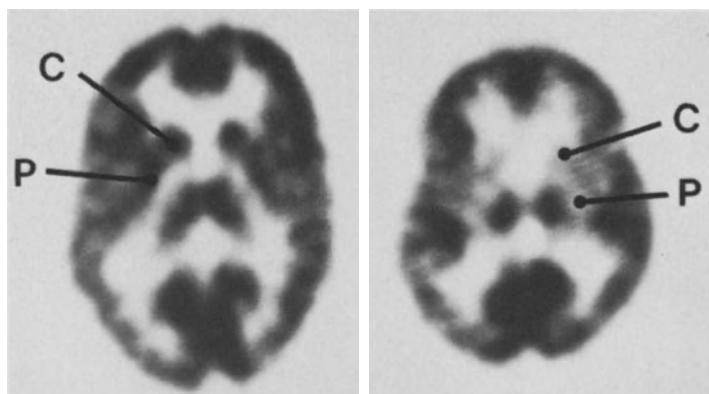


Fig. 6. FDG scans in a normal person (*left*) and in a patient with HD (*right*). In HD patients, the caudate (C) and putamen (P) are hypometabolic

Huntington's Disease

Huntington's disease (HD) is an inherited degenerative disorder of unknown cause characterized by development and progressive increase in chorea and dementia. There is evidence that at least the motor symptoms may result from an imbalance of the effects of dopamine, acetylcholine, and gamma-aminobutyric acid released from nerve terminals in the striatum (the putamen and caudate nucleus) and substantia nigra (Spokes 1981). At autopsy the brain typically shows widespread neuronal cell loss and glial proliferation, particularly in the caudate, putamen, and cerebral cortex (Forno and Jose 1973). Characteristic striatal atrophy, accompanied by ventricular dilatation and cortical sulcal widening, has been documented by CT (Terrence et al. 1977).

To learn the relationship of local cerebral metabolism and atrophy in this disease, we performed FDG scans on 13 patients with HD, 15 subjects at risk for HD, and 40 normal control subjects (Kuhl et al. 1981a, 1982c). These data were compared with CT measures of cerebral atrophy, with age, and with duration and severity of symptoms.

In this HD group, severity of dementia was closely associated with CT measures of central atrophy, suggesting that atrophy and symptoms worsen together as the disease progresses. A more significant finding was that overall CMR_{glc} , and particularly mean cortical CMR_{glc} , were normal in most HD patients and correlated with neither duration of disease nor severity of dementia. In contrast, patients with senile dementia have been found consistently

to have approximately 35% reductions in global CBF (Ingvar 1973), $CMRO_2$ (Frackowiak et al. 1981), and CMR_{glc} (Hoyer 1978) when dementia was severe, and lesser reductions when dementia was mild. In FDG scans of patients with AD, CMR_{glc} is symmetrically reduced in association cortex (Benson et al. 1981), but in HD patients we found only a minor decrease in frontal relative to parietal cortex metabolism in those with disease duration greater than 6 years, and the decrease did not correlate with the severity of dementia.

Finding nearly normal cortical CMR_{glc} in demented HD subjects is in agreement with both clinical and biochemical data suggesting that the primary dementing process in HD may not be cortical in location. The behavioral syndrome in HD includes preservation of vocabulary and facility with language, and is the so-called subcortical dementia in which the symptoms include apathy, mental slowing, memory disorder, and personality change (Albert 1978). Pathology is most prominent in subcortical structures, in contrast to the so-called cortical dementias, such as in AD, where pathology is most prominent in association cortex. In HD, there is a normal cortical level of acetyl choline transferase, the marker enzyme for cholinergic neurons (Spokes 1980); in AD it is decreased (Bowen et al. 1976).

In this same study, FDG scans showed that $LCMR_{glc}$ was markedly depressed in the caudate and putamen of HD patients (Fig. 6), even in the absence of caudate atrophy. Early in HD, hypometabolism is out of proportion to atrophy in the caudate and appears to be a process that occurs prior to bulk loss of striatal

tissue. Since the synapse is the primary site of glucose utilization in the brain (Sokoloff 1981), this disproportionate metabolic decline might result from an early reduction of striatal terminals, which may occur in HD before neuron death. Alternatively, early striatal hypometabolism may reflect only early neuron dropout, the volume of which has been balanced by glial cell replacement. Later, with increasing duration of HD, striatal neuron loss progresses further and symptoms appear and then worsen, explaining the subsequent correlation of caudate atrophy with duration and with severity of symptoms.

In asymptomatic subjects at risk we found normal CT scans and normal CMR_{glc} patterns, except for possible caudate hypometabolism in some. Striatal CMR_{glc} was usually markedly decreased (Fig. 6) soon after the first appearance of symptoms in HD patients, as noted above. Klawans et al. (1972) and Finch (1979) have proposed a hypothetical pre-existing deficit in number or function of striatal neurons in asymptomatic carriers; when this exceeds a critical threshold, choreiform movements should appear. Since no information is available about the time course of striatal neuron loss in carriers or in HD patients, it is not known if such a striatal deficit would be present at birth or would appear later. Our finding of elevations in caudate MR indices to greater than 2 SD above the normal mean in 6 of 15 at-risk subjects suggests the possibility that striatal glucose utilization may be reduced in asymptomatic carriers of the autosomal dominant HD gene. Careful long-term longitudinal studies will be important in asymptomatic offspring of parents who have HD in order to discover definitely whether striatal hypometabolism is common in carriers and, if so, to determine whether its presence increases the probability of developing the disease.

Epilepsy

Diagnosis and classification of epilepsy depends heavily on the use of electroencephalography (EEG), which records electrical activity associated with neuronal activity. Since altered cerebral function in epilepsy is seldom accom-

panied by changes in structure detectable by radiographic procedures, it has been of interest to explore other means of assessing the severity and extent of underlining cerebral dysfunction, not only to lateralize or localize epileptogenic tissue, but also to improve understanding of the disorder.

Initial findings in a UCLA study (Kuhl et al. 1980b) were that the interictal FDG scan effectively detected dysfunctional brain zones considered most likely to be responsible for seizures in patients with partial epilepsy (Fig. 7), even though these zones usually appeared normal on CT scans. This has now been further confirmed with studies on 50 patients for whom ictal as well as interictal EEG data were available (Table 1) (Engel et al. 1982a-c, to be published; Kuhl et al., to be published). The location of focally decreased CMR_{glc} in interictal FDG scans was closely associated with the epileptogenic focus, determined by a combination of interictal and ictal EEG recording. Only two of these patients had focal CT scan abnormalities at the site of the presumed epileptogenic lesion. No correlation could be demonstrated between the percentage decrease in CMR_{glc} within the zone of hypometabolism and the frequency of depth-EEG-recorded interictal epileptiform events; however, this was not surprising considering the complicated relationship between interictal electrical and metabolic processes.

There is a good correlation between metabolic and combined electrophysiological techniques, which has important diagnostic significance with respect to localization of the epileptogenic focus. Both techniques are capable of producing false positive as well as false negative results, but when used in combination yield more reliable localizing information. The EEG is useful for confirming if a hypometabolic zone is epileptogenic; the FDG scan helps determine whether an epileptic EEG focus is likely to represent a primary epileptogenic region or propagation from a distance. The FDG scan also serves as an independent confirmation of the epileptogenic site. In presurgical evaluation, confirmation of the EEG-identified epileptogenic region by demonstration of a zone of hypometabolism has proved to be a useful criterion for determining the site of resection. In 25 patients with partial epilepsy who under-

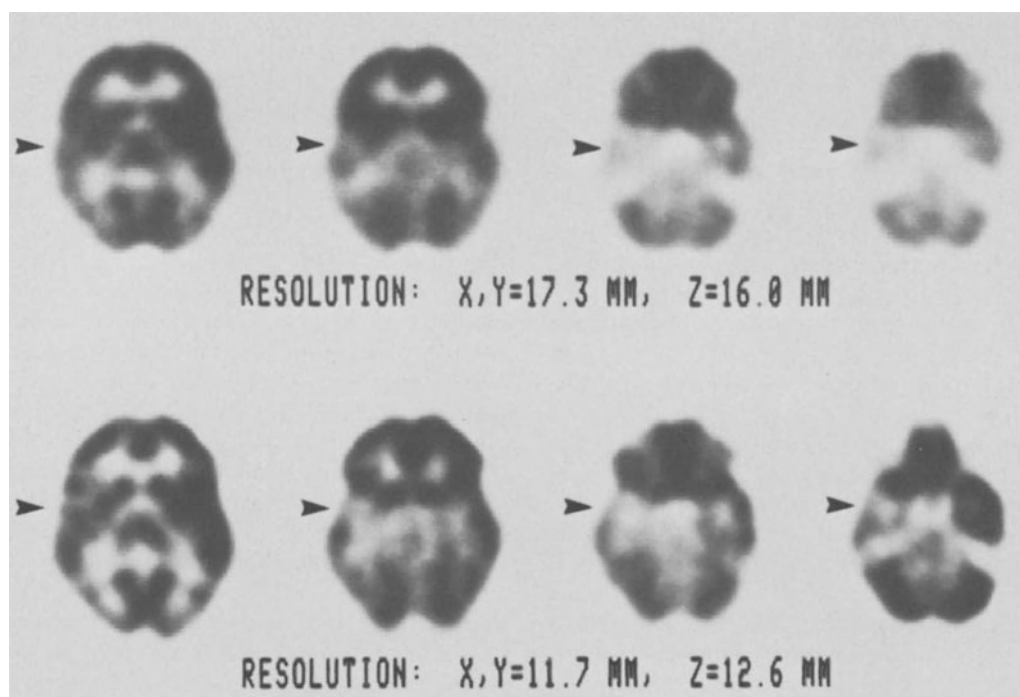


Fig. 7. Interictal FDG scans at different brain levels of a 22-year-old woman who had complex partial seizures originating in the left temporal lobe. The two studies are separated by a 4-month interval. Note the hypometabolic zone in the left anterior temporal lobe (arrows), which corresponds in location to the EEG-determined site of seizure onset, the unchanged appearance of this metabolic defect over a 4-month interval, and the degree of improved anatomic clarity

resulting from better spatial resolution of a newer tomograph (below). *Upper set:* ECAT II tomograph (EG & G/ORTEC Inc., Oak Ridge, Tennessee), 10 mCi FDG, 10 min per image, 1.5–2.4 million counts per image. *Lower set:* NeuroECAT tomograph (EG & G/ORTEC) 10 mCi FDG, 10 min per three images, 2.6–3.8 million counts per image. (From Kuhl et al. 1982b)

went anterior temporal lobectomies, the presence of lesions such as mesial temporal sclerosis, tumors, and heterotopia were highly correlated with hypometabolic zones detected in the preoperative interictal FDG scan (Table 2) (Engel et al., to be published b,) although preoperative CT scans had usually been normal.

In most instances, the extent of the hypometabolic zone was greater than the extent of the pathological changes noted in the excised specimen. The location and degree of hypometabolism has been unchanged in several patients who have undergone repeat scanning (Fig. 7), but whether the hypometabolic zone represents a relatively fixed deficit due to structural changes is still unsettled.

Interictal FDG scans carried out in patients with absence seizures who were diagnosed as having petit mal epilepsy have demonstrated

no abnormal patterns of CMR_{glc} (Engel et al., to be published).

In most patients, the FDG scan performed during partial seizures shows a marked local increase in CMR_{glc} at the site of EEG-demonstrated ictal onset (Kuhl et al. 1980b; Engel et al., to be published d), as shown in Fig. 8, but the magnitude of metabolic alteration has not correlated well with the duration or frequency of ictal EEG events. Ictal patterns of hypermetabolism and hypometabolism are more varied than the focal deficits found in the interictal state, and are less useful in predicting epileptic origin. One reason is that the ictal FDG scan does not distinguish between hypermetabolism associated with the seizure origin and that resulting from propagated neuronal activity, as illustrated in Fig. 8. It is difficult to obtain ictal FDG scans in partial epilepsy, and more expe-

rience is needed to understand better the significance of these patterns.

Ictal FDG scans have also been performed on patients during nonfocal seizures (Engel et al., to be published). When FDG is injected at the time of electroconvulsive shock treatment, there is a diffuse increase in CMR_{glc} , when injection is made at the end of the seizure and beginning of the postictal depression, there is a generalized decrease in CMR_{glc} . Ictal FDG scans have also been performed on patients during petit mal seizures induced by hyperventilation (Engel et al., to be published). These ictal episodes were associated with a diffuse increase in CMR_{glc} compared to control scans performed after successful treatment and when no absences occurred. Unlike the typical ictal FDG scan found in partial epilepsy, the relative distribution of activity appeared normal in ictal scans of petit mal patients.

Clinical Availability

There is good evidence now that the FDG scan performed by means of PECT is efficacious in localizing cerebral dysfunction undetected by CT in cerebrovascular, degenerative, and epileptic disorders. In its present state of development, the complexity and cost of this cyclotron-based technology will limit access to relatively few institutions. On the other hand, similar localization information might be provided by an equally effective single photon tomographic (SPECT) determination of the LCBF pattern in these patients. For example, in the UCLA series, local decreases in LCBF and $LCMR_{glc}$ were matched in the epileptogenic focus during the interictal PECT scans (Kuhl et al. 1980b). This suggests that in many clinical problems the same diagnostic information might be provided by a high-quality LCBF scan as by an $LCMR_{glc}$ scan. Recently, quantitative LCBF mapping has been accomplished using SPECT with a potentially readily available radiopharmaceutical labeled with ^{123}I , *N*-isopropyl-*p*-[^{123}I]-iodoamphetamine (IMP) (Kuhl et al. 1981b, 1982a). There is promise that less expensive SPECT scanners combined with more readily available radiopharmaceuticals such as IMP will make more accessible the kind of

physiological imaging previously restricted to the more expensive positron tomographic systems. At the present time, however, PECT is the more powerful method for research because it should permit an investigator to map quantitatively local cerebral values of blood flow, blood volume, oxygen and glucose utilization, protein synthesis, amino acid utilization, and receptor binding of neurotransmitters. Some of these determinations are possible now; others are potentially available. Hopefully, with time and experience the information learned from such studies will permit new insights into the pathogenesis of cerebral disorders. This will determine the eventual role of cerebral ECT in clinical management.

Acknowledgments. UCLA coinvestigators in work discussed here included J. R. Barrio, D. F. Benson, J. Engel Jr., R. Hawkins, E. J. Hoffman, S. C. Huang, N. MacDonald, J. C. Mazziotta, E. J. Metter, M. E. Phelps, and W. H. Riege. This project was supported in part by Department of Energy contract #DE-AM03-76-SF00012 and USPHS research grants R01-CA 32855 and P01-NS-15654.

References

1. Ackerman RH, Correia JA, Alpert NA et al. (1981) Positron imaging in ischemic stroke disease using compounds labeled with oxygen 15. *Arch Neurol* 38: 537-543
2. Albert ML (1978) Subcortical dementia. In: Katzman R, Terry RD, Bick KL (eds) *Alzheimer's disease: senile dementia and related disorders* (Aging, vol 7). Raven, New York pp 173-180
3. Baron JC, Bousser MG, Comar D et al. (1981) Noninvasive tomographic study of cerebral blood flow and oxygen metabolism in vivo. *Eur Neurol* 20: 273-284
4. Benson DF, Kuhl DE, Phelps ME, Cummings L, Tasai SY (1981) Positron emission computed tomography in the diagnosis of dementia. *Ann Neurol* 10: 76
5. Bowen DM, Smith CB, White P, Davison AN (1976) Neurotransmitter related enzymes and indices of hypoxia in senile dementia and other abiotrophies. *Brain* 99: 459-496
6. Brody H (1976) An examination of cerebral cortex and brainstem aging. In: Terry RD, Gershon S (eds) *Neurobiology of aging*. Raven, New York pp 177-181

Table 1. In partial epilepsy, the locations of metabolic defects in interictal FDG scans were closely associated with the localizations of epileptogenic foci determined with a combination of surface and depth EEG telemetry during interictal and ictal epileptiform activity. XCT scans were rarely abnormal at the epileptogenic focus. (From Kuhl et al., to be published; data from Engel et al. 1982a)

Epileptic patients (<i>n</i> = 50) <i>FDG scan</i>		
With defects (<i>n</i> = 35)	Agree with	EEG localization: 28
	Do not agree with	EEG localization: 3
	Nondiagnostic	EEG localization: 4
Without defects (<i>n</i> = 15)	Definite	EEG localization: 5
	Nondiagnostic	EEG localization: 10
<i>XCT scan</i>		
Focal abnormality at EEG localization site: 2		
Diffuse abnormalities or normal: 48		

Table 2. In patients with partial epilepsy who had undergone anterior temporal lobectomies, the presence of a lesion within the specimen (usually mesial temporal sclerosis) was highly correlated with decreased LCMR_{glc}, as detected in the preoperative interictal FDG scan. Preoperative CT scans were usually normal at the focus. Patient outcome grading (short follow-up; nine cases <1 year): 1, seizure-free since surgery; 2, few seizures, then seizure-free; 3, worthwhile improvement; 4, no improvement. (From Kuhl et al., to be published c; data from Engel et al. 1982b)

Anterior temporal lobectomies (<i>n</i> = 25)		
	Normal	Ab- normal
FDG scan	3	22
Pathological examination	6	19

Patient outcome	Grade	1	2	3	4
	<i>n</i>	20	2	2	1

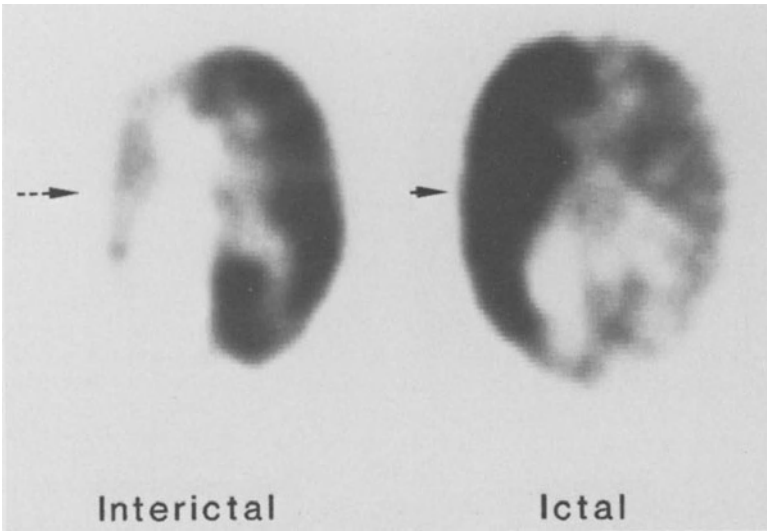


Fig. 8. Interictal and ictal FDG scans of a 12-year-old girl with a 6-year history of right-sided tonic-clonic seizures and hemiparesis. The interictal FDG scan showed marked *hypometabolism* in the cerebral cortex of the left hemisphere (*dotted arrow*). In contrast, the ictal FDG scan showed marked left cortical *hypermetabolism* (*solid arrow*) that coincided with bilat-

eral jerking of the extremities and face and epileptiform EEG spike activity, most marked in the left parietal region. It is likely that the seizure activity originated in the left parietal region, then spread to the more normal left frontal structures where hypermetabolism was greatest. (From Kuhl et al., 1980b)

7. Engel J Jr, Kuhl DE, Phelps ME, Mazziotta JC (1982a) Interictal local cerebral glucose metabolism in partial epilepsy and its relation to EEG changes. *Ann Neurol* 12: 510–517
8. Engel J Jr, Brown WJ, Kuhl DE, Phelps ME, Mazziotta JC, Crandall PH (1982b) Pathological findings underlying focal temporal lobe hypometabolism in partial epilepsy. *Ann Neurol* 12: 518–528
9. Engel J Jr, Kuhl DE, Phelps ME, Crandall PH (1982c) Comparative localization of the epileptic focus in partial epilepsy with PCT and EEG. *Ann Neurol* 12: 529–537
10. Engel J Jr, Kuhl DE, Phelps ME (to be published d) Patterns of ictal and interictal local cerebral metabolic rate studied in man with positron emission computed tomography. In: Akimoto H, Kazamatsuri H, Seino M (eds) *Advances in epileptology*. Raven, New York
11. Finch CE (1979) The relationships of aging changes in the basal ganglia to manifestations of Huntington's chorea. *Ann Neurol* 7: 406–411
12. Forno LS, Jose C (1973) Huntington's chorea: A pathological study. In: Barbeau A, Chase TN, Paulson GW (eds) *Advances in neurology*, Vol 1. Raven, New York pp 453–470
13. Frackowiak RSJ, Pozzilli C, Legg NJ, DuBoulay GH, Marshall J, Lenzi GL, Jones T (1981) Regional cerebral oxygen supply and utilization in dementia. A clinical and physiological study with oxygen-15 and positron tomography. *Brain* 104: 753–778
14. Gottstein U, Held K (1979) Effects of aging on cerebral circulation and metabolism in man: In: Gotoh F, Nagai H, Tazaki Y (eds) *Cerebral blood flow and metabolism*. *Acta Neurol Scand* 60 [Suppl 72]: 54–55
15. Heiss WD, Kloster G, Vyska K, Traupe H, Freundlieb C, Becker V, Feinendegen LE, Stoeklin G (1981) Regional cerebral distribution of ^{14}C -methyl-D-glucose compared with CT perfusion patterns in stroke. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S506–S507
16. Hoyer S (1978) Blood flow and oxidative metabolism of the brain in different phases of dementia. In: Katzman R, Terry RD, Bick KL (eds) *Alzheimer's disease: senile dementia and related disorders*. Raven, New York pp 219–226
17. Huang SC, Phelps ME, Hoffman EJ, Sideris K, Selin CJ, Kuhl DE (1980) Noninvasive determination of local cerebral metabolic rate of glucose in man. *Am J Physiol* 238: E69–E82
18. Ingvar DH (1973) Cerebral blood flow and metabolism in complete apallic syndromes, in states of severe dementia, and in akinetic mutism. *Acta Neurol Scand* 49: 233–244
19. Ingvar DH (1979) "Hyperfrontal" distribution of the cerebral grey matter flow in resting wakefulness; on the functional anatomy of the conscious state. *Acta Neurol Scand* 60: 12–25
20. Klawans HL, Paulson GW, Ringel SP, Barbeau A (1972) Use of L-DOPA in the detection of pre-symptomatic Huntington's chorea. *N Engl J Med* 286: 1331–1334
21. Kuhl DE (to be published) The effects of aging and stroke on patterns of local cerebral glucose utilization. In: Reivich M (ed) *Cerebrovascular diseases, 13th research (Princeton) conference*. Raven, New York
22. Kuhl DE, Phelps ME, Howell AP, Metter EJ, Selin C, Winter J (1980a) Effects of stroke on local cerebral metabolism and perfusion: Mapping by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 47–60
23. Kuhl DE, Engel J Jr, Phelps ME, Selin C (1980b) Epileptic patterns of local cerebral metabolism and perfusion in humans determined by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 348–360
24. Kuhl DE, Phelps ME, Markham C, Winter J, Metter J, Riege W (1981a) Local cerebral glucose utilization in Huntington's disease determined by emission computed tomography of ^{18}F -Fluorodeoxyglucose. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S459–S460
25. Kuhl DE, Wu JL, Lin TH et al. (1981b) Mapping cerebral blood flow by means of emission computed tomography of N-isopropyl-p-[^{123}I]-iodoamphetamine (IMP). *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S25–S26
26. Kuhl DE, Barrio JR, Huang SC, Selin C, Ackerman RF, Lear JL, Wu JL, Lin TH, Phelps ME (1982a) Quantifying local cerebral blood flow by N-isopropyl-p-[^{123}I]-iodoamphetamine (IMP) tomography. *J Nucl Med* 23: 196–203
27. Kuhl DE, Metter EJ, Riege WH, Phelps ME (1982b) Effects of human aging on patterns of local cerebral glucose utilization determined by the (^{18}F) fluorodeoxyglucose method. *J Cereb Blood Flow Metabol* 2: 163–171
28. Kuhl DE, Phelps ME, Markham CH, Metter EJ, Riege WH, Winter J (1982c) Cerebral metabolism and atrophy in Huntington's disease determined by FDG and CT scan. *Ann Neurol* 12: 425–434
29. Kuhl DE, Engel J Jr, Phelps ME (to be published) Emission computed tomography in the study of human epilepsy. In: Ward AA (ed) *Proceedings of the association for research in nervous and mental disease*. Raven, New York
30. Lammertsma AA, Frackowiak RSJ, Lenzi GL, Heather JD, Pozzilli C, Jones T (1981) Accuracy of the oxygen-15 steady state technique for measuring CBF and CMRO_2 : tracer modelling, statistics and spatial sampling. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S3–S4
31. Lenzi GL, Frackowiak RS, Jones T (1981) Regional cerebral blood flow (CBF), oxygen utilization (CMRO_2) and oxygen extraction ratio (OER) in acute hemispheric stroke. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S504–S505

32. Mazziotta JC, Phelps ME, Carson RE, Kuhl DE (1982a) Tomographic mapping of human cerebral metabolism: sensory deprivation. *Ann Neurol* 12: 435–444
33. Mazziotta JC, Phelps ME, Carson RE, Kuhl DE (1982b) Tomographic mapping of human cerebral metabolism: auditory stimulation. *Neurology (Minneapolis)* 32: 921–937
34. Metter EJ, Wasterlain CG, Kuhl DE, Hanson WR, Phelps ME (1981) 18 FDG positron emission computed tomography in a study of aphasia. *Ann Neurol* 10: 173–183
35. Phelps ME, Hoffman EJ, Huang SC, Kuhl DE (1978) ECAT: A new computerized tomographic imaging system for the positron emitting radiopharmaceuticals. *J Nucl Med* 19: 635–647
36. Phelps ME, Huang SC, Hoffman EJ, Selin CS, Sokoloff L, Kuhl DE (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with (F-18) 2-fluoro-2-deoxyglucose: validation of method. *Ann Neurol* 6: 371–388
37. Phelps ME, Mazziotta JC, Kuhl DE et al. (1981) Tomographic mapping of human cerebral metabolism. Visual stimulation and deprivation. *Neurology* 31: 517–519
38. Reivich M, Kuhl DE, Wolf A, Greenberg J, Phelps ME, Ido T, Casella V, Fowler J, Hoffman E, Alavi A, Sokoloff L (1979) The ^{18}F -fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
39. Smith CB, Gooch C, Rapoport SI, Sokoloff L (1980) Effects of aging on local rates of cerebral glucose utilization in the rat. *Brain* 103: 351–365
40. Sokoloff L (1981) The relationship between function and energy metabolism: its use in the localization of functional activity in the nervous system. *Neurosci Res Program Bull* 19: 159–210
41. Sokoloff L, Reivich M, Kennedy C, Des Rosiers H, Patlak CS, Pettigrew KD, Sakurada O, Shinohara M (1977) The (^{14}C) deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
42. Spokes EGS (1980) Neurochemical alterations in Huntington's chorea. A study of post-mortem brain tissue. *Brain* 103: 179–210
43. Spokes EGS (1981) The neurochemistry of Huntington's chorea. *Trends Neurosci* 4: 115–118
44. Terrence CF, Delaney JF, Alberts MC (1977) Computed tomography for Huntington's disease. *Neuroradiology* 13: 173–175
45. Williams CW, Crabtree MC, Burke MR et al. (1981) Design of the NeuroECAT. A high resolution, high efficiency positron tomograph for imaging the adult head or infant torso. *IEEE Trans Nucl Sci NS-28*: 1376–1741

Human Sensory Stimulation and Deprivation as Demonstrated by Positron Computed Tomography

By M. E. Phelps and J. C. Mazziotta

Since functional anatomy is revealed by positron computed tomography (PCT), the effects of sensory stimulation and deprivation can be examined in human subjects. Such data are essential in defining the neuroanatomical location and response characteristics of the normal brain and serve as a necessary prelude to future studies of pathological states. Examination of normal subjects in "resting" or sensory deprived states establishes the range of conditions that one can encounter when specific stimuli are not administered to a test subject. Stimulation studies using visual, auditory, or tactile inputs can define in humans the stimulus-response characteristics for the brain previously obtainable only from animal studies using the invasive neurophysiological techniques. Once a stimulus-response data base has been developed in normal subjects these same studies could then be performed in patients with known or suspected neuropathology to look for subtle aberrations in cerebral metabolism, blood flow, or other physiological parameters which might be overlooked during simple resting studies.

Among the first experiments performed with PCT was the use of sensory stimuli, including visual, auditory, and tactile modalities (Reivich et al. 1979a). In addition, an ever increasing amount of normative data is accumulating which defines the magnitude and range of local normal human cerebral glucose (Phelps et al. 1979, Huang et al. 1980, Reivich et al. 1979b; Kuhl et al. 1980; Mazziotta et al. 1981a) and oxygen metabolism (Baron et al. 1981; Frackowiak et al. 1980; Lebrun-Grandie et al., to be published) as well as cerebral blood flow (Baron et al. 1981; Frackowiak et al. 1980; Lebrun-Grandie et al., to be published). Such data demonstrates left-right symmetry for these parameters under resting conditions which minimize sensory inputs (Mazziotta et al. 1981a; Baron et al. 1981; Frackowiak et al. 1980; Lebrun-Grandie et al., to be published). An ex-

ception to this was reported by Finklestein et al. (1980), who described relative left > right oxygen and glucose metabolism in normal subjects under resting conditions.

Using ^{18}F -fluorodeoxyglucose (FDG) and PCT, the following study was undertaken to examine the effects on cerebral glucose metabolism of (1) selective or combined forced auditory and visual sensory deprivation (Mazziotta et al., 1982a), (2) visual stimulation of progressively increasing complexity (Phelps et al. 1981a, b), (3) visual stimulation in patients with lesions of the visual pathway (Phelps et al. 1981b), and (4) monaurally and binaurally presented verbal (factual story) and nonverbal (music, tone sequences, chord pairs) auditory stimuli (Mazziotta et al. 1982b).

Materials and Methods

Scanning Procedures

Basic scanning procedures have been described previously (Phelps et al. 1979; Huang et al. 1980), as has the preparation of FDG (Ido et al. 1978; Barrio et al. 1981). The tomograph used in most of the following studies was the ECAT II (EG&G/ORTEC, Oak Ridge Tennessee) PCT device (Phelps et al. 1978), which has an image spatial resolution of 1.6 ± 0.1 cm (full width at half maximum line source) in the plane and 1.6 ± 0.2 cm in the axial direction. Approximately 2-3 million counts were obtained per image in approximately 5-10 min. Three subjects were studied using the NeuroECAT PCT device (EG&G/ORTEC, Oak Ridge, Tennessee) (Hoffman et al., 1981) which has a spatial resolution of $9.8 \times 9.8 \times 13.5$ mm.

Typical subject FDG doses were 5-10 mCi. Following injection with FDG the subjects received stimulation for a 40-min period and this

was followed by obtaining four or five tomographic images. The images were obtained through the perisylvian region at 8-mm intervals (Fig. 1) parallel to the canthomeatal line using a specially designed head holder system (Mazziotta et al., 1982c).

Subjects

All subjects gave informed consent in accordance with the regulations of the UCLA Human Use Committee. All volunteer subjects were judged to be clinically normal by both general and neurological examination. The age range of the subjects was 21–28 years and all were strongly right-handed as evaluated by standard questionnaires. With the exception of the studies on patients with visual pathway lesions (see below), all stimulation studies were performed in pairs, that is a stimulation study was performed in either the auditory or visual mode and a control study was also performed during which the input modality was occluded (by either eye patches or earplugs and soundproof headphones).

The subjects were paid for their participation in the study. Regardless of the paradigm to which they were assigned, they were asked to lie supine on the padded scanner bed in a room with low ambient light and were asked not to move or speak. They were not spoken to and the only room noise was that of equipment fans and motors. All intravenous lines and scalp electrodes for EEG monitoring were put in place 10–15 min prior to FDG injection. The subjects were not touched and did not know the exact time of the FDG injection.

Subject Groups

Sensory Deprivation

Three subgroups were studied independently and will be referred to by the sensory modality which was eliminated from the subject's environment. The ears-closed group included eight subjects who had their eyes exposed, enabling them to see the white ceiling above the tomograph, but had their ears plugged with stoppers and covered with soundproof headphones. The

eyes-closed group contained six subjects who had their eyes patched with light proof material but had their ears exposed. The both-closed group contained nine subjects who had their eyes patched and their ears plugged. One subject was studied in both the ears-closed and the both-closed conditions.

Visual Stimulation in Normal Volunteers

All subjects were allowed 10–15 min of dark adaptation before the study was initiated. Except as noted all visual stimuli were presented binocularly. The first visual stimulus ($n = 5$) consisted of a bright, white light (570 lux). This stimulation was started 5 min before injection and continued throughout the study. The second level of stimulation consisted of a 2 cycle/s alternating black-and-white checkerboard pattern with a 20×20 array of 1-cm squares, a central fixation point, and a distance of 0.5 m from the subject ($n = 6$). Three of these subjects also had monocular checkerboard stimulation studies. The third and most complex stimulation ($n = 2$) consisted of allowing the subjects to view the surroundings of a park beside our laboratory.

Visual Stimulation of Patients with Lesions of the Visual Pathway

Seven patients had homonymous hemianopsia due to unilateral lesions of the visual pathway (i.e., involving the thalamus and/or optic radiation). The visual cortex of all these subjects appeared anatomically normal by X-ray CT. The age range of these subjects was 47–67 years, with a mean of 57 years.

Auditory Stimulation

Four types of auditory stimuli were used and the subjects were asked to adjust the stimulus intensity to the lowest level that was still easily perceptible. The stimuli were delivered monaurally through one channel of a pair of stereophonic headphones. The nonstimulated ear was occluded with an earplug. In each subgroup approximately equal numbers of subjects had left and right ear stimulations.

Four subjects received verbal stimuli, which consisted of a Sherlock Holmes story. These

subjects were told that following completion of the scanning procedure they would be tested on the content of the story and would be paid in proportion to their score. In addition, three separate subjects had the same stimulus but were scanned using the high-resolution NeuroECAT device.

Two types of nonverbal stimuli were used. The first was the tonal memory test consisting of pairs of tone sequences. The initial sequence contained three, four, or five tones of different frequencies but equal intensity, duration, and interval. The subjects heard this sequence and then after a brief pause a second sequence which was identical or differed in the frequency of one tone. Subjects were asked to identify different sequence pairs by depressing a microswitch with the right index finger.

The second type of nonverbal stimulus was called the timbre test and consisted of chord pairs of complex harmonic composition. When the chord pairs were different the subjects had to respond by using the microswitch under their right index finger.

The tonal memory group contained eight participants and the timbre group had four. In addition, two subjects received both the factual story and music (Brandenburg Concertos) presented binaurally.

In the stimulation studies data were analyzed to determine the percentage change (control to stimulated state) of the local cerebral metabolic rate for glucose (LCMRG1c) as well as the change of the local value from the overall

cross-sectional mean. In addition, the significance of left-right asymmetries was calculated for local regions.

Results

Sensory Deprivation

Subjects in the ears-closed and eyes-closed group demonstrated cerebral metabolic left/right symmetry. This symmetry was confirmed statistically and is presented in Table 1. The both-closed group demonstrated a left > right asymmetry. This asymmetry is illustrated in Fig. 1 and proved to be significant in five regions as well as for the overall hemispheric mean (Fig. 1, 2; Table 1). The regions that were significantly asymmetric included anterior inferior prefrontal, posterior inferior frontal, posterior superior temporal, and lateral occipital cortex. Hemispheric asymmetry in this group was significant at the $p < 0.05$ level.

For the single patient studied in two states of sensory deprivation, a change from left > right metabolic asymmetry in the both-closed state to a left-sided metabolic symmetry in the ears-closed state was observed.

When the overall metabolism from the four or five tomographic images of each subject in each sensory deprived state were averaged and compared between groups, a progressive and significant decrease in CMRGlc was found in

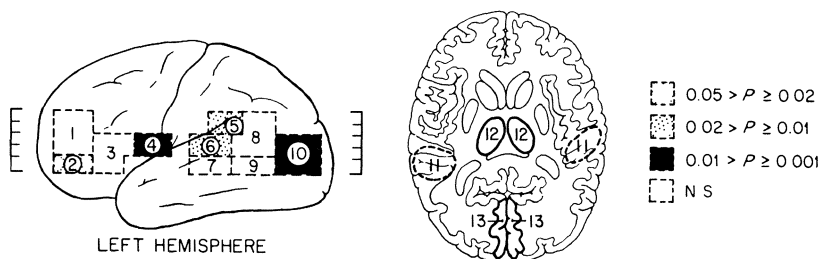


Fig. 1. The superficial and deep cerebral regions analyzed in the sensory deprivation and auditory stimulation studies. The numbers correspond to those used in Tables 1 and 3. The brackets on the *left* illustrate the approximate position, angle, and extent of the tomographic planes obtained in most subjects. Asymmetries are demonstrated from the both-closed subgroup of the sensory deprivation experiment group; the *shaded zones* represent cortical areas hav-

ing significant left > right asymmetries. *Unshaded areas* did not have significant asymmetries. The metabolic rates were always greater in the left hemisphere in the both-closed group and hence only the left hemisphere is shown. The percentage asymmetries corresponding to the p values are: $0.05 > p > 0.02 = 3\%$, $0.02 > p > 0.01 = 4\%-5\%$, and $0.01 > p > 0.01 = 5\%-7\%$. Data taken from Mazziotta et al. (1982a)

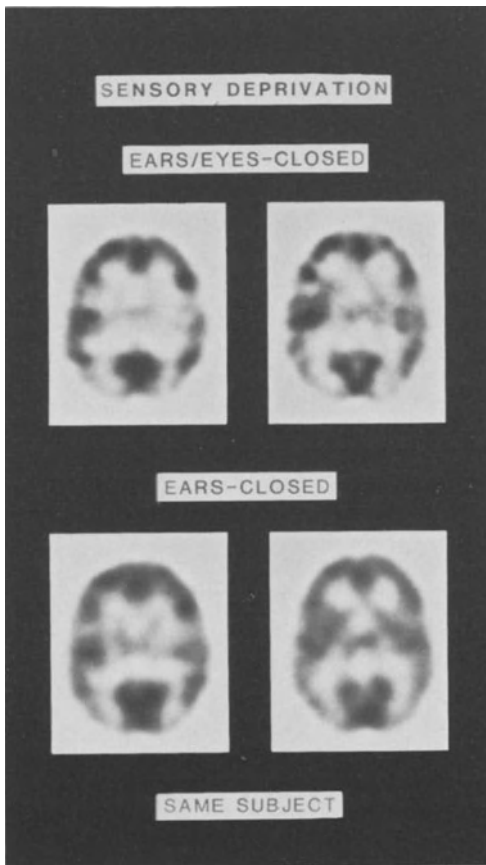


Fig. 2. Two sets of images from a normal individual studied with FDG to measure local cerebral glucose metabolic rate in two states of sensory deprivation. The *upper* row of images demonstrates the subject studied with both auditory input occlusion and light-excluding eye patches. Asymmetries are seen in this state with the right side being relatively hypometabolic compared with the left. The *lower* set of images was obtained from the same subjects with his ears closed but his eyes open. This set of images demonstrates left-right symmetry throughout, as was typical of the partially sensory deprived test states (Mazziotta et al., 1982a, Phelps et al. 1982)

going from the eyes-closed (6.99 ± 1.49 mg/100 g per minute) to the ears-closed (4.77 ± 0.73) state (32% ; $p < 0.001$) and from the ears-closed to the both-closed (3.79 ± 1.00) state (17% ; $p < 0.01$) (Mazziotta et al., 1982a). The decrease in metabolic activity in the right hemisphere was significantly greater than in the left in this last transition. The reason for the hemispheric asymmetry in the both-closed state was

therefore due to a larger right than left hemispheric metabolic depression in the most fully sensory deprived condition.

Anterior-posterior asymmetries were also calculated. The mid-prefrontal cortex was compared to the parietal and lateral occipital cortex in all states. Relatively increased frontal metabolism was seen in all states bilaterally. When compared to the parietal cortex, the percentage increase in frontal LCMRGlc (from 7.3% to 18.7%) did not reach statistical significance in any of the test conditions. However, when compared with the lateral occipital cortex, the frontal cortical relative hypermetabolism (16.9% to 48.3%; $p < 0.001$) was significant and progressively increased from the eyes-closed to the ears closed to the both-closed states.

Visual Stimuli in Normal Subjects

Progressive increases in both the primary (PVC) and the associative (AVC) visual cortex were demonstrated with visual stimuli of increasing complexity (Phelps et al. 1981a, b). That is, the metabolic rate of the visual cortex increased progressively from the eyes-closed state through the white light condition, and the alternating checkerboard stimulus to the complex scene ($p < 0.01$) (Figs. 3, 4; Table 2). The AVC reached a higher metabolic value in the most complex state when compared to the PVC and the rate of increase for the AVC exceeded that for the PVC as stimulus complexity increased.

In all test states, eyes-closed, white light, checkerboard, and complex scene, and in both monocular and binocular stimulations, metabolic left/right asymmetry was demonstrated for both the AVC and PVC.

Visual Stimulation of Patients with Lesions of the Visual Pathway

All hemianopic subjects exhibited reduced LCMRGlc in the visual cortex contralateral to the visual field deficit. The average LCMRGlc of the involved PVC and AVC was respectively $32\% \pm 7\%$ and $44\% \pm 6\%$ lower than the contralateral side (Phelps et al. 1981b). The metabolic response of the visual cortex was normal

Table 1. Local and overall metabolic rates for glucose utilization (mg glucose/min/100 g brain; mean $\pm$ SD) in three states of sensory deprivation. Region numbers refer to regions outlined in Fig. 1. Data taken from Mazziotta et al. (1982a)

Region	Eyes closed (<i>n</i> =6)		Ears closed (<i>n</i> =8)		Both closed (<i>n</i> =9)	
	Left	Right	Left	Right	Left	Right
1	8.55 $\pm$ 1.87	8.60 $\pm$ 1.85	5.91 $\pm$ 0.96	5.92 $\pm$ 0.88	5.13 $\pm$ 1.22	5.05 $\pm$ 1.17
2	8.93 $\pm$ 0.23	9.19 $\pm$ 0.05	5.63 $\pm$ 1.19	5.77 $\pm$ 1.08	4.71 $\pm$ 1.15	4.57 $\pm$ 1.13
3	9.92 $\pm$ 0.21	10.03 $\pm$ 0.11	6.13 $\pm$ 1.08	6.23 $\pm$ 0.98	5.24 $\pm$ 1.39	5.14 $\pm$ 1.36
4	8.55 $\pm$ 1.90	8.51 $\pm$ 2.20	5.59 $\pm$ 0.95	5.76 $\pm$ 0.81	4.80 $\pm$ 1.20	4.43 $\pm$ 1.00
5	8.35 $\pm$ 1.94	8.41 $\pm$ 2.00	5.38 $\pm$ 1.01	5.33 $\pm$ 0.90	4.71 $\pm$ 1.25	4.49 $\pm$ 1.35
6	9.09 $\pm$ 1.83	9.12 $\pm$ 1.92	5.93 $\pm$ 0.94	5.91 $\pm$ 0.92	4.96 $\pm$ 1.42	4.77 $\pm$ 1.46
7	9.42 $\pm$ 0.95	9.38 $\pm$ 0.87	5.75 $\pm$ 0.85	5.76 $\pm$ 0.85	4.84 $\pm$ 1.41	4.77 $\pm$ 1.33
8	7.96 $\pm$ 1.96	8.01 $\pm$ 2.05	4.90 $\pm$ 0.77	5.05 $\pm$ 0.81	4.48 $\pm$ 1.16	4.40 $\pm$ 1.04
9	8.40 $\pm$ 1.76	8.27 $\pm$ 1.80	4.97 $\pm$ 0.91	5.04 $\pm$ 0.84	4.19 $\pm$ 1.15	4.18 $\pm$ 1.04
10	7.13 $\pm$ 1.74	7.10 $\pm$ 1.81	4.21 $\pm$ 0.68	4.35 $\pm$ 0.64	3.12 $\pm$ 0.46	2.99 $\pm$ 0.51
11	9.72 $\pm$ 1.92	9.39 $\pm$ 1.97	6.57 $\pm$ 0.84	6.57 $\pm$ 0.77	5.27 $\pm$ 1.55	5.09 $\pm$ 1.58
12	7.51 $\pm$ 0.80	7.59 $\pm$ 0.68	5.90 $\pm$ 0.98	5.91 $\pm$ 0.95	4.48 $\pm$ 1.24	4.53 $\pm$ 1.21
13	8.45 $\pm$ 2.17	8.39 $\pm$ 2.09	6.50 $\pm$ 0.61	6.54 $\pm$ 0.63	4.39 $\pm$ 0.52	4.41 $\pm$ 0.51
Hemisphere	7.01 $\pm$ 1.55	6.97 $\pm$ 1.57	4.78 $\pm$ 0.76	4.75 $\pm$ 0.76	3.99 $\pm$ 1.04	3.96 $\pm$ 1.01

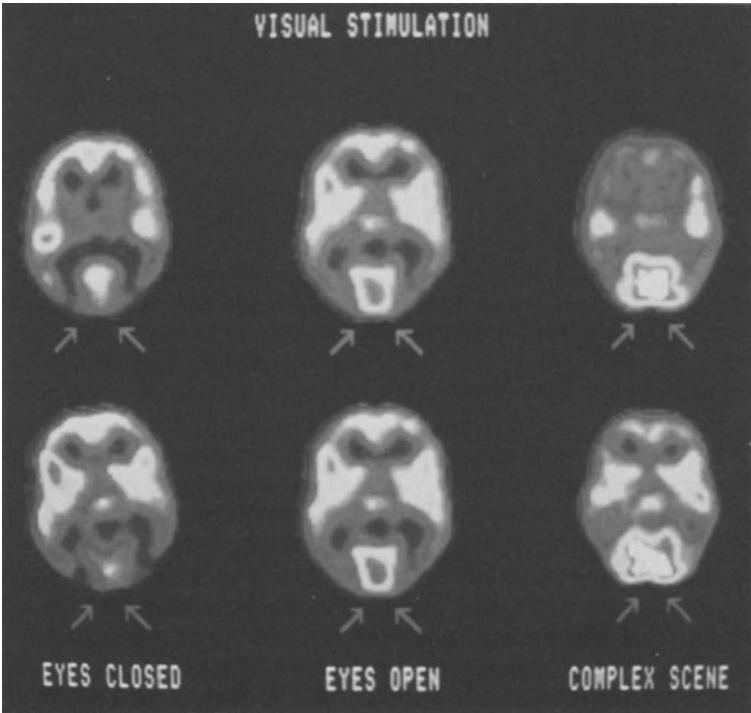


Fig. 3. Cerebral metabolic activity of the primary and associative visual cortex in the eyes-closed state compared to white light stimulation and the complex stimulation of an park. Note the progressive increase

in metabolic activity in both AVC and PVC as the visual stimulus complexity increases. (Phelps et al. 1981b)

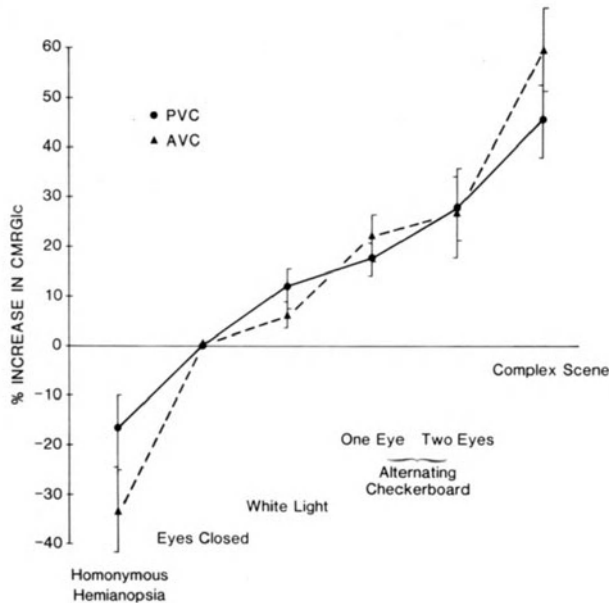


Fig. 4. Plot of the average percentage change in glucose metabolism of the PVC and AVC during three states of visual stimulation and in patients with homonymous hemianopsia due to optic radiation or thalamic lesions. The zero line represents the eyes-closed normal control state. Note the metabolic response of the AVC increases at a faster rate than the PVC as the visual scene becomes more complex. Maximal increases in local metabolic rates were typically about twice the average value shown. (Phelps et al. 1981b)

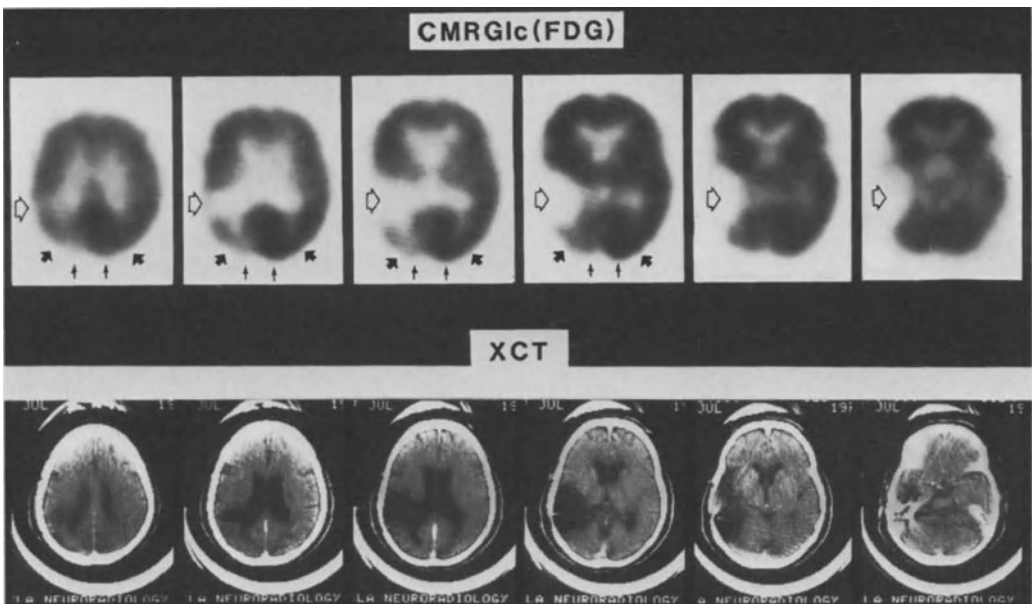


Fig. 5. X-ray CT (XCT) and PET studies of patient with right homonymous hemianopsia secondary to herpes encephalitis. X-ray CT demonstrates a large area of brain loss in the left posterior temporal regions disrupting the optic radiations. PET study demonstrates the anatomical defect in the same loca-

tion and distribution as the X-ray CT image. In addition, there is a striking asymmetry of the metabolic activity of the PVC and AVC. The visual cortex ipsilateral to the lesion is hypometabolic compared to the normal right side and correlates with the patient's right-sided visual field cut. (Phelps et al. 1981b)

Table 2. Progressive changes in LCMRGlc of the primary (PVC) and associative (AVC) visual cortex to visual stimulation of increasing complexity and in patients with homonymous hemianopsia (see text). Baseline values (mg glucose/min/100 g brain) in the eyes-closed state were 8.40 for the PVC and 6.66 for the AVC. Data obtained from Phelps et al. (1981b)

Visual stimulation	PVC(%)	AVC(%)
Complex (park)	+45.1 ± 7.2	+58.9 ± 9.8
Two-eye checkerboard	+27.6 ± 6.5	+26.7 ± 9.3
One-eye checkerboard	+17.4 ± 3.3	+21.9 ± 4.9
White light	+11.5 ± 4.1	+6.1 ± 2.8
Eyes-closed controls	0	0
Homonymous hemianopsia	-18.3 ± 8.1	-33.5 ± 9.2

on the functioning side, but no increase in LCMRGlc in the visual cortex ipsilateral to the lesion occurred during stimulation (as compared to the eyes closed state). The metabolic values of the involved visual cortex in these patients were lower than those found in unstimulated normal subjects with their eyes closed (Fig. 4, 5; Table 2).

Auditory Stimulation

Regardless of the stimulus, the highest relative (percentage of the cross-sectional mean) metabolic values were consistently in the transverse temporal cortex bilaterally, from 34% ± 8% to 40% ± 5% above the mean (Figs. 6–8). The second most active areas were the frontal regions; each test group had at least one frontal region greater than 30% above the hemispheric mean in the stimulated state. There was no consistent correlation between the patterns of LCMRGlc and the ear stimulated.

The distribution of the increased LCMRGlc differed between the left and right transverse temporal cortices. The area on the left was larger in five subjects, smaller in one subject and approximately equal in the others. In those subjects studied with the NeuroECAT during verbal stimulation, the area on the left side was more circular and more posteriorly situated than the diagonally placed and smaller rectangular band of activated tissue in the right transverse temporal cortex (Fig. 8) (Mazziotta et al., 1982b). These asymmetries are similar to those

previously described anatomically by Geschwind and Levitsky (1968).

Verbal Stimulation

Significant activations in the left frontal cortex (6% ± 8%) and bilaterally in the posterior (left 11% ± 7%, right 6% ± 10%) and transverse (17%–25%) temporal cortex were found in this group. A small but significant (6% ± 14%) activation also occurred in the left thalamus, but not in the right thalamus. Left > right metabolic asymmetries were demonstrated at the frontal, lateral occipital, and posterior superior temporal cortices when compared with symmetry. The cortical responses to verbal stimulation were consistently left-sided regardless of the ear stimulated (Mazziotta et al., 1982b).

Tonal Memory Stimulation

Significant increases above the hemispheric mean were measured in this group in the stimulated state for the right posterior superior (16% ± 12%) and middle (27% ± 6%) temporal cortices. Two subgroups were identified by the patterns of metabolic activity and by their response to a poststudy interview. One group ($n = 5$) reported no specific strategy in comparing identical or different tone sequences or said that they had mentally “resung” the tone sequence. These subjects had right > left asymmetries and activations (Fig. 6) (Mazziotta et al., 1982b). This was true for all measured frontal lobe regions as well as the parietal-temporal cortex. The second subgroup ($n = 3$) reported using a highly structured system of visual imagery (e.g., “visualizing a frequency histogram” or “seeing the notes on a musical scale”) to accomplish the tone sequence discrimination task. One of these subjects was a professional musician. In this subgroup, left > right metabolism occurred in the posterior superior temporal cortex and no right > left asymmetries were demonstrated (Fig. 6). Changes in left/right LCMRGlc values were a function of the strategies rather than the content (tonal memory test in all) or side of stimulation (Mazziotta et al., 1982b).

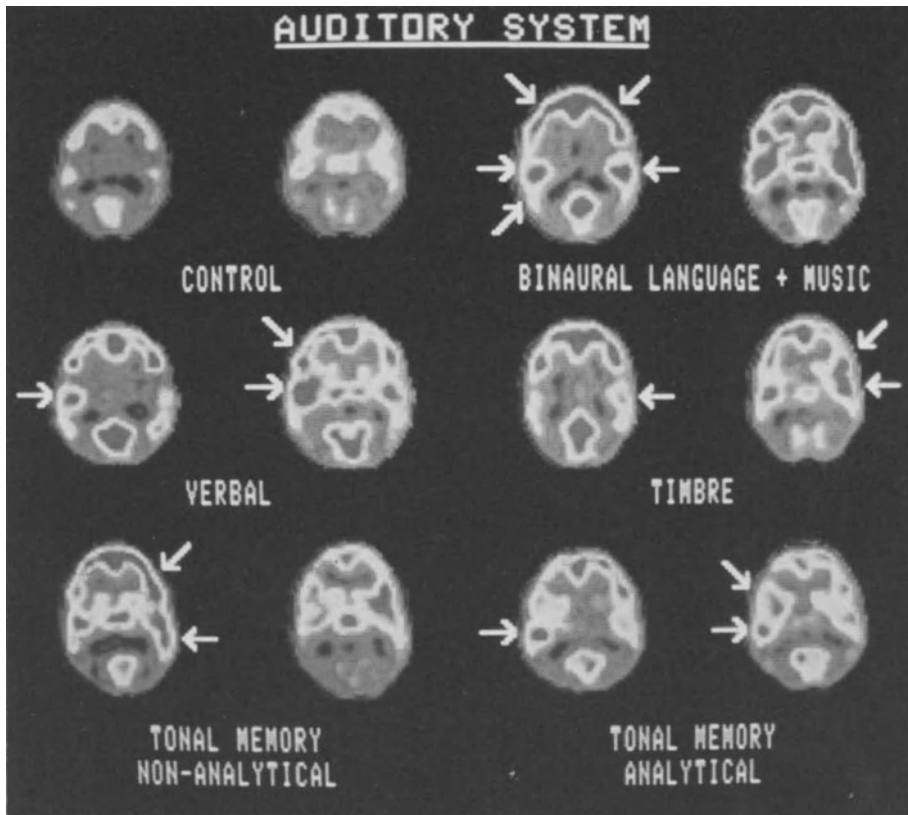


Fig. 6. Composite set of examples for individual subjects imaged in a variety of states of auditory stimulation. Note that the areas of asymmetry correlate with the stimulus content and/or the analysis strategy of the subjects. No correlation was found between the ear of stimulation and the metabolic response. While the complex nature of the auditory stimuli employed in this study resulted in complex metabolic responses, it also demonstrated the feasibility of studying primary and higher order central processing of auditory stimuli using PCT techniques. Note in particular the bilateral activation of the frontal and temporal cortical zones with combined language and music stimulation, whereas verbal stimuli produced

predominantly left-sided asymmetries and activations. Chords (timbre) produced predominantly right-sided asymmetries and activations. Results of stimulation with the tonal memory test produced two subgroups which differed by their analysis schemes in interpreting the auditory information. Analytical individuals who used visual imagery and/or were musically sophisticated had left-sided activations and asymmetries of the posterior temporal zone. Musically naive individuals who used subjective strategies without visual imagery had predominantly right-sided asymmetries and activations similar to subjects who listened to pairs of chords (timbre). (Mazziotta et al., 1982b)

Timbre (Chord) Stimulation

The most generalized right > left metabolic asymmetries occurred in this group. No subject demonstrated local or generalized left-sided relative hypermetabolism. Right > left asymmetries occurred in all three frontal regions examined, as well as diffusely in the posterior temporal and temporal-occipital regions. Activations occurred in the right superior posterior

temporal cortex ($10\% \pm 7\%$) and bilaterally in the temporal-parietal regions (left $16\% \pm 17\%$, right $22\% \pm 23\%$) (Mazziotta et al., 1982b).

Binaural Verbal and Nonverbal Stimulation

These subjects demonstrated diffuse frontal-temporal increases in metabolic activity. Because of the small number of subjects, no

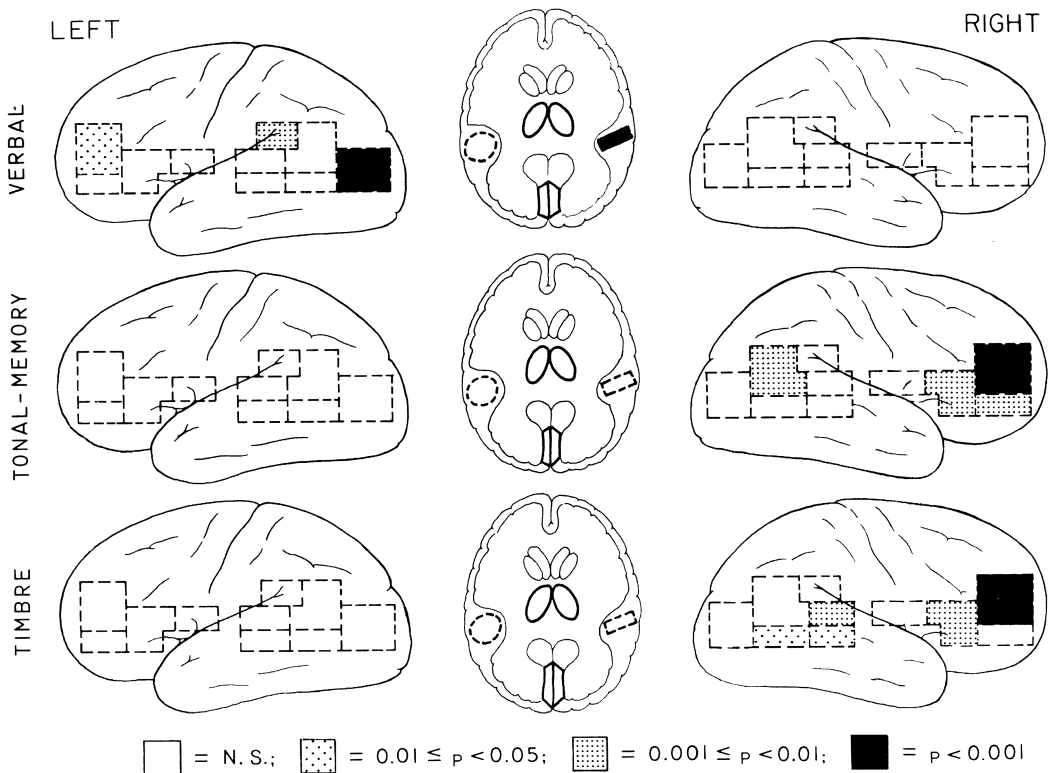


Fig. 7. Significant left/right metabolic asymmetries for each auditory test state. The degree of asymmetry is illustrated by the magnitude of its statistical significance. The percentage difference in LCMRGlc of these asymmetries is listed in Table 3. Note the tim-

bre test had the most consistent unilateral (i.e., right > left) asymmetry. The verbal stimuli produced more diffuse activations and therefore only a few regions in the left hemisphere were statistically asymmetric. (Mazziotta et al., 1982b)

Table 3. Percentage asymmetry of metabolic activity in normal subjects who received verbal and nonverbal auditory stimulation. These values were calculated as left-right/right and represent the significant asymmetries in each test group. Region numbers correspond to the anatomical regions depicted in Fig. 1. The errors are standard deviations. Values are tabulated under the hemisphere having the higher LCMRGlc. No significant asymmetries were found for the hemispheric means or regions 12 and 13 in any test state. The analytic subgroup of the tonal memory subjects had only one significant asymmetry, located at region 6 on the left. All values are significant at the $p < 0.05$ level or better. Data taken from Mazziotta et al. (to be published b)

Region	Tonal memory					
	Verbal		Nonanalytic		Timbre	
	Left	Right	Left	Right	Left	Right
1	3.0 ± 0.03			6.12 ± 0.12		7.25 ± 0.32
2				5.45 ± 0.47		
3				4.92 ± 0.19		6.85 ± 0.52
4						
5	9.4 ± 1.93					
6						13.3 ± 0.35
7						5.47 ± 0.24
8				4.30 ± 0.50		
9						6.88 ± 0.27
10	15.7 ± 0.08					
11						3.54 ± 0.01

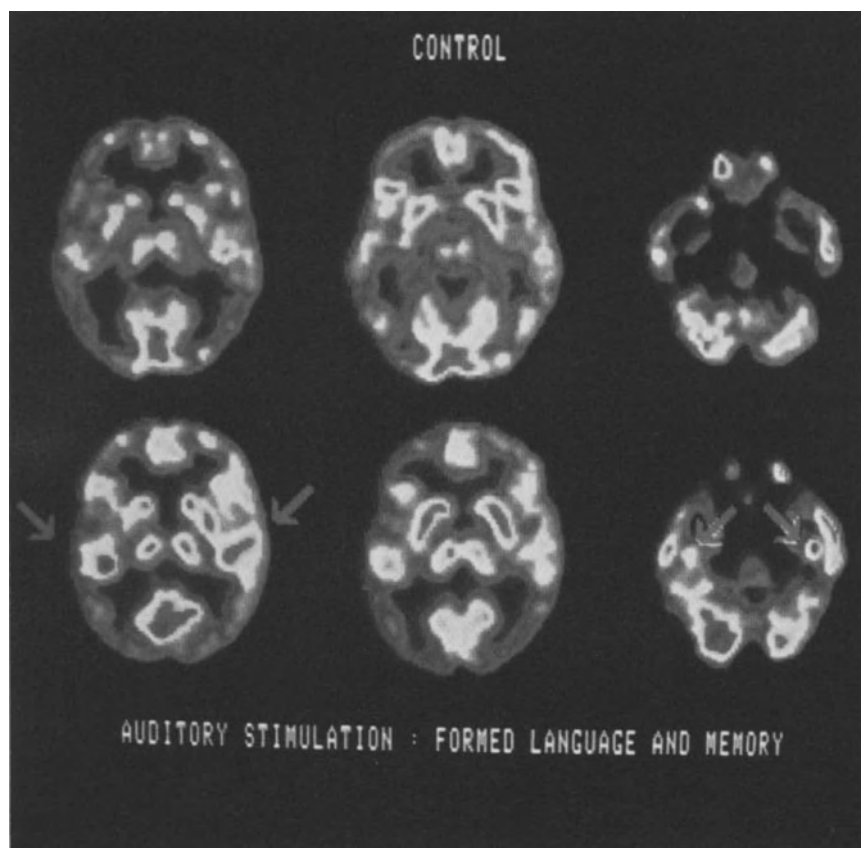


Fig. 8. High-resolution PCT-FDG images of cerebral glucose metabolism obtained with the NeuroECAT. The control subject was imaged with ears plugged and covered with soundproof headphones; another subject listened to a Sherlock Holmes story. Note that in the stimulated state the area of increased metabolic activity in the transverse temporal cortex appears to be distributed in an asymmetric fashion between the left and right hemispheres. In the left hemisphere this area is more circular, larger, and encompasses two tomographic planes. In addition, the area is more posteriorly situated on the left than the small rectangular band of activated tissue seen in the right

hemisphere. These asymmetries closely parallel known anatomical asymmetries for Heschl's gyri and the planum temporale. Note also the activation of the hippocampus only in the stimulated state (*bottom right*). This structure was not visualized in the control state, although it was consistently activated in the stimulated state using high-resolution PCT. It is unclear whether activation of the hippocampus reflects auditory tasks in general, specific analysis strategies in particular, or some aspects of memory and learning employed by these subjects to perform well on a poststimulus examination. (Mazziotta et al., 1982b)

quantitative analysis was performed. A quantitative left-right symmetry was obtained throughout the tomographic images (Fig. 6).

Discussion

Sensory stimulation and deprivation studies demonstrate the feasibility of using PCT to examine the brain's functional response to phy-

siological stimuli. They also reveal that small changes in ambient conditions can have dramatic effects on the PCT image and that "resting" states are actually misnomers (Mazziotta et al., 1982b). Physiological inputs or their absence must therefore be defined and controlled in any study attempting to examine the functional response of the brain to specific stimulation and in all states of pathological alteration.

The data obtained in this study also demonstrate that intersubject variability reduces the potential statistical significance of a given physiological manipulation. This situation is improved by comparing control and stimulated states. In addition, the use of left/right asymmetries serves as a very stringent and relatively invariable (left/right = 1.01 ± 0.03) (Mazziotta et al. 1981a; Lebrun-Grondie et al., to be published) indicator of activations and/or abnormalities in the metabolic landscape. Specific features of each of the paradigms examined are of interest, and these will be discussed separately.

Sensory Deprivation

The left > right metabolic asymmetries noted in the both-closed group could have four possible causes; they could be artifactual, anatomical, or functional, or arise from a combination of these factors. While we feel artifactual asymmetries have been ruled out (Mazziotta et al., 1982a), anatomical asymmetries have been demonstrated with regard to the size, shape, and orientation of a number of human cerebral structures, particularly in the perisylvian areas. In addition to these morphological asymmetries, tissue assays have disclosed biochemical asymmetries in both the temporal cortex and the thalamic nuclei.

It is possible that different amounts of gray matter in the left and right hemisphere could produce asymmetric partial volume effects (Hoffman et al. 1979; Mazziotta et al. 1981b) that could be misinterpreted as metabolic asymmetries. If these size differences were below the spatial resolution of the tomograph, one might not see the asymmetry but it would be manifested by differences in the measured metabolic rate (Mazziotta et al. 1981b).

Functional asymmetries have been well described in patients having a wide variety of lesions, in commissurotomy cases as well as from the results of intraoperative cortical stimulations (Heilman 1979; Heilman and Van den Abell 1979). While no specific functional explanation for the left > right metabolic asymmetries could be determined from this work, possible explanations include (1) selective tuning of language areas in the left hemisphere for potentially meaningful auditory in-

formation, (2) cerebral dominance for consciousness, (3) verbally based cognitive processes revealed only at low overall metabolic rates, (4) reduced right hemisphere surveillance and vigilance for sensory data during maximal sensory deprivation, and (5) combinations of these factors. Other functional explanations for our results are certainly possible and the above hypotheses merely represent an attempt to offer initial speculations for these observed asymmetries.

The relative frontal hypermetabolism correlates with similar findings demonstrated by a large number of investigators using ^{133}Xe to measure cerebral blood flow (Ingvar 1979). The fact that the degree of frontal hypermetabolism can be influenced by the ambient test conditions is important and indicates that studies which express findings as ratios of frontal to posterior cortical metabolism must interpret such data with regard to the test conditions at the time of the study.

Visual Stimulation Studies

Initial PCT studies of the metabolic response of the visual system employed both full- and half-field stimuli (black and white lines) (Reivich et al. 1979a). With full field stimulation, increases of 23% in the metabolic rate were reported for the visual cortex. Half-field stimulations resulted in asymmetries of $8\% \pm 3\%$ (Greenberg et al. 1981) and 15%–20% (Reivich et al. 1979a), with the higher value in the visual cortex contralateral to the field of stimulation. Visual stimulation in the present studies correlated visual scene complexity with metabolic response (Phelps et al. 1981a, b). The results demonstrate the following stimulus-metabolic response characteristics (Phelps et al. 1981a, b) for the primary and associative visual cortices: (1) increasing LCMRGlc with increasing stimulus complexity; (2) equal cortical functional inputs from each eye to each visual cortex, as demonstrated by metabolic symmetry in all test states; and (3) excellent clinical-PCT correlations of visual pathway lesions. Studies in patients with homonymous hemianopsia demonstrate the ability of PCT to reveal distant effects of chronic lesions resulting from interruptions of neural projection systems.

Auditory Stimulation

Auditory stimulation with verbal and nonverbal material results in activations and asymmetries that correspond to the content of the material and in some cases to the strategy employed in its interpretation (Mazziotta et al., 1982b). While the patterns of activations and asymmetries are complex (Figs. 6, 7), a consistently different pattern of responses was found within each subgroup. Details of these findings and their anatomical relationship to human lesion studies and animal experimentation have been previously described (Mazziotta et al., 1982b).

The stimuli used for these studies were complex and required the subjects to use multiple cognitive functions. This makes the interpretation of the specific relationship of the stimulus-cognitive processes to metabolic changes impossible. In broad terms, the verbal stimuli were acoustically complex, sequential, and symbolic in content. They resulted in predominantly left-sided activations and asymmetries. The tonal memory test had less acoustic complexity but contained sequencing information. The timbre test was composed of single chords of harmonic composition without sequences. The results demonstrated left > right asymmetries in metabolic activity for verbal stimuli and right > left changes for the timbre test. The tonal memory test was nonverbal but did contain specific temporal sequencing. Subjects who were musically sophisticated and/or used an analytic strategy consisting of visual imagery had left posterior temporal relative hypermetabolism and reductions in diffuse right-sided activations and asymmetries observed in musically naive subjects who used subjective nonanalytic strategies without visual imagery (Mazziotta et al., 1982b). Similar group differences have been found by Bever and Chiarello (1974) when testing musicians and nonmusicians with dichotically presented nonverbal material.

The fact that acoustically identical stimuli (tonal memory test) produced different metabolic responses (determined by strategy) suggests that hemispheric specialization can be varied not only by the stimulus parameters themselves but also by the cognitive strategy of the listener. The differences in metabolic pat-

terns between the analytic and nonanalytic subgroups represent different cognitive states, since the input and motor output were identical in both groups. These subjects were separable by the differences in their LCMRGlc and the highly stereotyped approach they used to solve the tonal memory task. Such hemispheric specialization does not necessarily signify unique properties of one hemisphere, but merely an advantage of one hemisphere over the other for a specific task in certain situations. Issues such as these await elucidation, but the present study demonstrates that selective auditory experiments using PCT are feasible.

Conclusions

The results of these studies demonstrate that PCT has the ability to examine stimulus-metabolic response characteristics of the human brain. Such investigations are not only feasible for the study of basic cortical responses, but also, as evidenced by the tonal memory test results, have the capacity to examine complex issues of higher cortical function.

The sensory deprivation studies demonstrate that cerebral metabolic responses are extremely sensitive to ambient test conditions and must be controlled in any PCT paradigm. The small magnitude of change from control to stimulate states indicates the necessity of carefully designed and accurate studies to reduce intersubject variability and maximize the reproducibility of data. The use of left-right asymmetries as well as control-stimulation paired studies will help in this regard. Rapidly repeatable measurements using continuous inhalation or intravenous injection of ^{15}O -labeled compounds to determine cerebral blood flow and oxygen metabolism would allow multiple studies to be performed within one test session, thereby minimizing day-to-day variability. In addition, studies where the experimental measurement period is reduced should minimize non-stimulus-related brain activity.

Since most of our knowledge of human brain function has been derived from lesion studies where large segments of the neuronal apparatus has been damaged, these findings may not reflect the normal state of brain physiology. Noninvasive PCT studies using physiologic sti-

muli may produce an entirely new set of rules to use in exploring normal brain function. It is not necessarily true that observations gained over the last century in patients will be wholly substantiated by noninvasive studies in normal subjects. In fact, the expectation may be the reverse. Whatever the outcome, PCT should provide new, previously unobtainable information about how the normal brain operates.

Acknowledgements. The authors wish to thank Dr. Norman S. MacDonald and the cyclotron staff and Dr. Jorge Barrio and his chemistry staff for preparation of the compounds used in this study; Joan Miller, Francine Aguillar, Ron Sumida, and Lawrence Pang for their technical assistance; Lee Griswold for the illustrations; and Maureen Kinney for preparation of the manuscript. This work was partially supported by DOE contract DE-AMO3-76-SF00012 and NIH grants ROI-6M-248389-01 and POI NS-15654-01. Dr. Mazziotta is the recipient of teacher/investigator award 1K07-0058801-NSBA from the NINCDS.

References

1. Baron JC, Steinling M, Tanaka T, Cavalheiro E, Soussaline F, Collard P (1981) Quantitative measurement of CBF, oxygen extraction fraction (OEF) and CMRO₂ with ¹⁵O continuous inhalation techniques and positron emission tomography (PET): experimental evidence and normal values in man. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S5-S6
2. Barrio JR, MacDonald NS, Robinson GD et al. (1981) Remote, semiautomated production of F-18-labeled 2-deoxy-2-fluoro-D-glucose. *J Nucl Med* 22: 372-375
3. Bever TG, Chiarello RJ (1974) Cerebral dominance in musicians and nonmusicians. *Science* 185: 537-539
4. Finklestein S, Alpers NA, Ackerman RH et al. (1980) Positron imaging of the normal brain: regional patterns of cerebral blood flow and metabolism. *Trans Am Neurol Assoc* 105: 8-10
5. Frackowiak SJ, Lenzi GL, Jones T, Heather JD (1980) Quantitative measurement of regional cerebral blood flow and oxygen metabolism in man using ¹⁵O and positron emission tomography: theory, procedure and normal values. *J Comput Tomogr* 4: 727-736
6. Geschwind N, Levitsky W (1968) Human brain: left-right asymmetries in temporal speech region. *Science* 161: 186-187
7. Greenberg J, Reivich M, Alavi A, Hand P, Rosenquist A, Rintelmann W, Stein A, Tusa R, Dann R, Christman D, Fowler J, MacGregor B, Wolf A (1981) Metabolic mapping of functional activity in human subjects with the [¹⁸F]fluorodeoxyglucose technique. *Science* 212: 678-680
8. Heilman KM (1979) Neglect and related disorders. In: Heilman KM, Valenstein E (eds) *Clinical Neuropsychology*. Oxford University Press, New York pp 268-307
9. Heilman KM, Van Den Abell T (1979) Right hemisphere dominance for mediating cerebral activation. *Neuropsychologia* 17: 315-321
10. Hoffman EJ, Huang SC, Phelps ME (1979) Quantitation in positron emission computed tomography: 1. Effect of object size. *J Comput Assist Tomogr* 3: 299-308
11. Hoffman EJ, Phelps ME, Huang SC, Kuhl DE, Crabtree M, Burke M, Burgess S, Keyser R, Highfill R, Williams C (1981) A new tomograph for quantitative positron emission computed tomography of the brain. *IEEE Trans Nucl Sci* NS-28: 99-103
12. Huang SC, Phelps ME, Hoffman EJ, Sideris K, Selin CJ, Kuhl DE (1980) Non-invasive determination of local cerebral metabolic rate of glucose in man. *Am J Physiol* 238: E69-E82
13. Ido T, Wan CN, Casella V, Fowler JS, Wolf AP, Reivich M, Kuhl DE (1978) Labeled 2-deoxy-2-fluoro-D-glucose, 2-deoxy-2-fluoro-D-mannose, and ¹¹C-2-deoxy-2-fluoro-D-glucose. *J Labelled Compd Radiopharm* 24: 174-183
14. Ingvar DH (1979) "Hyperfrontal" distribution of cerebral gray matter flow in resting wakefulness; on the functional anatomy of the conscious state. *Acta Neurol Scand* 60: 12-25
15. Kuhl DE, Phelps ME, Kowell AP, Metter EJ, Selin C, Winter J (1980) Effects of local cerebral metabolism and perfusion: mapping by emission computed tomography of ¹⁸FDG and ¹³NH₃. *Ann Neurol* 8: 47-60
16. Lebrun-Grandie P, Baron JC, Soussaline F, Loch C, Sastre J, Bousser MG (to be published) Regional blood flow, oxygen utilization and the coupling between the two in the normal human brain: a study with positron computed tomography and oxygen-15. *Arch Neurol*
17. Mazziotta JC, Phelps ME, Miller J, Kuhl DE (1981a) Tomographic mapping of human cerebral metabolism: normal unstimulated state. *Neurology* 31: 503-516
18. Mazziotta JC, Phelps ME, Plummer D, Kuhl DE (1981b) Quantitation in positron computed tomography: 5. Physical-anatomical effects. *J Comput Assist Tomogr* 5: 734-743
19. Mazziotta JC, Phelps ME, Carson RE, Kuhl DE (1982a). Tomographic mapping of human cerebral metabolism: sensory deprivation. *Ann Neurol* 12: 435-444, 1982
20. Mazziotta JC, Phelps ME, Carson RE, Kuhl DE

- (1982b) Tomographic mapping of human cerebral metabolism: auditory stimulation. *Neurology* 32: 921–937
21. Mazziotta JC, Phelps ME, Meadors AK, Ricci A, Winter J, Bentson JR (1982c) Anatomical localization schemes for use in positron computed tomography using a specially designed head holder. *J Comput Assist Tomogr* 6: 848–853
 22. Phelps ME, Hoffman EJ, Huang SC, Kuhl DE (1978) ECAT: A new computerized tomographic imaging system for positron emitting radiopharmaceuticals. *J Nucl Med* 19: 635–647
 23. Phelps ME, Huang SC, Hoffman EJ, Selin C, Sokoloff L, Kuhl DE (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with (F-18) 2-fluoro-2-deoxy-D-glucose: validation of method. *Ann Neurol* 6: 371–388
 24. Phelps ME, Kuhl DE, Mazziotta JC (1981a) Metabolic mapping of the brain's response to visual stimulation: studies in humans. *Science* 211: 1445–1448
 25. Phelps ME, Mazziotta JC, Kuhl DE, Nuwer M, Packwood J, Metter J, Engel J Jr (1981b) Tomographic mapping of human cerebral metabolism: visual stimulation and deprivation. *Neurology* 31: 517–529
 26. Phelps ME, Mazziotta JC, Huang SC (1982) Study of cerebral function with positron computed tomography. *J Cereb Blood Flow Metabol* 2: 113–162
 27. Reivich M, Greenberg J, Alavi A, Christman D, Fowler J, Hand P, Rosenquist A, Rintelmann W, Wolf A (1979a) The use of the ^{18}F -fluoro-deoxyglucose technique for mapping of functional neural pathways in man. *Acta Neurol Scand* 60 [Suppl 72]: 198–199
 28. Reivich M, Kuhl D, Wolf A, Greenberg J, Phelps ME, Ido T, Casella V, Fowler J, Hoffman E, Alavi A, Som P, Sokoloff L (1979b) The (^{18}F)-fluoro-deoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137

Measurement of Local Cerebral Glucose Metabolism: Application to the Study of Stroke

By M. Reivich, J. H. Greenberg, M. J. Kushner, and A. Alavi

Introduction

The measurement of regional cerebral blood flow (CBF) in humans has greatly increased our understanding of cerebral hemodynamics under normal and pathologic conditions. The introduction of the ^{18}F -fluorodeoxyglucose (^{18}F -FDG) method (Reivich et al. 1979) now enables us to measure regional cerebral glucose metabolism (LCMRgl) in humans. This has allowed information concerning LCMRgl to be obtained in a number of conditions, including normal physiological stimuli (Reivich et al. 1978a; Greenberg et al. 1981; Phelps et al. 1981), stroke (Kuhl et al. 1980b), seizures (Kuhl et al. 1980a), brain tumors (DiChiro et al. 1981), schizophrenia (Farkas et al. 1980), and senile dementia (Ferris et al. 1980; Alavi et al., to be published). In the majority of the studies with ^{18}F -FDG, single determinations of glucose consumption were made. In the few cases where repeat determinations were made in the same individual (Ferris et al. 1980; Kuhl et al. 1980a), at least a day between studies was required for the decay of ^{18}F to background levels. This required repeat vascular puncture for the administration of tracer and collection of blood samples.

Since a statistical advantage would result from making the control and experimental measurement in the same subject over a relatively short period of time, we have developed a method utilizing ^{14}C -deoxyglucose to measure LCMRgl (Reivich et al. 1982).

Measurement of Local Cerebral Glucose Consumption

The ^{18}F -FDG technique for the measurement of LCMRgl in humans (Reivich et al. 1979) is based upon the autoradiographic technique using ^{14}C -deoxyglucose developed by Sokoloff et

al. (1977) for the determination of LCMRgl in animals. [^{18}F]-2-Deoxy-2-fluoro-D-glucose is used as a tracer for the exchange of glucose between plasma and brain and its phosphorylation by hexokinase in the tissue. A mathematical model and derived operational equation are used which enable LCMRgl to be calculated in terms of measurable variables. This model considers that glucose and fluorodeoxyglucose share and compete for a common transport carrier between plasma and brain tissue. Once transported into a common homogeneous precursor pool, glucose and fluorodeoxyglucose either compete for a common carrier for transport back from brain to plasma or for hexokinase for phosphorylation to their respective hexose-6-phosphates. In this model it is assumed that fluorodeoxyglucose-6-phosphate, once formed, is not further metabolized and is trapped in the tissues. This assumption, however, is not necessary and, as shown by Phelps et al. (1979), the model can be extended to include the presence of phosphatase activity in the brain if required in a particular instance. However, since phosphatase activity is in very low concentrations in the brain this term can be omitted for studies performed over a period of 60 min. An intravenous bolus of ^{18}F -FDG is given and the arterial specific activity monitored for a predetermined period of from 30 to 60 min. Starting at 30 min, the activity in a series of sections through the brain is determined with three-dimensional resolution by an emission tomographic scanner.

This technique has been used to determine the rates of glucose consumption in various regions of the brain in humans (Reivich et al. 1979; Mazziotta et al. 1981). In the resting state there are only small differences in the metabolic rate of a given structure in the two hemispheres. The ratios of these left/right metabolic rates for a number of structures, with their coefficients of variation, are shown in Table 1. Because of the relatively small coefficients of va-

Table 1. ^{18}F -Fluorodeoxyglucose: L/R ratio

	Mazziotta et al. (1981)		Present work	
	Mean	Coefficient of variation (%)	Mean	Coefficient of variation (%)
Frontal cortex	1.00	5.0	1.01	9.9
Auditory cortex	1.02	5.9	0.96	5.2
Visual assoc. cortex	0.97	6.2	0.99	7.1
Primary visual cortex	1.02	4.9	1.00	4.0
Thalamus	1.01	4.0	1.01	5.0
Caudate nucleus	1.02	7.8	1.03	3.9

Table 2. ^{18}F -Fluorodeoxyglucose: LCMRgl (mg/100 g/min)

	Mazziotta et al. (1981)		Present work	
	Mean	Coefficient of variation (%)	Mean	Coefficient of variation (%)
Frontal cortex	7.75	24.4	6.5	24.6
Auditory cortex	7.94	26.4	6.1	21.3
Visual association cortex	6.66	25.8	5.4	22.2
Primary visual cortex	8.40	23.8	6.1	21.3
Thalamus	6.60	19.5	5.7	22.8
Caudate nucleus	6.93	23.7	6.1	23.0

riation it is possible to detect small changes in glucose consumption when a unilateral effect is produced. This enables the structure on one side to serve as a control within the same subject. The following studies of visual and auditory stimuli were designed to use this paradigm, and illustrate the relatively small changes that can be reliably measured with this technique.

In the visual study (Greenberg et al. 1981), either the left ($n = 4$) or right ($n = 6$) visual hemifield was stimulated. After positioning the subject in the tomograph, a plexiglass hemisphere (diameter, 18 in.) was placed 70 cm from him. He was instructed to fixate on a small light located at the center of the hemisphere, which was dimmed at random. The luminance of the fixation stimulus was adjusted so as to be detectable only by foveal vision. The subject's hemifields could thus be defined relative to the hemisphere and stimulation could be limited to the desired hemifield. Subjects reported more than 95% of the dimming events, indicating good visual fixation. One half of the hemisphere was painted black and the space around the painted side darkened with black cloth to eliminate visual input from that hemifield. The stimulus consisted of a well-illuminated, slowly

moving, high-contrast black-and-white pattern of small lines at various orientations, as well as abstract color images presented into one visual hemifield. The subjects wore earplugs.

The auditory system was studied in six subjects with normal hearing who listened to a tape-recorded factual story presented through earphones to only one ear (left ear, $n = 3$; right ear, $n = 3$). Attentiveness to the story was assessed by testing the subject's recall. These subjects were also blindfolded. Six subjects who were blindfolded and wore earplugs acted as controls for all the studies.

The visual stimulus caused the visual cortex contralateral to the stimulated hemifield to become $8\% \pm 3.0\%$ more active than the ipsilateral visual cortex. The asymmetry is significant in comparison with the controls [$t(14) = 4.06$, $p < 0.01$], who showed a left-right asymmetry of only $0.5\% \pm 3.0\%$.

The monaurally presented auditory stimulation elevated the metabolic rate in the temporal cortex contralateral to the stimulated ear. This cortex had a metabolic rate of $7\% \pm 2.5\%$ higher than the ipsilateral temporal cortex. This asymmetry is significant in comparison with the controls [$t(8) = 6.02$, $p < 0.001$], who

Table 3. ^{11}C -Deoxyglucose: repeated measures

Structure	Gray matter	
	Mean % difference	Coefficient of variation of repeat measures (%)
Frontal pole	− 2.7	6.5
Frontal gyrus	− 0.8	8.3
Somatomotor cortex	+ 2.0	7.9
Auditory cortex	− 2.3	6.6
Caudate nucleus	+ 1.8	7.7
Primary visual cortex	+ 1.3	8.7
Visual associative cortex	− 5.0	6.9
Thalamus	− 1.0	5.2
White matter		
Frontal white	− 5.0	10.2
Occipital white	+ 1.0	14.0
Corpus callosum	+ 0.6	9.7

showed a left-right asymmetry of only $1\% \pm 2\%$.

However, when it is necessary to compare one group of subjects with another, the changes in glucose metabolism need to be much larger in order to be detected, since the coefficients of variation of groups means are larger (Table 2). The ability to make repeat measurements in the same subject would reduce the variance due to intersubject differences, allowing smaller changes in LCMRgl to be detected with confidence. The use of ^{11}C -2-deoxy-D-glucose (^{11}C -DG) would enable this to be done, since the shorter half-life of ^{11}C (20.4 min) would allow repeat measurements to be made 2 h apart. For this reason we have investigated the use of ^{11}C -DG for the repeat measurement of LCMRgl (Reivich et al. 1982).

The ^{11}C -DG was prepared at Brookhaven National Laboratory as described previously (MacGregor et al. 1981). The radiochemical purity was 97%–99% as determined by thin-layer chromatography and the specific activity (at time of injection) 14–45 mCi/mg. The determination of LCMRgl was carried out in a series of eight normal male subjects aged 19–26 years. The subjects had their eyes open and ears plugged. Each subject was studied on two separate occasions 120 min apart. Thirty minutes after the injection of ^{11}C -DG, scanning for the determination of LCMRgl was initiated using

the PETT III scanner at Brookhaven National Laboratory.

The results of these repeat measurements of LCMRgl are shown in Table 3. The mean percentage difference between the repeat measurements varied from +0.6% in the corpus callosum to -5.0% in frontal white matter and the visual association cortex. The coefficients of variation ranged from 5.2% in the thalamus to 14.0% in the occipital white matter. The variation in repeated measures in the same subject is thus comparable to that of the L/R ratio of metabolic rates in a single measurement and significantly less than the variation of mean values for a given structure in a group of individuals. The pattern of CMRgl is thus relatively constant in a given individual over this interval of time when the conditions of the study are maintained constant. The ability to make repeat measurements in the same subject minimizes intersubject variability, allowing smaller changes in LCMRgl to be detected reliably.

Local Cerebral Glucose Consumption in Stroke

Animal studies of focal ischemia in a cat middle cerebral artery occlusion model (Ginsberg et al. 1977) have shown markedly heterogeneous alterations in LCMRgl. There was a consistent central zone of decreased glucose utilization within the basal ganglia and a zone of increased glucose utilization surrounding this central region (Fig. 1). In the overlying cortex, regions of decreased and increased glucose consumption were present in some animals. The areas of increased glucose utilization were thought to represent regions of anaerobic metabolism of glucose. Mild but significant depression of glucose consumption was also present in the contralateral, nonischemic cortex (Fig. 2). This depression of metabolism at a distance from a focus of brain ischemia was thought to correspond to the clinical phenomenon of diaschisis, or a depression of brain function in areas remote from a focal injury, first described by von Monakow (1914). A corresponding depression of cortical blood flow contralateral to the ischemic hemisphere (Fig. 3) has been observed in this same model

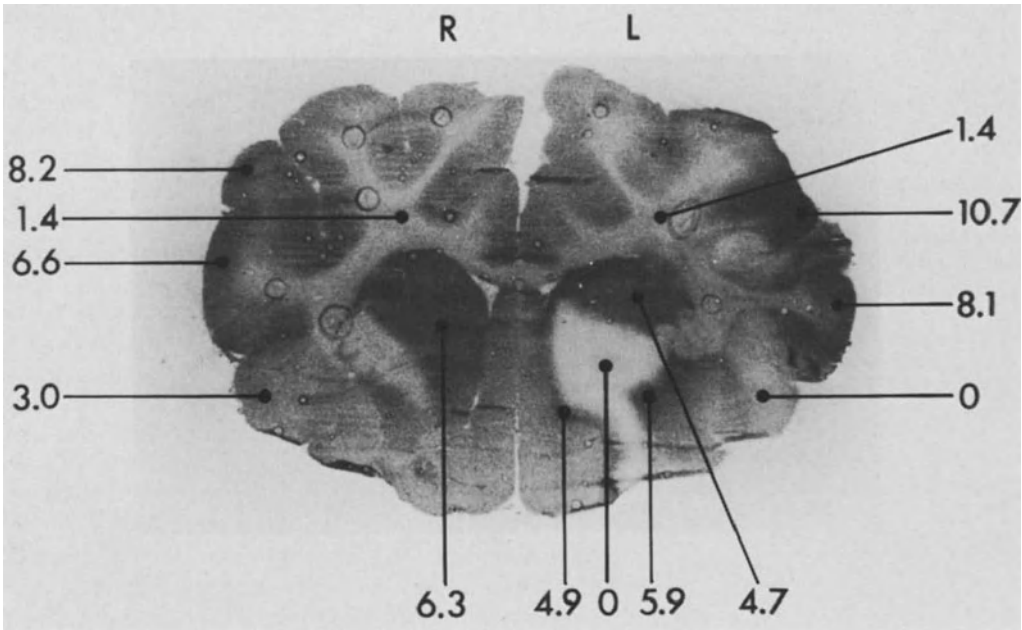
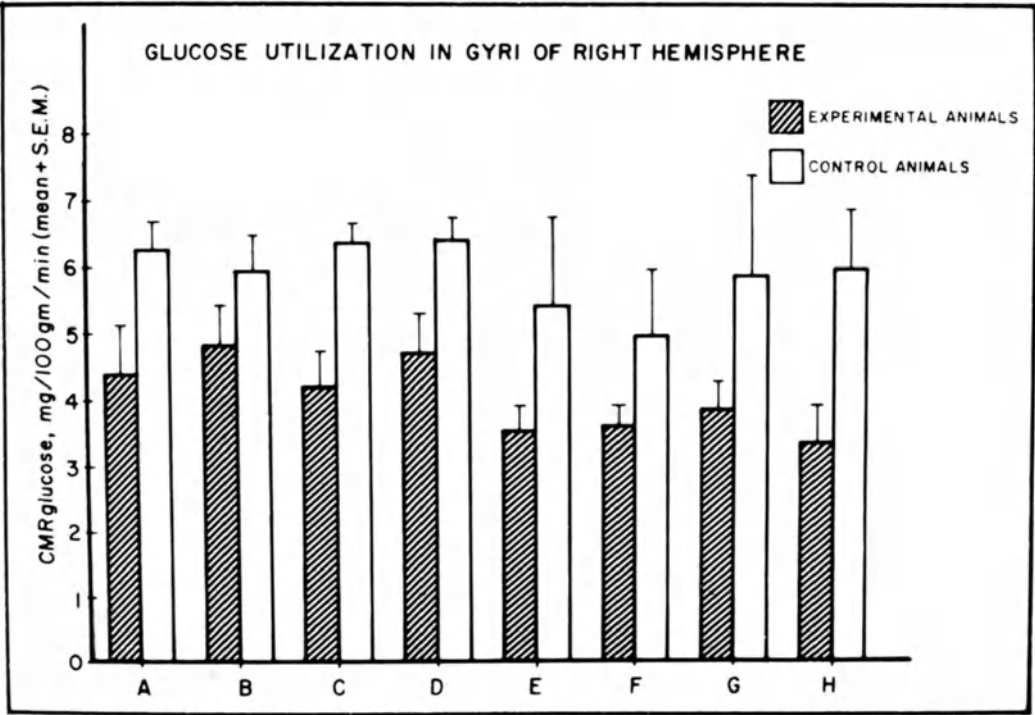


Fig. 1. ¹⁴C-deoxyglucose autoradiogram from a cat subjected to left middle cerebral artery and common carotid artery occlusion 90 min earlier. An irregular zone of depressed glucose utilization involving the caudate, internal capsule, and inferior frontotemporal cortex is present. This zone is ringed by a thin re-

gion of enhanced glucose utilization. The lateral portion of the cerebral cortex is diffusely affected as well, showing large zones of moderately depressed LCMRgl bordered by zones of normal or mildly elevated LCMRgl. (Ginsberg et al. 1977)



(Reivich et al. 1978b). This reduction in flow in the contralateral cortex was of the order of 38% and agrees quite well with the observed depression of glucose consumption of 31% in the contralateral cortex. There have been several reports of bilateral reduction in CBF in patients with unilateral cerebral infarction (Hoedt-Rasmussen and Skinhoj 1964; Skinhoj 1965; Meyer et al. 1970; Melamed et al. 1975; Slater et al. 1977). A depression of cerebral metabolism in the nonischemic hemisphere has also been observed in these patients (Meyer et al. 1970). The time course of the hemodynamic alterations in the nonischemic hemisphere has been examined by Slater et al. (1977). They ob-

served that the depression of regional CBF in the contralateral hemisphere was not maximal immediately, but progressed gradually during the 1st week post infarction (Fig. 4). These data suggest that acute neuronal depression secondary to disconnection from afferent stimulation cannot be the only cause of diaschisis, as has been postulated. Nonetheless, some of the remote effects of a focal lesion may be mediated transneuronally.

Similar abnormalities in LCMRgl have been observed in PET studies of patients with stroke. Regions of decreased glucose consumption and perfusion have been consistently observed even when the CT scan was normal in those re-

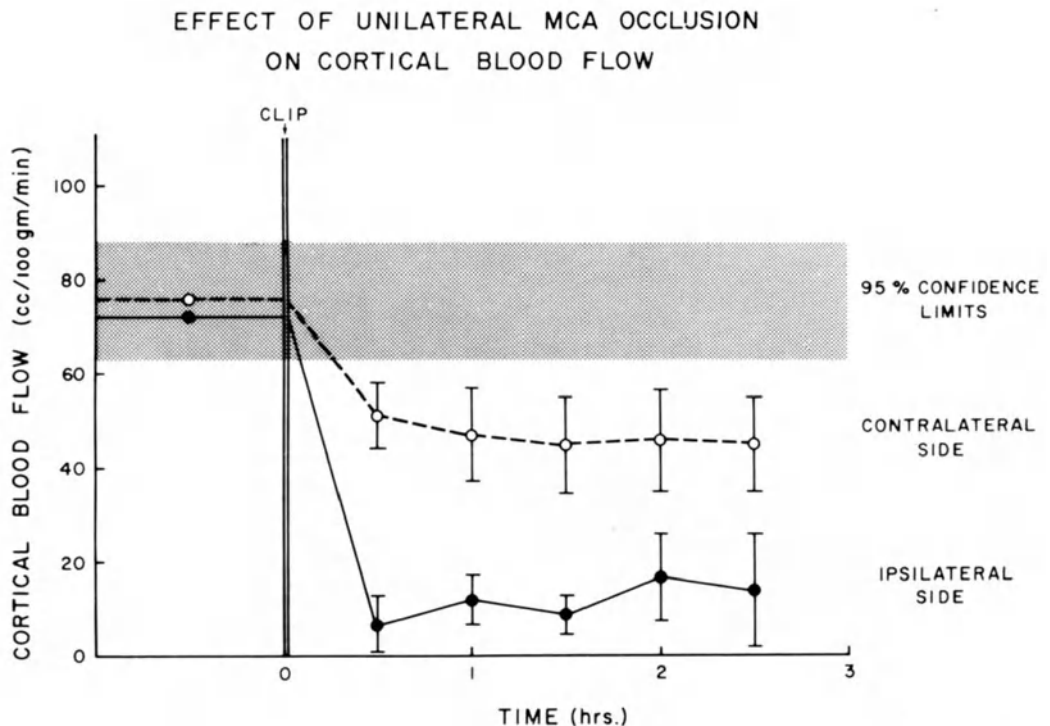


Fig. 3. Time course of change of blood flow in middle ectosylvian gyri in ischemic hemispheres (filled circles, $\pm$ SE) and nonischemic hemispheres (open

circles, $\pm$ SE) following left middle cerebral artery occlusion in the cat. (Reivich et al. 1978)

Fig. 2. LCMRgl values from eight representative cerebral gyri of the nonischemic hemisphere of experimental and control animals (mean $\pm$ SEM). The experimental group of cats had their left middle cerebral and common carotid arteries occluded 90 min earlier. For each gyrus, the mean LCMRgl of

the control group exceeds that of the experimental group. A, sigmoid gyrus; B, anterior suprasylvian gyrus; C, anterior ectosylvian gyrus; D, cingulate gyrus; E, lateral gyrus, medial portion; F, lateral gyrus, lateral portion; G, middle suprasylvian gyrus; H posterior sylvian gyrus. (Ginsberg et al. 1977)

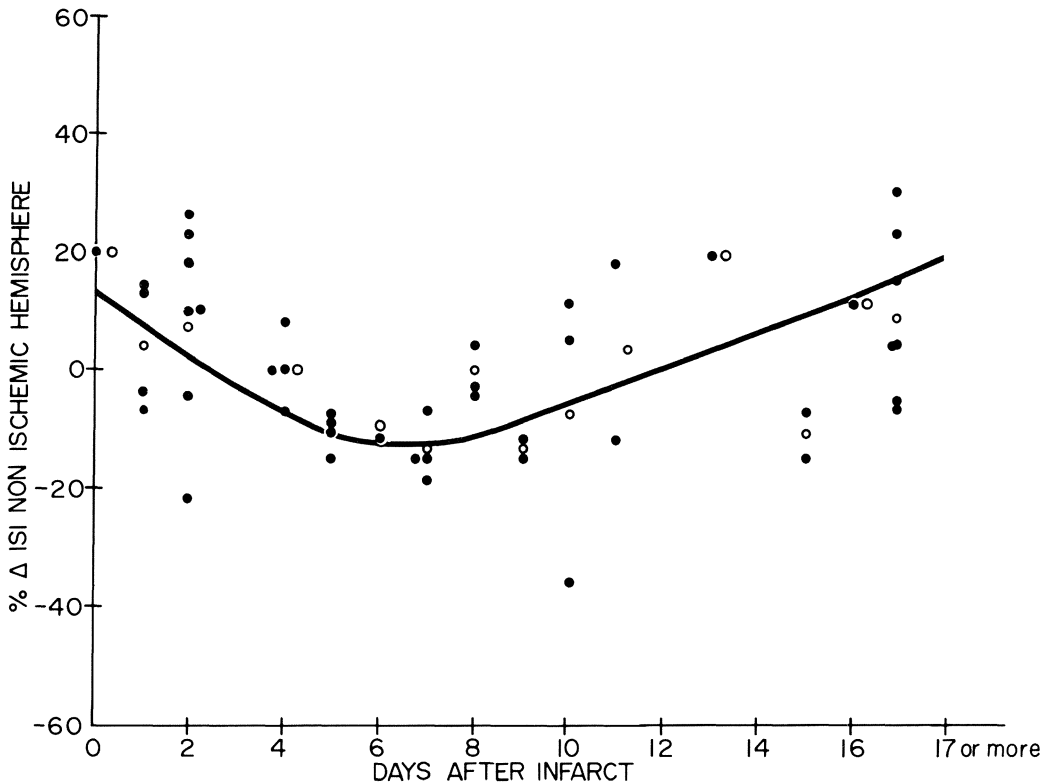


Fig. 4. Normalized flow in the nonischemic hemisphere as a function of time in patients with acute strokes. The *solid circles* are the individual data

points. The *open circles* represent the average values on each day. (*ISI*, initial slope index. Slater et al. 1977)

gions (Kuhl et al. 1980b). Areas of hyperemia in the infarct margins have been observed during the first 2 weeks following the stroke. This is often accompanied by a decrease in glucose consumption so that the flow appears to be in excess of the metabolic requirement, the so-called luxury perfusion syndrome. In one case reported by Kuhl et al. (1980b), studied within 1 week after a unilateral stroke, there was a 32% reduction in LCMRgl in the contralateral hemisphere, which had returned to normal when studied again 3 months later. This may represent the metabolic effects of diaschisis. Baron et al. (1981) observed a significant decrease in regional CBF and oxygen consumption in the cerebellar hemisphere contralateral to a supratentorial infarction, present only in patients who were studied less than 2 months after their strokes. They postulated that these effects were a manifestation of diaschisis.

We have observed a significant asymmetry of LCMRgl (> 10% difference) in a group of pa-

tients with acute unilateral cerebral hemispheric lesions (Fig. 5). When present, the hypometabolism occurred in the cerebellar hemisphere contralateral to the cortical lesion ($p < 0.05$ sign test). The likelihood of observing this crossed cerebellar hypometabolism seemed to be related to the extent of the hemispheric hypometabolism rather than to the degree of motor impairment. A well-defined cerebellar syndrome was not clinically apparent. The magnitude of the differences between the cerebellar hemisphere metabolic rates was significantly greater in these patients than in normal control subjects ($p < 0.05$, Wilcoxon sign rank test). These data indicate that cerebellar hemisphere metabolism can be remotely affected by lesions in the contralateral hemisphere, probably on the basis of transneuronal depression following interruption of cerebro-cerebellar pathways. This mechanism may underly such clinical syndromes as crossed cerebellar atrophy, ataxic hemiparesis, and Brun's ataxia.

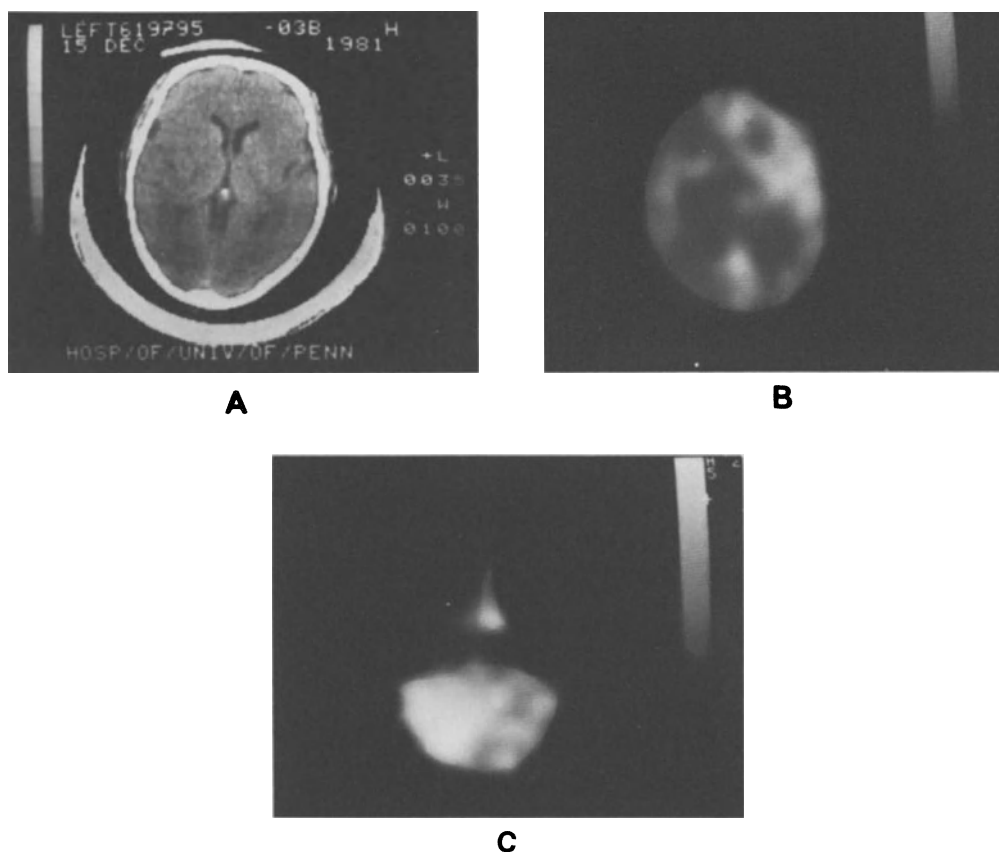


Fig. 5. The CT scan (*A*) was normal in this patient with a stroke in the distribution of the left middle cerebral artery. The PET scan, however, revealed significant reduction in glucose consumption in the left hemisphere (*B*). In addition, there was a signifi-

cant decrease in glucose metabolism in the contralateral cerebellar hemisphere (*C*). The patient had no symptoms referable to the cerebellar hypometabolism, which was thought to represent the metabolic concomitant of diaschisis

Correlative studies of the results of positron emission tomography (PET) scans and clinical course in acute stroke patients suggest that the metabolic alterations observed are a more consistent and reliable index of the severity of tissue injury than are the hemodynamic alterations present (Ackerman et al. 1981a, 1981b). Oxygen metabolism appeared to be uniformly depressed through out the evolution of the infarct, but CBF alterations evolved in a patchy, heterogeneous fashion. Areas of relative hyperemia and ischemia often coexisted. It appears that it may be possible to categorize acute strokes, on the basis of PET data, into those that have an ischemic lesion but are metabolically intact, those that are metabolically impaired but with viable tissue, and those with infarcted tissue. In the first instance regional flow

alone is depressed. In the second case both regional flow and oxygen metabolism are reduced, but the latter is less severely affected, so that the oxygen extraction fraction is increased in the area of injury. In the last case either flow and oxygen metabolism are both severely reduced, or the oxygen metabolism is more severely affected than flow so that regional oxygen extraction is depressed.

Further study is necessary to confirm these preliminary findings and to determine whether these metabolic data will prove to be helpful in the management of patients with acute strokes.

Acknowledgement. This work was supported in part by USPHS Program Project Grant NS-14867-04.

References

- Ackerman RH, Alpert NM, Correia JA, Finklestein S, Buonanno F, Davis SM, Chang JY, Brownall GL, Taveras JM (1981a) Importance of monitoring metabolic function in assessing the severity of a stroke insult (CBF: an epiphenomenon?). *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 502–503
- Ackerman RH, Correia JA, Alpert NM, Baron JC, Gouliamos A, Grotta JC, Brownell GL, Taveras JM (1981b) Positron imaging in ischemic stroke disease using compounds labeled with oxygen 15. *Arch Neurol* 38: 537–543
- Alavi, A, Ferris S, Wolf A, Christman D, Fowler J, MacGregor R, Farkas T, Greenberg J, Dann R, Reivich M (to be published) Determination of regional cerebral metabolism in dementia using F18 deoxyglucose and positron emission tomography. *Excerpta Medica*, Amsterdam
- Baron JC, Bousser MG, Comar D, Duquesnay N, Sastre J, Castaigne P (1981) Crossed cerebellar diaschisis: a remote functional depression secondary to supratentorial infarction in man. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 500–501
- DiChiro G, DeLaPaz R, Smith B, Kornblith P, Sokoloff L, Brooks R, Blasberg R, Cummins C, Kessler R, Wolf A, Fowler J, London W, Sever J (1981) ^{18}F -2-fluoro-2-deoxyglucose positron emission tomography of human cerebral gliomas. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 11–12
- Farkas T, Reivich M, Alavi A, Greenberg JH, Fowler JS, MacGregor RR, Christman DR, Wolf AP (1980) The application of ^{18}F -2-deoxy-2-fluoro-D-glucose and positron emission tomography in the study of psychiatric conditions. In: Passonneau JV, Hawkins RA, Lust WD, Welsh FA (eds) *Cerebral metabolism and neural function*. Williams and Wilkins, Baltimore pp 403–408
- Ferris SH, DeLeon MJ, Wolf AP et al. (1980) Positron emission tomography in the study of aging and senile dementia. *Neurobiol Aging* 1: 127–131
- Ginsberg MD, Reivich M, Giandomenico A, Greenberg JH (1977) Local glucose utilization in acute focal cerebral ischemia: Local dysmetabolism and diaschisis. *Neurology* 27: 1042–1048
- Greenberg JH, Reivich M, Alavi A, Hand P, Rosenquist A, Rintelmann W, Stein A, Tusa R, Dann R, Christman D, Fowler J, MacGregor B, Wolf A (1981) Metabolic mapping of functional activity in human subjects with the [^{18}F] fluorodeoxyglucose technique. *Science* 212: 678–680
- Hoedt-Rasmussen K, Skinhoj E (1964) Transneuronal depression of the cerebral hemispheric metabolism in man. *Acta Neurol Scand* 40: 41–46
- Kuhl DE, Engel J Jr, Phelps ME, Selin C (1980a) Epileptic patterns of local cerebral metabolism and perfusion in humans determined by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 348–360
- Kuhl DE, Phelps ME, Kowell AP, Metter EJ, Selin C, Winter J (1980b) Effects of stroke on local cerebral metabolism and perfusion: Mapping by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 47–66
- MacGregor RR, Fowler JS, Wolf AP et al. (1981) A synthesis of 2-deoxy-D-[^{11}C] glucose for regional metabolic studies: concise communication. *J Nucl Med* 22: 800–803
- Mazziotta JC, Phelps ME, Miller J, Kuhl DE (1981) Tomographic mapping of human cerebral metabolism: normal unstimulated state. *Neurology* 31: 503–516
- Melamed E, Lavy S, Portnoy Z (1975) Regional cerebral blood flow response to hypocapnia in the contralateral hemisphere of patients with acute cerebral infarction. *Stroke* 6: 503–508
- Meyer JS, Shinohara Y, Kanda T, et al. (1970) Diaschisis resulting from acute unilateral cerebral infarction. *Arch Neurol* 23: 241–247
- Phelps ME, Huang SC, Hoffman EJ, Selin C, Sokoloff L, Kuhl DE (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with (F18)2-fluoro-2-deoxy-D-glucose: Validation of method. *Ann Neurol* 6: 371–388
- Phelps ME, Kuhl DE, Mazziotta JC (1981) Metabolic mapping of the brain's response to visual stimulation: studies in humans. *Science* 211: 1445–1448
- Reivich M, Alavi A, Greenberg J, Fowler J, Christman D, Wolf A, Rosenquist A, Hand P (1978a) Metabolic mapping of functional cerebral activity in man using the ^{18}F -2-fluoro-2-deoxyglucose technique. *J Comput Assist Tomography* 2: 656
- Reivich M, Jones S, Ginsberg M, Slater R, Greenberg J (1978b) Regional hemodynamic and metabolic alterations in focal cerebral ischemia: studies of diaschisis. In: Silver A, Erecinska M, Bicher HI (eds) *Oxygen transport to tissue – III*. Plenum, New York pp 617–622
- Reivich M, Kuhl D, Wolf A, Greenberg J, Phelps M, Ido T, Casella V, Fowler J, Hoffman E, Alavi A, Som P, Sokoloff L (1979) The [^{18}F]Fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
- Reivich M, Alavi A, Wolf A, Greenberg JH, Fowler J, Christman D, MacGregor R, Jones SC, London J, Shiue C, Yonekura Y (1982) Use of 2-deoxy-D-[^{11}C] glucose for the determination of local cerebral glucose metabolism in humans: Variation within and between subjects. *J Cereb Blood Flow Metabol* 2: 307–319
- Skinhoj E (1965) Bilateral depression of CBF in unilateral cerebral diseases. *Acta Neurol Scand* [Suppl 14] 41: 161–163
- Slater R, Reivich M, Goldberg H, Banka R, Greenberg J (1977) Diaschisis with cerebral infarction. *Stroke* 8: 684–690

25. Sokoloff L, Reivich M, Kennedy C, DesRosiers MH, Patlak CS, Pettigrew KD, Sakurada D, Shinohara M (1977) The (^{14}C) Deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure, and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
26. von Monakow C (1914) Die Lokalisation im Grosshirn und der Abbau der Funktion durch kortikale Herde. JF Bergmann, Wiesbaden pp 26–34

Remote Functional Depression of Glucose Metabolism in Stroke and its Alteration by Activating Drugs

By W.-D. Heiss, H. W. Ilse, R. Wagner, G. Pawlik, and K. Wienhard

Applying the [^{18}F]fluorodeoxyglucose (FDG) method (Reivich et al. 1979a; Phelps 1981) and positron emission tomography (PET) in man, increased local cerebral metabolic rates for glucose (LCMRGI) during functional activation of nervous tissue have been demonstrated for the visual system (Reivich et al. 1979b; Phelps et al. 1981a), for the auditory system (Alavi et al. 1981; Phelps et al. 1982), and for the somatosensory system (Greenberg et al. 1981). By means of the ^{77}Kr inhalation technique, an increase in local cerebral blood flow (LCBF) was documented in the motor area after complicated finger movements (Roland et al. 1981). Some of these striking results and prospects on future studies have been reported by Phelps and Mazziotta (this volume) and Reivich et al. (this volume). In other studies of cerebral metabolism in patients suffering from ischemic stroke, a depression of oxygen uptake or glucose metabolism was observed in morphologically intact brain regions: Kuhl et al. (1980) described a reduction of glucose uptake in the thalamus ipsilateral to a cortical infarct. Using FDG (Phelps et al. 1981b) or [^{11}C]3-methyl-D-glucose (Heiss et al. 1982) as metabolic tracer, decreased glucose uptake was found in the CT-intact visual cortex of patients with an infarction in the territory of the middle cerebral artery by which the visual pathway was interrupted. A reduction of LCBF and oxygen consumption was observed in the hemisphere contralateral to supratentorial infarcts (Lenzi et al. 1981) and in the cerebellar hemisphere contralateral to infarcts in the cerebral cortex supplied by the carotid artery (Baron et al. 1981).

Stimulated by these observations, we systematically investigated the functional depression of glucose metabolism in patients who had suffered an ischemic stroke, and also studied the effect of activating drugs on these remote depressions of glucose metabolism.

Methods

The routine described by Phelps et al. (1979, 1981b) was used to measure LCMRGI: 3–6 mCi FDG was injected intravenously and arterialized blood was sampled from the vein of a heated hand from the time of injection to the end of the positron imaging period. Plasma FDG and glucose concentrations were determined and entered into the computer of the tomographic device (VAX 11/780, Digital Equipment Corporation). Tomographic positron images of the brain were recorded with a two-ring/three-slice system (Positron Camera PC 384-7B, Scanditronix) designed by Eriksson et al. (1982), starting 30 min after FDG injection. This device has a spatial resolution of 8 mm FWHM in 13-mm slices (Eriksson et al., this volume). A total of 9–12 tomographic images at different levels of the brain (0–11 cm above the canthomeatal line) were obtained. In several cases with larger ischemic lesions, specific rate constants were determined in individual regions of interest by consecutive scanning of these regions in periods of 3 min from the injection to the time of tomographic recording under steady state conditions (Hawkins et al. 1981). The model for LCMRGI calculation and average values of rate constants and lumped constants (Phelps 1981) are part of the software permitting calculation and display of metabolic maps or extraction of LCMRGI values from selected regions of interest (ROIs). For statistical analysis of LCMRGI values, up to 30 ROIs were used in several slices from individual cases. In five patients FDG studies were performed before and 20 min after an infusion of 12 g piracetam (Nootrop; UCB Chemie) over a 20-min period. Again, LCMRGI was calculated for selected ROIs and the values with and without medication were compared.

Results

LCMRGI in Normal Subjects

Maps of LCMRGI in a normal volunteer, measured under standard laboratory conditions (dim light, moderate noise of laboratory activity, no sensory deprivation), are shown in Fig. 1.

Metabolic values, which can be read in $\mu\text{mol}/100\text{ g}$ per minute by comparing the screen brightness (or color) to a reference gray (color) scale, are well within the range of values reported by Reivich et al. (1979 a), Kuhl et al. (1980), and Mazziotta et al. (1981). The highest metabolic rates were found in the basal ganglia and thalamus: 43–50 $\mu\text{mol}/$

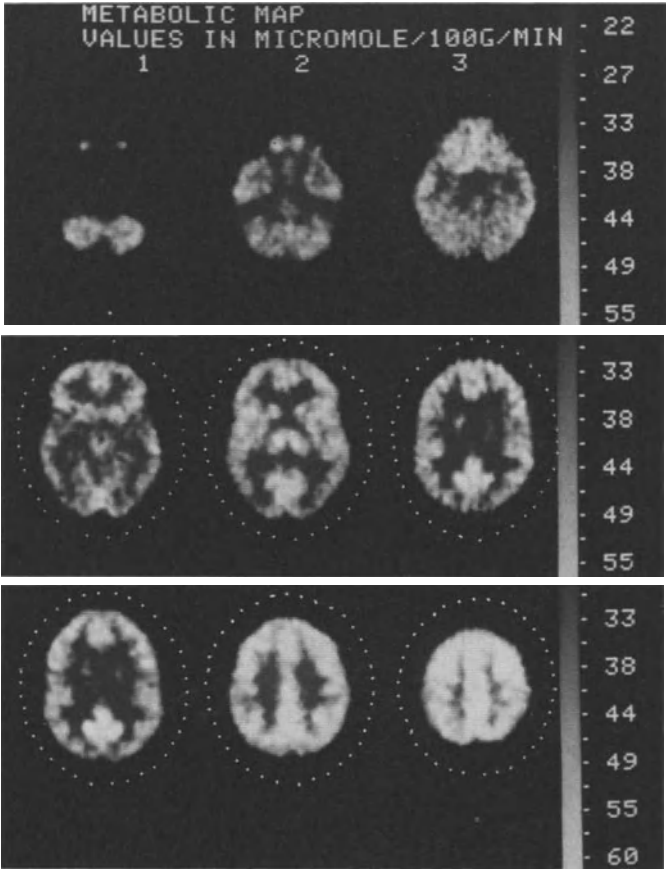


Fig. 1. Maps of LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) of brain slices from 0 to 10 cm above the canthomeatal line in a normal subject. Metabolic values can be determined by comparing the brightness of a region to the reference scale on the *right*. Note the high values in the basal ganglia and thalamus, the visual cortex, and the frontal lobe

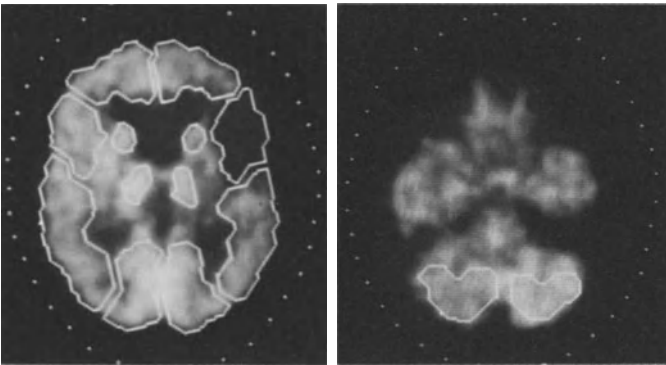


Fig. 2. LCMRGI calculated in individual regions of interest by using standard rate constants and rate constants determined from dynamic scans (see Table 1)

100 g per minute), in the visual cortex (55 $\mu\text{mol}/100\text{ g}$ per minute, slightly activated by room light), and in other cortical areas (approx. 45 $\mu\text{mol}/100\text{ g}$ per minute). The cerebellum had a lower glucose metabolism (35 $\mu\text{mol}/100\text{ g}$ per minute on average), and white matter structures consumed about 20 $\mu\text{mol}/100\text{ g}$ per minute.

LCMRGI in Ischemic Infarcts

In ten patients cerebral glucose metabolism was studied during the postacute to chronic phase after an ischemic stroke. In all these cases the ischemic infarct was clearly outlined by X-ray CT, and corresponding regions were selected in the PET images. For these ROIs,

Table 1. LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) in individual regions of interest

	Standard rate constants (0.102, 0.130, 0.062, 0.0068)		Calculated rate constants (G: 0.10, 0.20, 0.04, $\equiv$ 0) (I: 0.05, 0.14, 0.03, $\equiv$ 0)	
	Ipsi-lateral	Contra-lateral	Ipsi-lateral	Contra-lateral
Infarction	3	22	12	24
Frontal	17	20	19	22
Temporal	14	16	19	20
Occipital	17	20	23	25
Thalamus	25	32	27	32
Caudate nucleus	25	26	23	27
Cerebellum	21	18	23	19

LCMRGI was calculated using standard rate constants and rate constants determined individually from dynamic scanning (Fig.2). As shown in Table 1, the use of individually fitted rate constants resulted in slightly higher LCMRGI values in morphologically intact brain structures. In morphologically damaged brain tissue, however, i.e., within the ischemic infarction, differences were much more pronounced. LCMRGI computed for infarcted cortex as localized in CT was between 0 and 20 $\mu\text{mol}/100\text{ g}$ per minute, with a mean of $8.2 \pm 7.03\text{ }\mu\text{mol}/100\text{ g}$ per minute (Table 2).

LCMRGI in Inactivated Brain Structures

Morphologically intact brain structures with reduced glucose consumption were identified by comparing CT and metabolic images. As shown in Fig.3, the hypometabolic tissue volume was much larger than the ischemic infarct and comprised brain regions far away from the primary lesion. In general, a decrease in glucose metabolism was found in ipsilateral cortical as well as subcortical (e.g., thalamus) areas beyond the ischemically damaged tissue, but also in the cerebral cortex contralateral to the lesion and in the contralateral cerebellar hemisphere. Table 2 shows the mean values and standard deviations of LCMRGI from ROIs selected in corresponding brain regions of either side. A significant influence of the ischemic lesion on the metabolic rates in homolateral cortical areas and subcortical structures and on the contralateral cerebellum is demonstrated. The values of LCMRGI in the hemisphere contralateral to the infarct were signifi-

Table 2. LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) in individual brain regions in 10 stroke patients

	<i>n</i>	LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) Mean $\pm$ standard deviation		Sign test <i>p</i> <
		Ipsilateral	Contralateral	
Infarction	17	8.2 $\pm$ 7.03	29.6 $\pm$ 11.84	0,00001
Frontal	16	24.6 $\pm$ 10.49	30.0 $\pm$ 13.74	0.01
Temporal	18	24.6 $\pm$ 7.89	31.5 $\pm$ 12.07	0.001
Parietal	15	26.2 $\pm$ 9.27	31.9 $\pm$ 11.93	0.02
Occipital	19	26.9 $\pm$ 10.68	33.2 $\pm$ 13.56	0.005
Thalamus	14	25.2 $\pm$ 7.36	31.9 $\pm$ 7.77	0.002
Striatum	14	25.6 $\pm$ 9.79	35.0 $\pm$ 11.72	0.001
Cerebellum	8	27.8 $\pm$ 9.65	21.7 $\pm$ 10.62	0.005

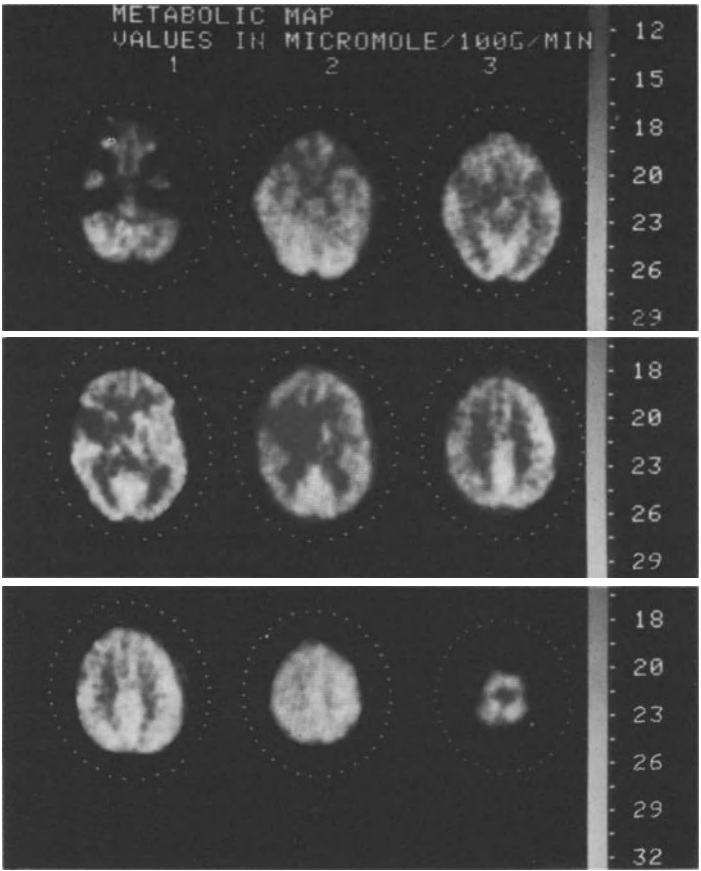


Fig. 3. Maps of LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) of brain slices from 0 to 11 cm above the canthomeatal line in a patient with an ischemic infarction. The infarcted area has the lowest LCMRGI value, but metabolism in other, even distant, areas is also decreased

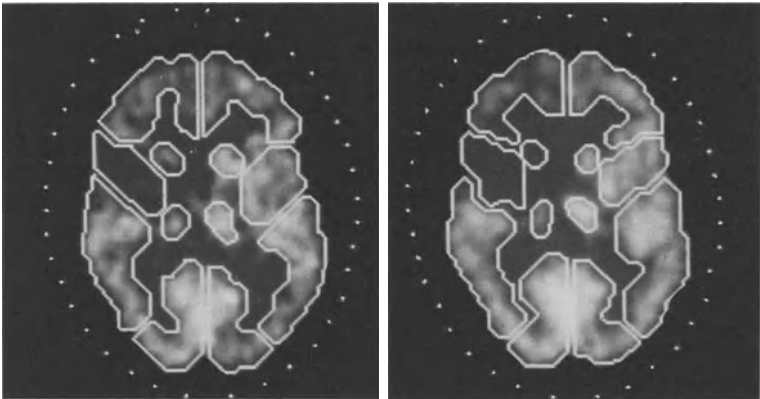


Fig. 4. Selected regions of interest in comparable metabolic images of a patient with ischemic stroke before

and after 12 g piracetam. The respective LCMRGI values are given in Table 3

cantly lower than LCMRGI in normal controls, but LCMRGI values in morphologically intact cortical areas homolateral to the infarct were on the average 17.9%–21.9% below the values in corresponding contralateral regions. The ho-

molateral thalamus was 21% and the homolateral striatum 26.9% below their contralateral counterparts. The depression of glucose metabolism in the contralateral cerebellum averaged 21.9%. Of all the contralateral cortical areas,

the region corresponding to the infarct had the lowest LCMRGI value; it was 6.6% below the mean value of the rest of the cortex contralateral to the ischemic lesion.

Effect of Piracetam on LCMRGI

In a preliminary study the metabolic effect of 12 g piracetam, infused over a period of 20 min was investigated in five stroke patients. Selected ROIs were used for comparison of LCMRGI measured with and without medication. As shown for one patient in Fig.4 and Table 3, LCMRGI is increased in all regions. Table 4 summarizes the results obtained in the five cases. Depressed glucose metabolism in the infarcted area and in functionally inactivated brain structures was significantly increased (paired t test, $p < 0.05$), while the change in the

corresponding region contralateral to the infarct did not reach the level of statistical significance. In unaffected brain structures no marked changes of LCMRGI were induced by piracetam.

Discussion

Accuracy of LCMRGI Measurements with FDG

The FDG model permits reliable determinations of LCMRGI in normal tissue, as demonstrated by the largely congruous values reported in the literature (for a review see Phelps et al. 1982). However, when rate constants differ from the routinely used standard values (Phelps 1981), e.g., due to altered kinetics in ischemic tissue (Hawkins et al. 1981); when metabolic rates change during the investigation (Huang et al. 1981); or when even the lumped constant is different, as has been observed in early experimental strokes (Crane et al. 1981; Mies et al., this volume), LCMRGI cannot be determined with sufficient accuracy. In these conditions it is necessary to determine both actual rate constants and blood volume by dynamic scanning. Only if rate constants are calculated individually for each ischemic region can LCMRGI be measured reliably and reproducibly. Such precautions are particularly important when repeated determinations of glucose metabolism are made in the course of a stroke or when treatment effects are to be quantified.

Table 3. LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) in individual brain regions of a stroke patient before and after piracetam

	LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$)			
	Control		Piracetam	
	Ipsi-lateral	Contra-lateral	Ipsi-lateral	Contra-lateral
Infarction	1	19	3	24
Frontal	11	13	15	19
Temporal	16	17	21	22
Occipital	21	18	26	24
Thalamus	13	22	16	28
Caudate nucleus	11	21	9	22
Cerebellum	14	11	19	15

Table 4. LCMRGI ($\mu\text{mol}/100\text{ g}/\text{min}$) in individual brain regions of 5 stroke patients before and after piracetam

	<i>n</i>	LCMRGI (μmol/100 g/min)		t test
		Mean ± standard deviation		<i>p</i> <
		Control	Piracetam	
Infarction	7	8.5 ± 7.84	10.8 ± 7.75	0.05
Ipsilateral cortex	22	23.7 ± 7.77	25.9 ± 8.21	0.05
Ipsilateral basal ganglia + contralateral cerebellum	8	19.4 ± 7.94	21.4 ± 8.42	0.05
Contralateral to infarct	7	23.1 ± 6.15	27.4 ± 9.12	N.S.
Contralateral cortex	22	29.1 ± 12.05	29.7 ± 9.91	N.S.
Contralateral basal ganglia + ipsilateral cerebellum	8	26.3 ± 8.81	27.9 ± 7.83	N.S.

Functional Inactivation and Glucose Metabolism

Positron emission tomography makes possible the detection of functionally impaired human brain structures exhibiting a disturbance of energy metabolism, even in regions where X-ray CT shows an intact morphology. While in most cases of such remote metabolic depressions an effect on the function of these brain regions is clinically apparent, the cause of this functional inactivation is not so obvious and may vary between cases. Patients with localized cerebral lesions for instance may develop a depression of neuronal function in contralateral regions of the brain. This phenomenon, known as diaschisis, was first described by von Monakow (1914) and later confirmed in CBF studies by Hoedt-Rasmussen and Skinhoj (1964), Meyer et al. (1970), and Ginsberg et al. (1977). This term, however, should be used only in its original definition by von Monakow (1914) to characterize the transcallosal depression of function, metabolism, and flow in contralateral brain regions corresponding to cortical infarcts. Such an effect was demonstrated in this study. However, remote effects other than the transcallosal deactivation in the area contralateral to the infarct were even more pronounced. They involved the basal ganglia and the thalamus, other cortical regions, and also the contralateral cerebellar hemisphere. These effects may be related to an interruption of interacting fiber systems in complex functional loops: disruption of the corticothalamic projections deactivates the homolateral thalamus and basal ganglia, thus depressing local metabolism (Kuhl et al. 1980). On the other hand the ischemic destruction of the basal ganglia and the thalamus inactivates the cortex. Morphologically intact cortical areas also are inactivated by disruption of afferent sensory fibers, e.g., the visual pathway (Heiss et al. 1982), or by interruption of other afferent fiber systems, e.g., the activating system to the frontal brain, and of associating pathways. The effect of retrograde degeneration after destruction of efferent fibers on number and metabolism of cells and flow in the cerebral cortex remains to be ascertained (Mies et al., to be published).

The depression of oxygen consumption (Baron et al. 1981) and glucose metabolism in the con-

tralateral cerebellum is probably due to the interruption of corticopontocerebellar projections. The crossed cerebellar atrophy observed in some cases long after a supratentorial infarction or hemorrhage may result from the depressed metabolism due to remote functional deactivation (Baron et al. 1981).

These observations of remote effects on glucose metabolism in patients with localized ischemia indicate the involvement of large and widely distributed brain areas in the syndrome of stroke. Some of the neurological symptoms and their potential rehabilitation may well be influenced by such remote functional disturbances.

Effect of Activating Drugs

As shown in our preliminary study of the effect of piracetam on cerebral glucose metabolism, PET can be used for quantification of drug effects on regional brain metabolism. The limitations inherent in regional flow measurements with conventional isotope techniques and in metabolic studies based on arteriovenous differences of substrates do not exist. However, PET is so far performed in only a few centers, and both the equipment required and the cost of tracers limit its applicability. Our results indicate that LCMRGI can be improved significantly in areas with a decreased metabolism. The mechanism by which piracetam induces metabolic changes is still obscure, but may be related to an activation of oxidative phosphorylation and an increased production of ATP and other high-energy phosphates (Pede et al. 1971).

While the benefits of an increase in LCMRGI in infarcted tissue are questionable, a stimulation of depressed metabolism in remote regions of the brain may alleviate neurologic deficits unrelated to the primary ischemic lesion and may therefore improve prospects for rehabilitation. However, the clinical value of such therapy, indicated in one of our cases by a remarkable recovery from long-lasting neurologic deficits after initiation of piracetam treatment, remains to be proved in controlled trials. A significant improvement of neurologic symptoms should be demonstrated to parallel the increase in metabolism induced by piracetam.

References

- Alavi A, Reivich M, Greenberg J et al. (1981) Mapping of functional activity in brain with [^{18}F]fluorodeoxyglucose. *Semin Nucl Med* 11: 24–31
- Baron JC, Bousser MG, Comar D et al. (1981) Crossed cerebellar diaschisis: A remote functional depression secondary to supratentorial infarction in man. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 500–501
- Crane PD, Pardridge WM, Braun LD et al. (1981) The interaction of transport and metabolism on brain glucose utilization: A reevaluation of the lumped constant. *J Neurochem* 36: 1601–1604
- Eriksson L, Bohm Ch, Kesselberg M et al. (1982) A four ring positron camera system for emission tomography of the brain. *IEEE Trans Nucl Sci* 29: 539–543
- Ginsberg MD, Reivich M, Giandomenico A et al. (1977) Local glucose utilization in acute focal cerebral ischemia: local dysmetabolism and diaschisis. *Neurology (Minneapolis)* 27: 1042–1048
- Greenberg J, Reivich M, Alavi A et al. (1981) Metabolic mapping of functional activity in human subjects with the [^{18}F]fluorodeoxyglucose technique. *Science* 212: 678–680
- Hawkins R, Phelps ME, Huang SC et al. (1981) Effect of ischemia on quantification of local cerebral glucose metabolic rate in man. *J Cereb Blood Flow Metabol* 1: 37–52
- Heiss WD, Vyska K, Kloster G et al. (1982) Demonstration of decreased functional activity of visual cortex by [^{11}C]-methylglucose and positron emission tomography. *Neuroradiology* 23: 45–47
- Hoedt-Rasmussen K, Skinhøj E (1964) Transneuronal depression of the cerebral hemispheric metabolism in man. *Acta Neurol Scand* 40: 41–46
- Huang SC, Phelps ME, Hoffman EJ et al. (1981) Error sensitivity of fluorodeoxyglucose method for measurement of cerebral metabolic rate of glucose. *J Cereb Blood Flow Metabol* 1: 391–401
- Kuhl DE, Phelps ME, Kowell AP et al. (1980) Effects of stroke on local cerebral metabolism and perfusion: mapping by emission computed tomography of ^{18}F FDG and $^{13}\text{NH}_3$. *Ann Neurol* 8: 47–60
- Lenzi GL, Frackowiak RS, Jones T (1981) Regional cerebral blood flow (CBF), oxygen utilization (CMRO $_2$) and oxygen extraction ratio (OER) in acute hemispheric stroke. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 504–505
- Mazziotta JC, Phelps ME, Miller J et al. (1981) Tomographic mapping of human cerebral metabolism: normal unstimulated state. *Neurology (Minneapolis)* 31: 503–516
- Meyer JS, Shinohara Y, Kanda T et al. (1970) Diaschisis resulting from acute unilateral cerebral infarction. Quantitative evidence for man. *Arch Neurol* 23: 241–247
- Mies G, Auer LM, Ebhardt G et al. (to be published) Flow and neuronal density in tissue surrounding chronic infarction. *Stroke*
- Pede JP, Schimpfessel L, Crokaert R (1971) The action of piracetam on oxidative phosphorylation. *Arch Int Physiol Biochem* 79: 1036
- Phelps ME (1981) Positron computed tomography studies of cerebral glucose metabolism in man: theory and application in nuclear medicine. *Semin Nucl Med* 11: 32–49
- Phelps ME, Huang SC, Hoffman EJ et al. (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with (F-18) 2-fluoro-2-deoxyglucose-D-glucose: validation of method. *Ann Neurol* 6: 371–388
- Phelps ME, Kuhl DE, Mazziotta JC (1981a) Metabolic mapping of the brain's response to visual stimulation: studies in humans. *Science* 211: 1445–1448
- Phelps ME, Mazziotta JC, Kuhl DE et al. (1981b) Tomographic mapping of human cerebral metabolism: Visual stimulation and deprivation. *Neurology (Minneapolis)* 31: 517–529
- Phelps ME, Mazziotta JC, Huang SC (1982) Study of cerebral function with positron computed tomography. *J Cereb Blood Flow Metabol* 2: 113–162
- Reivich M, Kuhl D, Wolf A et al. (1979a) The [^{18}F]fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
- Reivich M, Greenberg J, Alavi A et al. (1979b) The use of the [^{18}F]fluoro-deoxyglucose technique for mapping of functional neural pathways in man. *Acta Neurol Scand* 60 [Suppl 72]: 198–199
- Roland P, Meyer E, Yamamoto Y et al. (1981) Dynamic positron emission tomography as a tool in neuroscience: Functional mapping in normal human volunteers. *J Cereb Blood Flow Metabol* 1, Suppl 1: 463–464
- von Monakow C (1914) Die Lokalisation im Großhirn und der Abbau der Funktion durch kortikale Herde. Bergmann, Wiesbaden

Regional Perfusion and Glucose Uptake Determination with ^{11}C -Methyl-Glucose and Dynamic Positron Emission Tomography

By K. Vyska, G. Kloster, L. E. Feinendegen, W.-D. Heiss, G. Stöcklin, A. Höck, C. Freundlieb, A. Aulich, F. Schuier, H. U. Thal, V. Becker, and A. Schmid

Introduction

Recent studies have emphasized that quantitative assessment of different brain disorders may require the knowledge of at least two parameters. One is local perfusion, and the second should relate to tissue metabolism, for example to the glucose utilization rate (Sokoloff et al. 1977; Phelps et al. 1979) or the local unidirectional glucose transport rate (LUGTR) (Vyska et al. 1981, 1982; Heiss et al. 1981). All of these parameters may be determined by PET.

There is experimental evidence that the glucose utilization rate, in contrast to the LUGTR, might be normal even after prolonged anoxia (Betz et al. 1974). Thus during the first 2 min anoxia the rate of net glucose uptake was more than doubled, but after 10 min anoxia it declined to the preanoxic level, to remain unaltered for up to 30 min anoxia and during the first hour of postanoxic recovery. The LUGTR, on the other hand, was unchanged for the first 2 min anoxia. During the following 10 min it decreased to 39% of the control rate, remaining reduced up to the end of 30 min anoxia. During the recovery period of 1 hour, the LUGTR increased to 59% of the control value. The persistently reduced LUGTR but normal net glucose uptake in the postanoxic period suggested the impairment of unidirectional glucose transport systems. This conclusion agrees with the observation of long-lasting depression of functional activity after prolonged complete ischemia with no insufficiency of energy-supplying mechanisms (Hinzen et al. 1972). Thus the LUGTR is obviously more sensitive for detecting injury to viable tissue than the glucose utilization rate.

The LUGTR may be measured by ^{11}C -methyl-D-glucose (CMG) and dynamic positron emission tomography (dPET).

The aim of this chapter is to describe the application of the CMG technique to the measure-

ment of the rate constants for CMG influx (κ_1^*) and CMG efflux (k_2^*). Moreover, it will be shown that these parameters permit the simultaneous determination of the local perfusion rate (LPR) and the LUGTR.

Material and Methods

CMG is a glucose analogue in which the hydrogen atom of the hydroxyl group at C-3 is replaced by a C-11 methyl group (Kloster et al. 1981).

In our studies, 2–5 mCi CMG was injected into an antecubital vein and the transaxial activity distribution in one selected slice of brain was recorded with the ECAT II scanner at 2-min intervals for 40 min. Different regions of interest (ROIs) were selected in the brain scans and time-activity curves created. Since the blood volume in brain is very small (cortex 4%, white matter 2%) the time-activity curves registered over brain were considered as being representative for the changes in brain tissue CMG concentration. These were evaluated in relation to the blood CMG concentration, for which the activity registered over the superior longitudinal sinus (SLS) was considered as being representative. This approach is possible because:

1. The nonmetabolizable CMG is slowly eliminated from the blood pool, so that its concentration in blood remains relatively high during the whole measurement period. Consequently, the CMG concentration in the blood is nearly the same as that in the brain cortex.
2. According to the model shown below, it is not the absolute blood CMG concentration but its ratio to the tissue CMG concentration that is needed for measuring transport and flow.
3. From the spatial point of view, the SLS can be considered as being integrated into the body of the brain cortex.

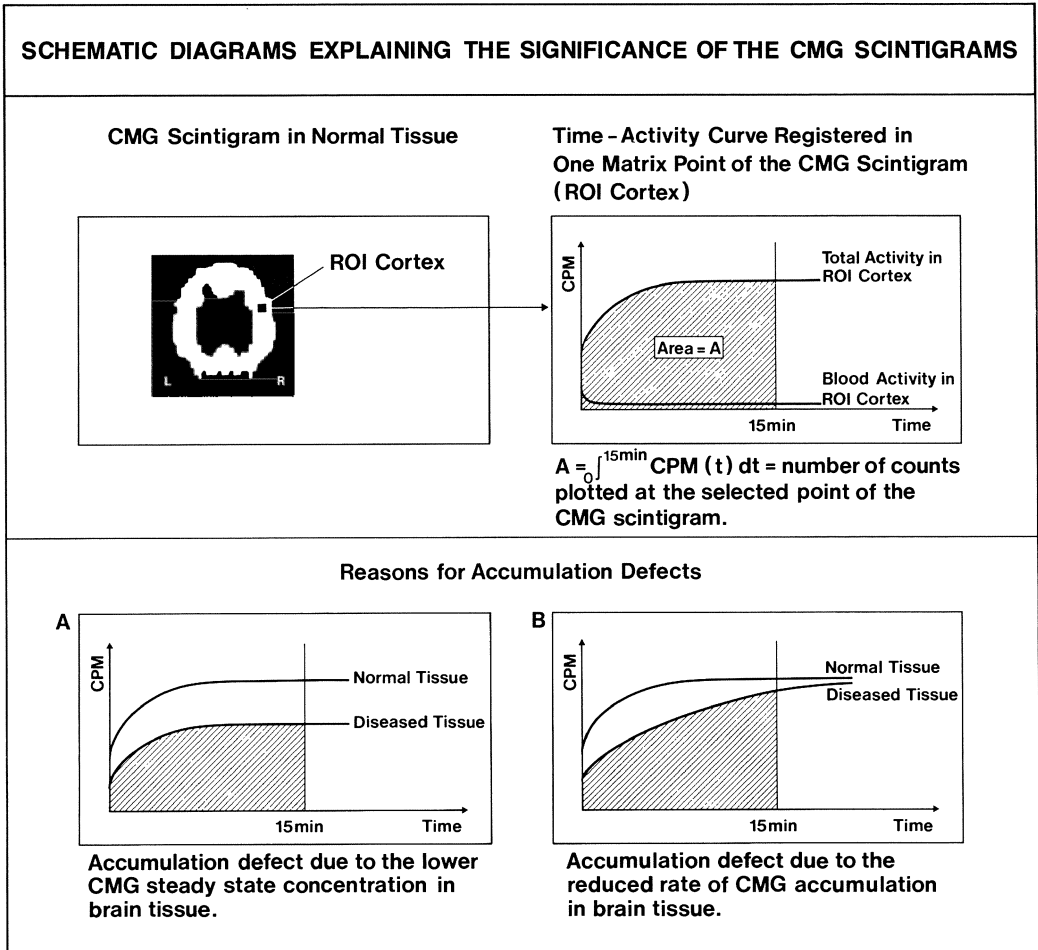


Fig. 1. The significance of the CMG scintigrams

Under these conditions the partial volume and crossover effects in the SLS and brain cortex ROIs would be practically the same, provided that the size of the two ROIs is comparable. They would practically cancel out in the ratio of two ROI activities. Consequently, the accuracy of the ratio of tissue to blood CMG concentration thus determined would be higher than the accuracy of the absolute activities measured in the neighborhood and/or in the areas of accumulation defects.

In order to analyze the regional distribution of CMG in brain tissue, the first six images registered in the dynamic study were summed up.

In our studies, four healthy volunteers and 50 patients suffering from ischemic stroke (two of them before and after an extra-intracranial

bypass operation) were examined. In all cases a CT investigation was performed before the CMG study.

Results

The CMG was found to be effectively accumulated in normal brain cortex; a significantly lower accumulation was observed in white matter (Fig. 1).

The time-activity curve registered over the SLS ROI usually showed two exponential components (Vyska et al. 1982). The initial rapid decrease of activity probably reflects mixing of indicator in the blood pool and its equilibration

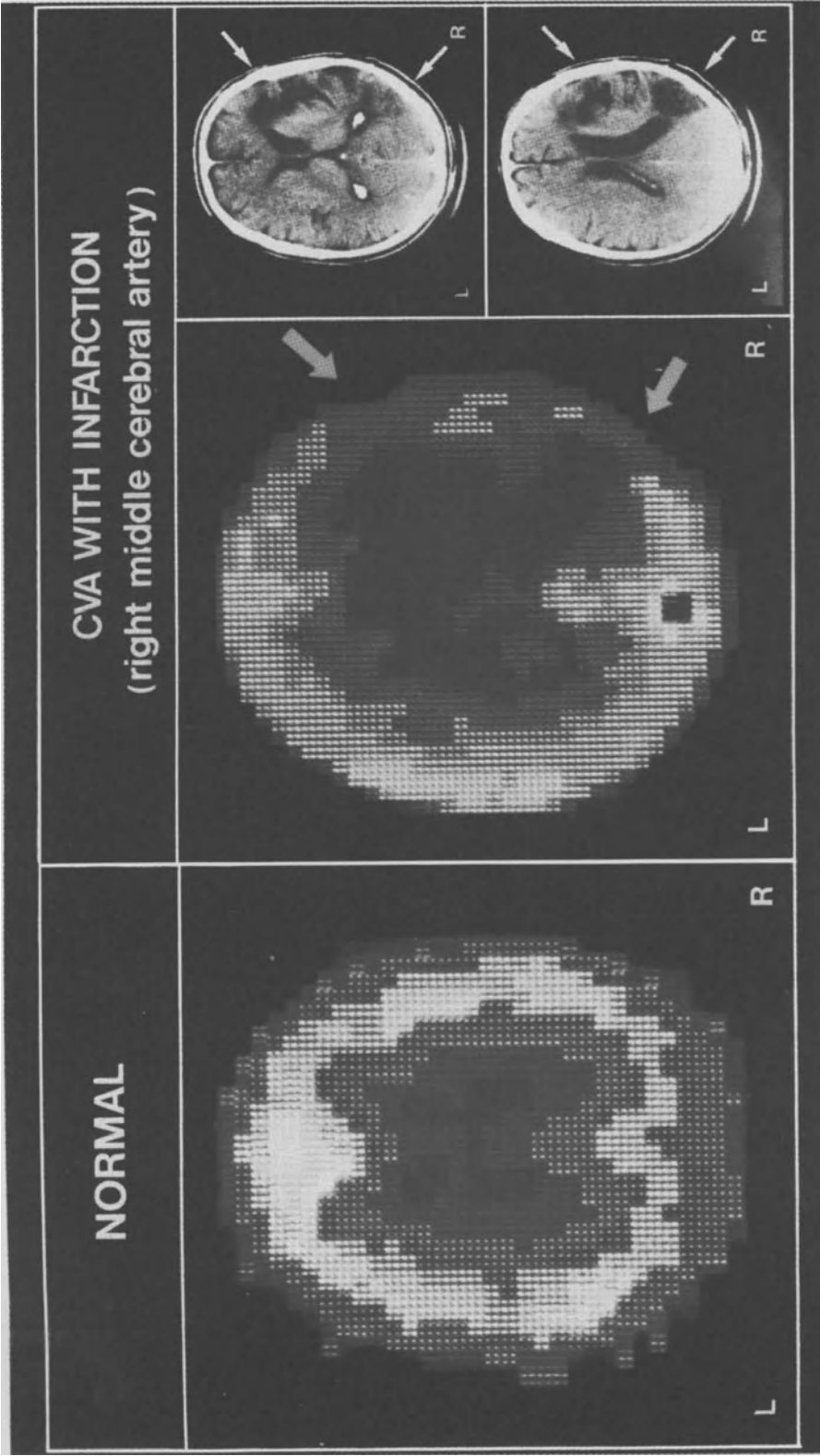


Fig. 2. CMG distribution pattern in normal brain (*left*) and in infarction (*right*)

with tissue. This was followed by slow indicator elimination ($T_{1/2} > 90$ min), indicating a high retention of nonmetabolizable indicator in the blood.

The time-activity curves registered over different brain regions exhibit a rapid accumulation phase and a slow elimination phase. The accumulation half-times in brain ranged from 1.8 to 3.2 min for normal cortex and from 5 to 7 min for normal white matter. These times were significantly prolonged in ischemic areas. The values obtained from areas with destroyed blood-brain barrier (BBB) exceeded 10 min in cortex and 16 min in white matter.

At steady-state distribution of the activity between tissue and blood, the CMG elimination from the brain tissue follows the time activity curve in blood. The CMG elimination half-time from brain tissue was 90 min or more.

In CMG scintigrams of stroke patients clear accumulation defects are recognized (see Fig. 2). Frequently the accumulation defects appearing in CMG scans were larger in size than the hypodense zones in computer tomograms. Since the CMG scintigrams were reconstructed from the data registered over the first 15 min after i.v. CMG administration, the accumulation defects may reflect both the reduced steady-state CMG concentration and the reduced rate of CMG accumulation in brain tissue (see Fig. 1).

Quantitative Evaluation of the Data

CMG is transported across the BBB by the same carrier as glucose, but it is not phosphorylated and further metabolized (Bidder 1968; Czaky and Wilson 1956). In the model presented in Fig. 3, c_B^* [cpm/ml] and c_2^* [cpm/g] are the CMG concentrations in blood and in brain tissue respectively, κ_1^* [min^{-1}] is the observed rate constant for CMG influx, and k_2^* [min^{-1}] the rate constant for CMG efflux.

In plasma, CMG competes with glucose for a common carrier for transport from plasma into a primary precursor pool in brain tissue. The rate of CMG accumulation in brain tissue is equal to the difference between the rate of CMG influx ($\kappa_1^* \cdot c_B^*$) and CMG efflux ($k_2^* \cdot c_2^*$) (Vyska et al. 1982):

$$dc_2^*/dt = \kappa_1^* \cdot c_B^* - k_2^* \cdot c_2^*. \quad (1)$$

The formal solution of this equation is

$$\begin{aligned} c_2^*(t) &= \kappa_1^* \cdot \int_0^t c_B^*(\tau) \cdot e^{-k_2^*(t-\tau)} d\tau + c_2^*(0) \cdot e^{-k_2^* \cdot t} = \\ &= (\kappa_1^* \cdot c_B^*(t) + c_2^*(0) \cdot \delta(t)) * e^{-k_2^* \cdot t} \end{aligned} \quad (2)$$

where $c_2^*(0)$ is the tissue CMG concentration at time $t=0$.

Since Eqs. (1) and (2) are valid for all t , they permit the determination of the unknowns κ_1^* and k_2^* , provided that $c_2^*(t)$ and $c_B^*(t)$ are known. For this various numerical methods (e.g., the least-square method) are available.

With the CMG technique, the blood CMG concentration, $c_B^*(t)$, may be determined either by blood sampling or by analysis of the time-activity curve registered over the SLS. The brain tissue CMG concentration $c_2^*(t)$ is determined by the time-activity curves registered over different brain areas.

In the present studies we used the time-activity curve registered over the SLS as a measure for CMG blood concentration. This curve could usually be approximated by two exponential functions. Under these conditions the ratio κ_1^*/k_2^* and the value of k_2^* can be determined from the tissue time-activity curves as described previously (Vyska et al. 1982).

In order to demonstrate the physical meaning of Eqs. (1) and (2), a simple case of CMG behavior in blood is given below for which the analytical solution of Eq. (2) is possible.

In this case the elimination of the nonmetabolizable CMG from the blood pool is so slow that it can be neglected and the mixing of the indicator in the blood is practically instantaneous, so that the CMG blood concentration $c_B^*(t)$ can be considered to be constant.

In steady state ($\frac{dc_2^*}{dt} = 0$) the influx is equal to efflux, i.e.:

$$\chi_1^* \cdot c_B^* = k_2^* \cdot c_{2ss}^* \text{ and } \kappa_1^*/k_2^* = c_{2ss}^*/c_B^* \quad (3)$$

where c_{2ss}^* [cpm/g] is the CMG concentration in brain tissue at steady state.

This means that in a steady state the ratio κ_1^*/k_2^* is equal to the ratio of tissue and blood CMG concentrations.

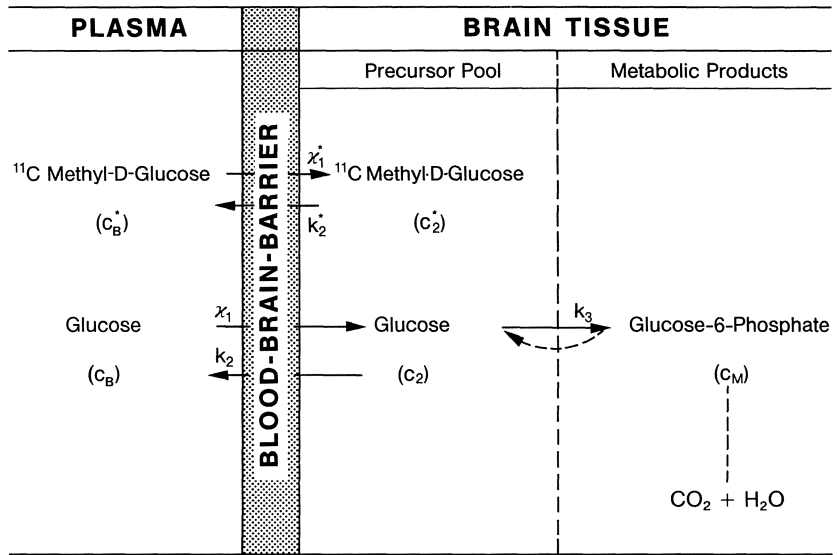


Fig. 3. The model used for quantitative analysis of CMG kinetics in brain tissue [modified according to Sokoloff et al. (1977)]

Because CMG influx is directly proportional to blood CMG concentration, CMG influx is constant when blood CMG concentration is constant. Therefore in this case Eq. (1) can be written as follows:

$$\frac{dc_2^*}{dt} = \kappa_1^* \cdot c_B^* - k_2^* \cdot c_2^* = \text{constant} - k_2^* \cdot c_2^* = \frac{k_2^* \cdot (\text{const}/k_2^* - c_2^*)}{(1)} \quad (4)$$

This equation demonstrates that under the condition of $c_B^*(t)$ being constant, the rate of CMG accumulation in tissue is governed solely by CMG efflux.

If in Eq. (4) the term const/k_2^* is designated as const_1 , Eq. (4) can be rearranged as follows:

$$\frac{1}{\text{const}_1 - c_2^*} \cdot \frac{d(\text{const}_1 - c_2^*)}{dt} = -k_2^* \quad (5)$$

This means that in the case of $c_B^*(t) = \text{const}$ the slope of approach of CMG tissue concentration to the steady state is equal to k_2^* .

Ratio κ_1^*/k_2^*

Using the approach described above the values of κ_1^*/k_2^* in normal cortex were determined to be 0.80–0.98. In normal white matter values of

0.3–0.4 were observed. These values agree favorably with the data reported in the literature for measurements in vivo (Sokoloff et al. 1977; Phelps et al. 1979; Kessler 1982, personal communication). They disagree, however, with experimental findings by Lund-Andersen and Kjeldsen (1976) in in vitro experiments on thick rat brain slices, which are supported by the theoretical analysis of Narahara et al. (1960) and which suggest that in in vitro experiments k_1 (as k_1 is designated the rate constant for glucose influx observed in in vitro experiments) is equal to k_2 , i.e., $k_1/k_2 = 1$ in all brain areas.

In order to explain this apparent discrepancy between the data obtained in in vivo and in vitro experiments, we considered the data reported by Betz et al. (1973), who studied the glucose influx rate in isolated dog brains under three different conditions.

In the first study they determined the glucose influx rate by different arterial glucose concentrations and different plasma flows. These experiments demonstrated that the glucose influx rate in brain tissue, Φ_i [$\mu\text{mol}/\text{min}$ per gram], is proportional to plasma flow f_p [ml/min per gram], arterial glucose plasma concentration c_p [$\mu\text{mol}/\text{ml}$], and fractional extraction $E[1]$, i.e.

$$\Phi_i = f_p \cdot c_p \cdot E \quad (6)$$

In the second experiment the glucose influx rate was determined at a constant plasma flow rate f_p (ml/min per gram), but at different glucose arterial plasma concentrations c_p (tested range of c_p was 3–50 $\mu\text{mol/ml}$). In these experiments the authors demonstrated that the fractional extraction E follows exactly the hyperbolic function $\frac{a}{b+c_k}$, where c_k is the mean capillary glucose concentration. This observation suggests that at a constant flow the glucose influx behaves as a first-order enzyme-catalyzed reaction. Since the values of constants a^1 (1.94) and b (5.4) characterizing this hyperbolic function agree closely with values of the constants V_M (2 $\mu\text{mol/min per gram}$) and K_M (5 $\mu\text{mol/min per gram}$) observed for the activity of the carrier system in in vitro studies (Bachelard 1971), it can be expected that the fractional extraction E is directly proportional to the rate constant for glucose influx observed in in vitro preparation, k_1 , i.e.

$$E = m \cdot k_1 \quad (7)$$

where m is the proportionality constant having the dimension [min]. (The physical meaning of m is discussed below.)

Under these conditions Eq. (6) can be written as follows:

$$\Phi_i = f_p \cdot c_p \cdot m \cdot k_1 \quad (8)$$

This means that if Eq. (7) is valid, one must expect on the basis of Eq. (8) that at constant arterial glucose plasma concentration (c_p), Φ_i must be directly proportional to the plasma flow f_p . This direct proportionality between Φ_i and f_p could be demonstrated by Betz et al. (1973) in their third experiment, in which they determined the glucose influx at a constant c_p but at different plasma flow rates (tested range of plasma flows was 0.20–0.80 ml/min per gram). Since the plasma flow f_p is related to the blood flow f [ml/min per gram], by the following relationship:

$$f_p = (1 - H_t) \cdot f \quad (9)$$

where H_t is the hematocrit, Eq. (8) can also be written as follows:

$$\Phi_i = m \cdot k_1 \cdot f \cdot (1 - H_t) \cdot c_p \quad (10)$$

This relationship indicates that only the glucose in plasma, not the glucose in erythrocytes, is available for transmembrane transport. This conclusion is in close agreement with the findings of Whitfield et al. (1974), who demonstrated that the exchange of glucose between plasma and erythrocytes is very slow.

Since under these conditions, the blood (c_B) and plasma (c_p) concentrations of available glucose are related to each other by

$$c_B = (1 - H_t) \cdot c_p$$

Eq. (10) can be written as follows:

$$\Phi_i = m \cdot k_1 \cdot f \cdot c_B \quad (11)$$

If this equation is compared with the commonly used formulation of glucose influx rate ($\kappa_1 \cdot c_B$) it becomes evident that:

$$\kappa_1 = m \cdot k_1 \cdot f \quad (12)$$

This equation demonstrates that the rate constant for glucose influx observed in vivo experiments, κ_1 , is proportional not only to the rate constant k_1 characterizing the catalytic activity of the carrier system for glucose influx, but also to the flow. This explains the difference between in vivo and in vitro experiments.

With respect to m , the data of Betz et al. (1973), were used and the value of m was determined to be 1 min.

Concerning the meaning of m , it must be considered that the product $m \cdot f$ is equal to $m \cdot F_i / W_i$, where F_i is the flow per W_i grams of tissue and $m = 1$ min. In this ratio ($m \cdot F_i / W_i$) $m \cdot F_i$ expresses the volume of the blood passing through the volume of W_i grams of tissue in 1 min. Thus $m \cdot F_i / W_i$ represents the ratio of two volumes and might be understood as the ratio of distribution volumes in the case of semiopen systems.

¹ In these calculations E was corrected for diffusion as described by Betz et al. (1973)

Rate Constant k_2^*

The value of k_2^* was determined to be 0.235 min^{-1} in normal cortex and 0.121 min^{-1} in normal white matter. These values are in good agreement with the data reported for rat brains (Pardridge and Oldendorf 1975; Sokoloff et al. 1977) and for humans (Kessler 1982, personal communication). They are, however, higher than the values reported for the humans by Phelps et al. (1979).

Determination of LPR

According to Eq. (11) the rate constant for glucose influx observed in in vivo experiments, κ_1 , is

$$\kappa_1 = k_1 \cdot f \cdot m \text{ (see Eq. 12)} \quad (13)$$

Since CMG is administered in tracer amounts, the rate constants κ_1^* and k_1^* may be considered as true first-order rate constants which are independent of plasma and tissue CMG concentration and which relate to the rate constants κ_1 and k_1 as follows:

$$\kappa_1^*/k_1^* = \kappa_1/k_1$$

After substitution of this relationship into Eq. (13) one obtains

$$\kappa_1^* = k_1^* \cdot f \cdot m$$

Dividing both sides of this equation by k_2^* leads to

$$\kappa_1^*/k_2^* = m \cdot f \cdot k_1^*/k_2^* \quad (14)$$

Based on: 1) the theoretical analysis by Narahara et al. (1960), which suggests that the glucose in tissue competes by back transport across the cell membrane with forward reaction (glucose influx) for combination with carrier molecule, so that the glucose transport across the cell membrane has to be treated as a reversible unimolecular enzyme catalysed reaction, 2) experiments by Pardridge et al. (1975) which indicate that the glucose transport across the BBB is symmetrical, and 3) the experiments of Lund-Andersen and Kjeldsen (1976) which

demonstrate that the ^{14}C -labeled 3-O-methyl glucose equilibrates in thick rat brain slices at a concentration which relates to concentration of methyl glucose in the incubation medium in a ratio of 0.95:1, we assumed that the rate constant characterizing the catalytic activity of carrier system for glucose influx, k_1 , is equal to the rate constant characterizing the catalytic activity of carrier system for glucose efflux, k_2 , i.e., $k_1 = k_2$. Under these conditions it is also true that $\kappa_1^* = k_2^*$. Based on this assumption Eq. (14) can be written as follows:

$$f = 1/m \cdot \kappa_1^*/k_2^* \quad (15)$$

In this relationship f is directly proportional to the ratio of the rate constants κ_1^* and k_2^* . Since both these parameters can be determined by CMG and dPET, this relationship was used for the determination of LPR in the present study. The validity of this approach was tested by comparing the values for flow obtained by the use of Eq. (15) in controls with the data reported in literature. The close agreement between these two values (see below) suggests that the above assumption is correct.

Determination of LUGTR

According to equations (10) and (12), the glucose influx rate, Φ_i , is given by:

$$\Phi_i = \kappa_1 \cdot (1 - H_i) \cdot c_p \quad (16)$$

Whereas H_i and c_p can be easily determined by blood analysis, the value of κ_1 must be obtained by the CMG technique. Since the affinities of CMG and glucose to the common carrier are slightly different (Pardridge et al. 1975), the rate constant κ_1^* , which is determined by the means of CMG technique, is not identical to the rate constant κ_1 . If, however, it is considered that $\kappa_1/\kappa_1^* = k_1/k_1^*$ and that according to Sokoloff et al. (1977) k_1 and k_1^* relate to each other as follows:

$$\frac{k_1}{k_1^*} = \frac{V_M \cdot K_M^*}{V_M^* \cdot K_M} = \xi \quad (17)$$

the following relationship is obtained:

$$\kappa_1 = \xi \cdot \kappa_1^*$$

If the values of K_M^* , V_M^* , K_M , and V_M determined by Pardridge et al. (1975) in rat brains are considered, the value of ξ can be calculated to be 1.11. Consequently,

$$\kappa_1 = \xi \cdot \kappa_1^* = 1.11 \cdot \kappa_1^* \quad (18)$$

After substitution of Eq. (18) into Eq. (16), the following relationship is obtained for the LUGTR (Φ_i):

$$\Phi_i = 1.11 \cdot \kappa_1^* \cdot (1 - H_i) \cdot c_p \quad (19)$$

Evaluation and Discussion of Data

Using the approach described above, the LPR [Eq. (15)] and the LUGTR [Eq. (19)] were calculated. In normal persons the LPR was 0.8–0.98 ml/min per gram in cortex and 0.3–0.4 ml/min per gram in white matter. The values for the cortex are in close agreement with data obtained with ^{133}Xe by inhalation and arterial blood sampling (Obrist et al. 1967) or following internal carotid injection (Ingvar et al. 1965). The values for the white matter are higher than those determined by ^{133}Xe (Obrist et al. 1967). This is probably due to the inclusion of larger vessels in the white matter ROIs in the present study.

The glucose influx rate (LUGTR) was 0.43–0.6 $\mu\text{mol}/\text{min}$ per gram in normal cortex and 0.09–0.12 $\mu\text{mol}/\text{min}$ per gram in white matter. These values are higher than those reported with ^{18}F -deoxyglucose (FDG) (Phelps et al. 1982). This is to be expected, because CMG measures the glucose influx and FDG the net glucose transport rate. The cortex to white matter ratio of the glucose influx rates was approximately 4. This value is in close agreement with data reported by Kennedy et al. (1978), who used FDG in monkeys. It is, however, higher than the ratio measured in humans by Phelps et al. (1979) with FDG.

The data obtained in stroke patients demonstrate that the accumulation defects in CMG scintigrams are frequently larger than the hypodense zones in CT.

The analysis of the time-activity curves, which were observed over the areas of accumulation defects in morphologically intact cortex, indi-

cated that in these areas the relative changes of LPR may equal, exceed, or be lower than the relative changes of LUGTR.

Parallel changes of LPR and LUGTR could be observed, for example, in a patient with left homonymous hemianopia caused by infarction in the distribution area of the right middle cerebral artery, but with an intact posterior cerebral artery. The CMG image, which was taken 5–6 cm above the orbitomeatal line, clearly showed two accumulation defects. One of them, a large frontotemporal area extending from the surface into the depth of the brain, closely correlated with CT findings (Heiss et al. 1982). The second was in the occipital lobe, where CT did not show any abnormality. In this area both the LPR (0.68 ml/min per gram) and LUGTR (0.38 $\mu\text{mol}/\text{min}$ per gram) were 22% lower than the corresponding values in the opposite hemisphere. The rate constant for the glucose carrier system in this area was, however, the same as that in the opposite hemisphere. (The rate constant for the glucose carrier system was taken to be equal, $k_2^* = \kappa_1^*/f$.) This means that the reduction of LUGTR in morphologically intact visual cortex was solely due to the reduction of LPR. Reivich et al. (1979), and Phelps et al. (1981), observed a similar degree of decrease in net glucose flux into visual cortex in controls when visual stimulation was shut off. The similarity of data suggests that the accumulation defects observed in visual cortex demonstrate the transport and perfusion response of the morphologically intact cortex to the destruction of visual pathways.

In a patient (P.L.) exposed to transient ischemia, the neurological symptoms (sensory loss) suggested an alteration of corresponding brain tissue, but there were no CT findings. In CMG scans a clear accumulation defect in temporo-parietal cortex was recognized. In this area, the LPR (0.93 ml/min per gram) was normal. The LUGTR, however, was 40% lower when compared with the opposite hemisphere. This reduction was due solely to a 40% reduction of the rate constant for the glucose carrier system. This observation indicates that residual impairment of the glucose transport system may follow transient ischemia and should be discussed as one of the reasons for deactivation of a morphologically intact cortex.

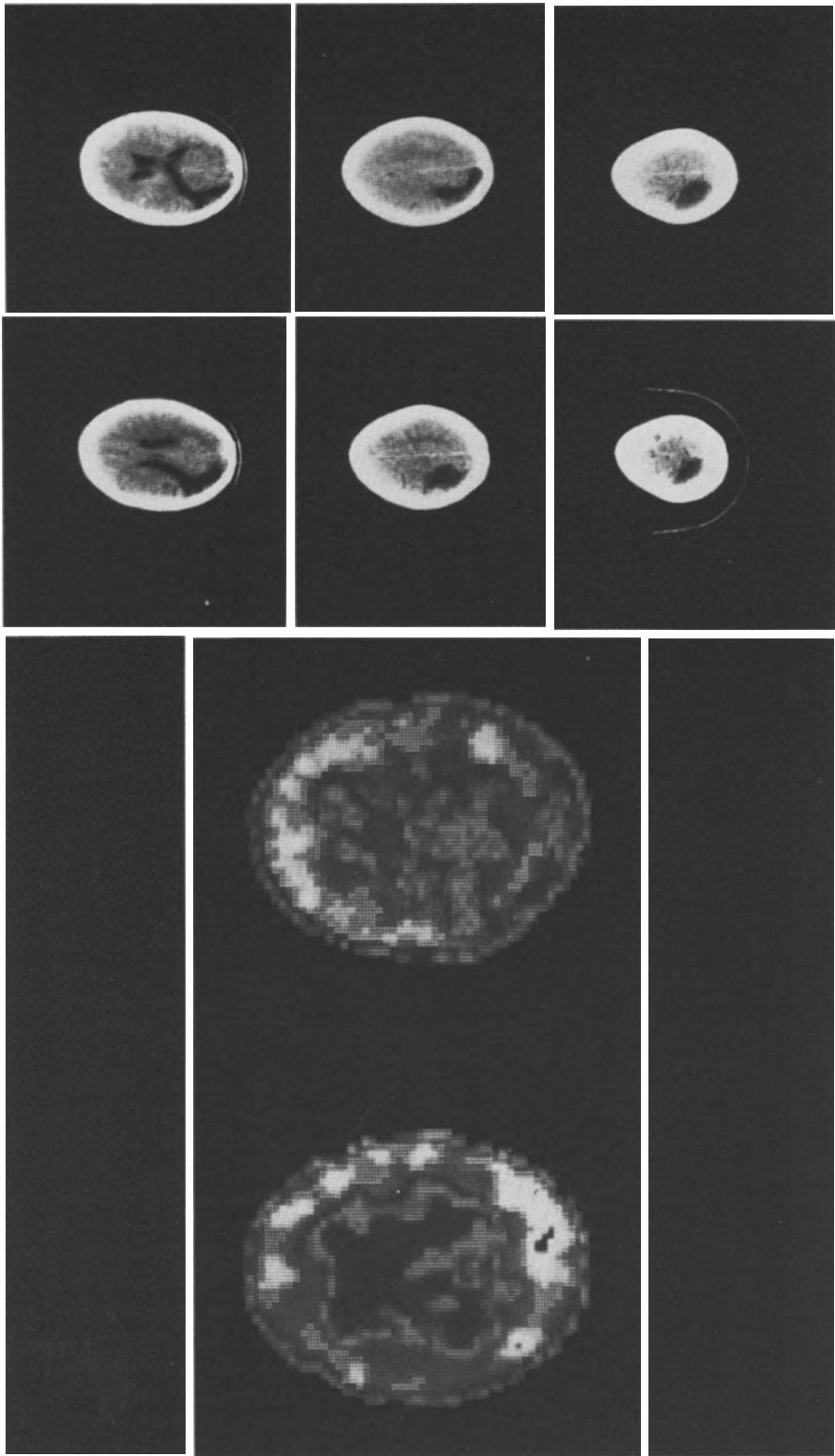


Fig. 4. CMG distribution images and CT findings from a patient with moyamoya disease

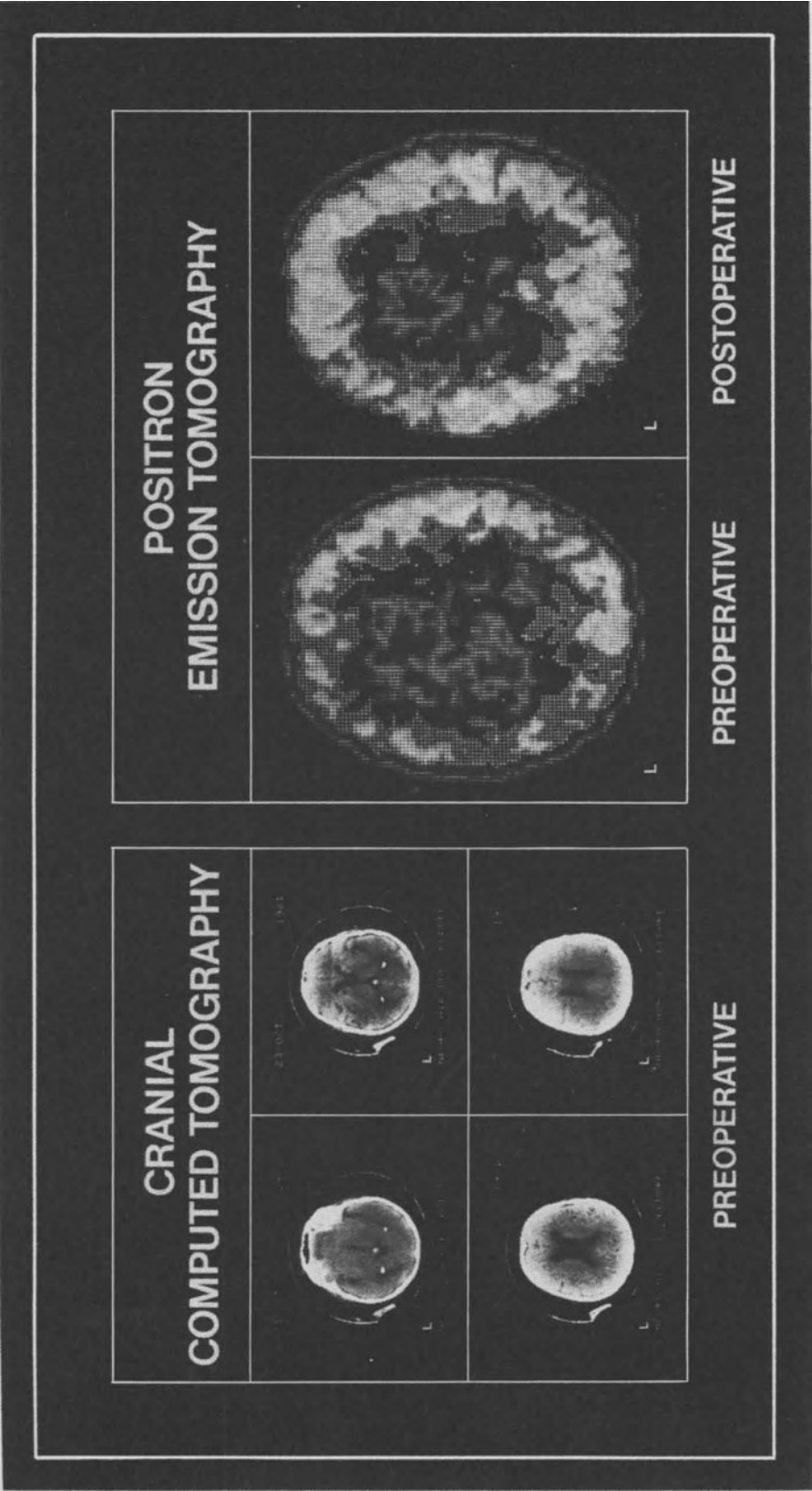


Fig. 1. CMG distribution images from a patient with ischemic stroke registered before and after an extracranial bypass operation

The results obtained in a 22-year-old female patient with moya-moya disease (multiple progressive intracranial arterial occlusion) are shown in Fig. 4. The only neurological finding in this case was homonymous hemianopia. The CT showed a hypodense area in the left occipital lobe indicating an old cerebral ischemic damage. Angiographically, a stenosis of the left internal carotid artery and of the left middle cerebral artery associated with formation of a fine network of abnormal collateral vessels was demonstrated, as well as a mild stenosis of the right middle cerebral artery. In CMG scans the activity accumulation defects could be recognized in both left and right hemisphere at different levels. These defects were significantly larger than the hypodense area in CT. In the residual noninfarcted left cortex that corresponded to the distribution of the left middle cerebral artery, the LPR (0.7 ml/min per gram) was reduced to 79% when compared with the opposite nonaffected hemisphere (0.88 ml/min per gram). The LUGTR, however, was practically the same as that in the opposite hemisphere. This was due to the significant increase of the rate constant for the glucose carrier system (140% when compared with the opposite hemisphere). This probably reflects a mechanism compensating for slowly developing perfusion deficit.

The results of a study done before and after extra-intracranial bypass surgery are demonstrated in Fig. 5. This study was carried out on a 72-year-old female patient (M.G.). In this case the initial symptom was a sudden onset of aphasia which lasted some minutes. In the following 4 months seven attacks were registered. In the later attacks the aphasia lasted up to 1 h: Angiographically, a "kinking" of the internal carotid artery without significant hemodynamic alteration in the Doppler sonogram and a severe stenosis of the left middle cerebral artery were demonstrated. In the CT scan no pathological evidence was found. In a preoperative CMG glucose scintigram large accumulation defects in the left cortex were evident. In this area, the LPR (0.79 ml/min per gram) was 10% lower than in the contralateral hemisphere (0.88 ml/min per gram). The LUGTR in this area was 44% lower than that in the opposite hemisphere. This unproportionally lower LUGTR was due to a lower value of the rate

constant for the glucose carrier system. Subsequent to the CMG scintigraphy, an extra-intracranial bypass was introduced. Postoperatively, a minor aphasic episode appeared for 2 days and then diminished. A Doppler sonogram indicated that the anastomosis was patent. The LPR in the treated hemisphere became normal (0.93 ml/min per gram) and was almost identical to the LPR in the opposite hemisphere (0.96 ml/min per gram). The LUGTR also increased, but did not achieve the values observed in the opposite hemisphere. The persisting lower value of LUGTR (18% reduction when compared with opposite hemisphere) was due to the persisting lower value of the rate constant for the glucose carrier system.

References

1. Bachelard HS (1971) Specificity and kinetic properties of monosaccharide uptake into guinea pig cerebral cortex. *J Neurochem* 18: 213–222
2. Betz LA, Gilboe DD, Yudilevich DL et al. (1973) Kinetics of unidirectional glucose transport into the isolated dog brain. *Am J Physiol* 225: 586–592
3. Betz AL, Gilboe DD, Drewes LR (1974) Effects of anoxia on net uptake and unidirectional transport of glucose into the isolated dog brain. *Brain Res* 67: 307–316
4. Bidder TG (1968) Hexose transport across the blood-brain interface: configurational aspects. *J Neurochem* 15: 867–874
5. Czaky TZ, Wilson JE (1956) The fate of 3-O- $^{14}\text{CH}_3$ -glucose in the rat. *Biochim Biophys Acta* 22: 185–186
6. Heiss WD, Vyska K, Kloster G et al. (1982) Demonstration of decreased functional activity of visual cortex by [^{11}C]methylglucose and PET. *Neuroradiology* 23: 45–47
7. Heiss WD, Kloster G, Vyska K et al. (1981) Regional cerebral distribution of ^{11}C CMG compared with CT perfusion patterns in stroke. *J Cereb Blood Flow Metabol* 1, Suppl 1: 506–507
8. Hinzen DH, Müller V, Sobotka P et al. (1972) Metabolism and function of dogs brain recovering from longtime ischemia. *Am J Physiol* 223: 1158–1164
9. Ingvar DA, Cronquist S, Ekberg R et al. (1965) Normal values of regional cerebral blood flow in man including flow and weight estimates of gray and white matter. *Acta Neur Scand* 41, Suppl 14: 72–84
10. Kennedy C, Sakurada O, Shinohara M et al. (1978) Local cerebral glucose utilisation in the normal conscious macaque monkey. *Ann Neurol* 4: 293–301

11. Kloster G, Müller-Platz C, Laufer P (1981) 3-¹¹C-Methyl-D-Glucose a potential agent for regional cerebral glucose utilisation. *J Lab Comp Radiopharm* 18: 855–863
12. Lund-Andersen H, Kjeldsen CS (1976) Kinetic analysis of the uptake of glucose analogs by rat brain cortex slices from normal and ischemic brain. In: Levi G, Battistin L, Lajtha A (eds) *Transport phenomena in the nervous system*. Plenum, New York London, 265–272
13. Narahara HT, Özand P, Cori CF (1960) Studies of tissue permeability. *J Biol Chem* 235: 3370–3378
14. Obrist WD, Thompson HK, King CH et al. (1967) Determination of regional cerebral blood flow by inhalation of ¹³³Xenon. *Circ Res* 20: 124–135
15. Pardridge WM, Oldendorf WH (1975) Kinetics of blood brain barrier transport of hexoses. *Biochim Biophys Acta* 382: 377–392
16. Phelps ME, Huang SC, Hoffman EJ et al. (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with FDG. *Ann Neurol* 6: 371–388
17. Phelps ME, Kuhl DE, Mazziotta JC (1981) Metabolic mapping of the brain's response to visual stimulation. *Science* 211: 1445–1448
18. Phelps ME, Mazziotta JC, Huang SC (1982) Study of cerebral function. *J Cereb Blood Flow Metabol* 2: 113–162
19. Reivich M, Greenberg J, Alavi A (1979) The use of fluorodeoxy-glucose technique for mapping of functional neural pathways in man. *Acta Neurol Scand* 60, Suppl 72: 198–199
20. Sokoloff L, Reivich M, Kennedy C et al. (1977) The ¹⁴C deoxyglucose method for the measurement of local cerebral glucose utilisation. *J Neurochem* 28: 897–916
21. Vyska K, Freundlieb C, Höck A et al. (1981) The measurement of glucose transport across the blood brain barrier in man by use of CMG. *J Cereb Blood Flow Metabol* 1, Suppl 1: 42–43
22. Vyska K, Freundlieb C, Höck A et al. (1982) Analysis of LPR and LUGTR in brain and heart in man by means of CMG and dPET. In: Höfer R, Bergmann H (eds) *Radioaktive Isotope in Klinik und Forschung*. Verlag H. Egermann, 15: 129–142
23. Whitfield CF, Rames RS, Morgan HE (1974) Acceleration of sugar transport in avian erythrocytes by catecholamines. *J Biol Chem* 249: 4181–4188

Glycolytic Rate and Histologic Grade of Human Cerebral Gliomas: A Study with [^{18}F]Fluorodeoxyglucose and Positron Emission Tomography

By G. DiChiro, R. A. Brooks, L. Sokoloff, N. J. Patronas, R. L. DeLaPaz, B. H. Smith, and P. L. Kornblith

Introduction

The spectrum of malignancy in cerebral gliomas is quite vast, from slow-growing, relatively benign lesions (e.g., astrocytoma grade I and certain oligodendrogliomas) to highly malignant tumors characterized by a fast growth rate (e.g., astrocytoma grade IV and glioblastoma multiforme) (Kernohan and Sayre 1952). In a number of cases the preoperative recognition of the various types of cerebral gliomas represents a diagnostic challenge. Among the neuro-radiological techniques, cerebral angiography and particularly transmission X-ray computed tomography (CT) provide substantial information in this area (Tchang et al. 1977; Joyce et al. 1978). Difficulties are encountered, however, even with CT, which may be inconclusive in the detection of the early tumor and in the discrimination of neoplastic tissue from edema (Kendall et al. 1979; Lilja et al. 1981). Discrepancy between histology and the preoperative CT diagnosis is frequent (Lilja et al. 1981). Finally, the assessment of tumor recurrence following surgery, radiotherapy, and chemotherapy may be a formidable task. Quite often we are not able to ascertain if the tumor is recurring and/or if the histological type of the lesion has changed.

A positive correlation between degree of malignancy and glycolysis has been reported in non-CNS tumors (Warburg 1930; Dickens and Simer 1930; Warburg 1956; Macbeth and Bekesi 1962; Weinhouse 1972; Weber 1977) and some evidence suggesting a similar relationship for experimental and human cerebral gliomas is available in the literature (Heller and Elliott 1955; Perria et al. 1964; Wolleman 1972; Kirsch et al. 1978; Timperley 1980).

Sokoloff et al. (1977) have developed a method for the quantitative determination of the local rates of glucose utilization throughout the central nervous system. This method is based on a kinetic model of the biochemical properties of

glucose and its analogue 2-deoxyglucose in brain tissue and utilizes tracer doses of [^{14}C]deoxyglucose and autoradiography.

2-Deoxyglucose competes with glucose for transport across the blood-brain barrier and for hexokinase-mediated phosphorylation to 2-deoxyglucose-6-phosphate (2-DG-6- PO_4). The further metabolism of deoxyglucose stops at this point, however, because in contrast to glucose-6-phosphate (G-6- PO_4), 2-DG-6- PO_4 is not a substrate for further metabolism, and remains essentially "trapped" in the cell. The Sokoloff method (1977) has been adapted for use with positron emission tomography (PET) by tagging 2-DG with ^{18}F , a positron emitter, to obtain the radiopharmaceutical 2-[^{18}F]fluoro-2-deoxy-D-glucose (FDG). PET with FDG as a tracer allows assessment of local cerebral glucose utilization in human patients (Reivich et al. 1979).

Considering the correlation between malignancy and glycolysis in primary cerebral tumors, we decided to carry out our FDG-PET studies in patients with those tumors (DiChiro et al. 1982). Our goals included detection of the gliomas, more precise characterization (grading), assessment of peritumoral or even remote (projected) metabolic alterations, and early recognition of recurrence and change of grade. Correlation between *in vivo* analysis of basic metabolic features (glycolysis) of the gliomas with similar tumor tissue culture studies under way in our laboratory represented an additional intriguing possibility.

Materials and Methods

Patients

Forty-four adult patients¹ harboring, or suspected of harboring, primary cerebral tumors

1 Our NIH-approved protocol does not allow studying patients below 18 years of age

were examined at the National Institutes of Health (NIH) with FDG-PET between May 1980 and December 1981. A number of these patients were studied more than once; comparison of different examinations in the same patient will not, however, be dealt with here. When multiple studies were available, the one closest in time to the verification or the most recent was selected for the purpose of this review. Table 1 lists the low-grade gliomas (grade I and II astrocytoma), while Tables 2 and 3 deal with the high-grade tumors (Table 2, grade III astrocytoma; Table 3, grade IV astrocytoma). In these tables, together with the peak tumoral metabolic rate, we have included the results of CT, technetium scans, and cerebral arteriography, as well as the mode of diagnosis and the available histologic grades. Whenever possible, CT and RN scans were carried out in close proximity to the FDG-PET scan, in most instances within 24 h to 1 week. The available angiograms, however, are dated at quite variable times from the PET scans.

At the time of the PET scan, all patients were being treated with anticonvulsants, most commonly phenytoin and phenobarbital, but also valproic acid, carbamazepine, and primidone; a number of patients were also being treated with oral steroids.

Radiopharmaceutical and Scan Procedure

The radiopharmaceutical (FDG) was prepared by the method of Ido et al. (1978), either at the nearby cyclotron facility of the Naval Research Laboratory (Washington, D.C.) or at Brookhaven National Laboratory (Upton, N.Y.), from which it was sent by special air carrier to an airport close to the NIH. Sterile and pyrogen-free (limulus test) FDG was administered intravenously over a 30-s period and, more recently, a 10-min interval. A maximum dose of 5 mCi (range 1.9–5.0 mCi) was administered. We have not seen any adverse reactions to FDG. Arterialized venous blood samples were obtained for the purpose of monitoring the FDG and glucose concentration in the plasma as a function of time. Sampling was done through a catheter placed in a hand vein (contralateral to the injection), which was heated to 40°C to produce arteriovenous shunting at the

fingertips as suggested by Phelps et al. (1979). In four cases, sampling was done directly from the cannulated radial artery.

All PET scans were done with the single-plane ECAT II positron emission computed tomograph (EG+G Ortec, Oak Ridge, Tennessee) (Williams et al. 1979) with standard shadow shields (medium resolution). The axial-transverse (horizontal) scan plane was oriented along the canthomeatal line by visual alignment with the laser beam built into the scanner. In most cases, scanning began 1 min after the start of the injection, with the scans taken at 2.5-min intervals for 30 min at the tumor level (determined by CT) to follow the kinetics of the FDG uptake. Subsequently, a series of longer scans (5–20 min each) was done at the same and other levels, usually at 10-mm increments, in order to obtain static images of the trapped tracer. The number of coincidence counts per image was generally 500 000–1 000 000. In a few instances scans in the coronal plane (perpendicular to the canthomeatal line) were obtained. The entire scanning procedure generally required 2–2.5 h.

Image reconstruction was performed on the ECAT console with the standard ellipse method of attenuation correction and the medium resolution convolution filter. The resulting spatial resolution in the image plane for the stated scanning mode and filter is 1.7 cm full width at half maximum (FWHM). The slice thickness was 1.95 cm FWHM. Images may be presented either in black and white or in a coded color display, each hue corresponding to a specific narrow range of metabolic rates (Gooche et al. 1980).

Calculation of Metabolic Rate

Image analysis was performed by the “region of interest” program provided with the ECAT instrument, as well as by a separate computer facility at the NIH. When discernible, the area of peak activity within the tumor was determined, with the CT images used as a guide, and numerical values (ECAT numbers) were obtained. If no clear peak area was evident, the highest reading within the tumor area was used, while excluding normal gray matter areas as much as possible (see treatment of the par-

tial volume effect in Discussion). The region of measurement was a circle with a diameter varying from 0.75 cm to 1.5 cm, depending on the size of the "hot" spot. The peak readings were used because the grade of a tumor is generally determined by its most malignant part. Therefore, just as the pathologist does with the histological grading, we attempted to identify and measure that part of the tumor which was the most metabolically active and presumably most viable. Normal gray and white matter values were obtained for this study from the frontal lobe of the opposite hemisphere in cases with only unilateral, low-grade tumors.

The ECAT numbers were converted to tissue activity in $\mu\text{Ci/cc}$ by means of calibration values obtained by scanning a phantom filled with a uniform FDG solution. The phantom concentration was determined with the same scintillation well counter used for plasma counting. The tissue activity was then converted to metabolic rate by the operational equation of the method. This equation utilizes the interpreted specific activity of the precursor (i.e., ratios of deoxyglucose to glucose concentrations) in the tissues, determined from the specific activity in plasma with a correction for the lag in the equilibration of the tissue pool with that of the plasma. It also corrects for the residual free unphosphorylated FDG remaining in the tissues at the time of the scan. An additional correction is required for dephosphorylation and loss of FDG-6- PO_4 ; this correction is very small for scans done within 1 h of injection, because the dephosphorylation is carried out by the enzyme, glucose-6-phosphatase, which is present in only small amounts in the brain (Sokoloff et al. 1977).

The equation for metabolic rate which incorporates all these factors is that of Sokoloff et al. (1977) and Huang et al. (1980), as modified by Brooks (to be published):

$$R = \frac{c_t}{LC} \frac{c_t(t) - B \int_0^t \exp[-\alpha_2(t-t')] c_p(t') dt'}{\int_0^t \beta \exp[-\alpha_1(t-t')] c_p(t') dt'} \quad (1)$$

In this equation $c_t(t)$ and $c_p(t)$ are the tissue and plasma concentrations of FDG at time t , c_g is the plasma concentration of glucose, LC is Sokoloff's lumped constant (Heller and Elliot

1955), and β , B , α_1 , and α_2 are simple constants related to the rate constants (k_1-k_4) of the model by the equations

$$\alpha_1 = 0.5 [k_2 + k_3 + k_4 - \sqrt{(k_2 + k_3 + k_4)^2 - 4k_2k_4}] \quad (2)$$

$$\alpha_2 = 0.5 [k_2 + k_3 + k_4 + \sqrt{(k_2 + k_3 + k_4)^2 - 4k_2k_4}] \quad (3)$$

$$\beta = [1 + (k_4 - \alpha_1)/k_3] [(1 + k_4/(k_2 + k_3))^2 - 4k_2k_4 / (k_2 + k_3)^2]^{-1/2} \quad (4)$$

$$B = k_1(\alpha_2 - k_3 - k_4)/(\alpha_2 - \alpha_1). \quad (5)$$

The values of LC and $k_1 - k_4$ reported by Huang et al. (1980) for normal white and gray matter were used in the calculations, with gray matter constants used for tumor calculations. The values of glucose metabolic rate are presented in conventional units of mg glucose/100 g tissue per minute, since the specific gravity of brain tissue is close to unity.

Results

The mean glucose utilization rates and the standard errors of the mean for the 13 cases measured for "normal" tissue were $R_{WM} = 3.3 \pm 0.2$ mg/100 g per minute (white matter) and $R_{GM} = 7.1 \pm 0.5$ mg/100 g per minute (gray matter).

The pathological results are grouped into three categories: grades I-II, grade III, and grade IV astrocytomas. The average rate of glucose utilization (Table 1) and the standard error of the mean for the 21 cases of low-grade tumor are: $R_{I-II} = 3.6 \pm 0.3$ mg/100 g per minute. This value is very close to that of normal white matter. For the high-grade cases (Tables 2, 3), we have $R_{III} = 5.2 \pm 0.9$ mg/100 g per minute and $R_{IV} = 7.1 \pm 1.1$ mg/100 g per minute.

The larger variation in rates for the high-grade gliomas (58% standard deviation) in grade IV may be partly due to differing degrees of partial volume artifact (see Discussion), as well as to a diversity of tumor stages. A plot of all cases is shown in Fig. 1.

Several statistical analyses were performed of the significance of the difference in mean metabolic rate among the three groups: a one-way

Table 1. Low-grade gliomas (astrocytomas I and II)

Case	Peak tumoral glucose utilization (mg/100 g per minute)	CT		RN scans (^{99m} Tc uptake)	Arterio-graphic findings ^a	Mode of diagnosis	Histologic grade
		Atten-uation (↑↓=)	Enhance-ment (+ -)				
1	1.2	↓	-	-	NA	Biopsy	I
2	2.9	=	+	NA	NA	Biopsy	II
3	4.4	=	+	+	NA	Biopsy	II
4	6.3	=	+	+	++	Biopsy	II
5	2.5	↓	-	NA	NA	Clinical and CT	I or II
6	3.5	↓	-	-	+	Clinical and CT	I or II
7	5.2	↑	-	NA	-	Clinical and CT	I or II
8	3.3	↓	-	-	-	Clinical and CT	I or II
9	3.9	↑	-	+	+	Biopsy	II
10	2.5	↓	-	NA	NA	Clinical and CT	I or II
11	3.3	↑	-	-	+	Biopsy	I
12	5.7	↑	-	NA	NA	Clinical and CT	I or II
13	3.9	↓	-	-	-	Clinical and CT	I or II
14	2.9	↑	-	+	NA	Clinical and CT	I or II
15	1.9	↑	-	-	-	Clinical and CT	I or II
16	1.3	↓	-	-	NA	Clinical and CT	I or II
17	2.5	↓	-	+	++	Biopsy	II
18	2.2	↓	-	NA	+	Clinical and CT	I or II
19	5.8	↓	-	NA	+	Clinical and CT	I or II
20	4.8	↓	-	-	++	Clinical and CT	I or II
21	5.3	↓	-	-	++	Biopsy	II
Mean	3.6						
SD	1.5						

^a ++, Vessel displacement and pathological vessels and/or blush; +, vessel displacement only

Table 2. High-grade gliomas (astrocytomas III)

Case	Peak tumoral glucose utilization (mg/100 g per minute)	CT		RN scans (^{99m} Tc uptake)	Arteriographic findings ^a	Mode of diagnosis
		Atten-uation (↑↓=)	Enhance-ment (+ -)			
1	11.6	↓	+	NA	NA	Biopsy
2	6.4	↓	+	NA	+	Biopsy
3	4.5	=	+	NA	NA	Biopsy
4	5.9	=	+	NA	NA	Biopsy
5	3.0	=	+	NA	+	Biopsy
6	2.8	↓	+	++	++	Biopsy
7	4.1	↓	+	+	+	Biopsy
8	2.5	↓	+	++	+	Biopsy
9	5.7	=	+	+	++	Biopsy
Mean	5.2					
SD	2.8					

^a ++, Vessel displacement and pathological vessels and/or blush; +, vessel displacement only

Table 3. High-grade gliomas (astrocytomas IV)

Case	Peak tumoral glucose utilization (mg/100 g per minute)	CT		RN scans (^{99m} Tc uptake)	Arteriographic findings ^a	Mode of diagnosis
		Attenuation (↓=)	Enhancement (+ -)			
1	10.2	=	+	++	++	Biopsy
2	7.5	=	+	++	+	Biopsy
3	15.9	↓	+	+	+	Biopsy
4	8.9	=	+	+	NA	Biopsy
5	5.9	=	+	++	++	Biopsy
6	5.1	=	+	NA	NA	Biopsy
7	7.9	=	+	++	NA	Biopsy
8 ^b	5.0	=	+	++	NA	Clinical and RN
9	14.9	=	+	+	++	Biopsy
10	4.5	=	+	++	NA	Biopsy
11	5.0	=	+	+	++	Biopsy
12	2.5	↓	+	+	NA	Biopsy
13	3.3	↓	-	+	++	Autopsy
14	3.2	↓	+	NA	NA	Biopsy
Mean	7.1					
SD	4.1					

^a ++, Vessel displacement and pathological vessels and/or blush; +, vessel displacement only.

^b Histological grade not known with certainty, but probably IV

analysis of variances on the rates, a similar one-way analysis on the log rates (because of the differences in sample variances among the groups), and a Kurskal-Wallis three-sample test using the ranks (because of possible non-normality). All tests were significant at the $p < 0.1$ level.

The visual distinction between the two categories was particularly striking. Most scans of high-grade tumors III and IV exhibited visually distinct hot spots (high activity) associated with the tumor. The appearance of the hot spots varied from large and sometimes multiple areas to barely distinguishable foci on the rim of the tumor. In cases where the tumor was adjacent to normal gray matter, especially cortex, which also has a high metabolic rate, it was sometimes not possible to discern a separate hot region. On the other hand, one of the low-grade tumors exhibited separate identifiable hot spots associated with the tumor.

Figure 2 shows a high-grade lesion (Table 3, case 4), a grade IV astrocytoma with a peak glucose metabolic rate of 8.9 mg/100 g per minute. Note the high activity in the viable tumor rim posteriorly and low uptake in the necrotic anterior portion. These findings were confirmed at

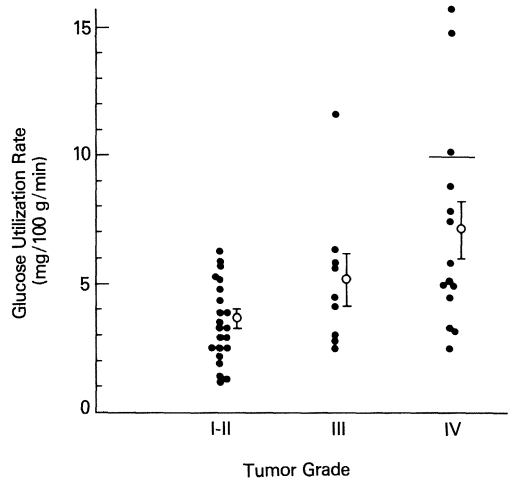


Fig. 1. Experimental values of glucose utilization rates for gliomas plotted as a function of tumor grade. The open circles represent the mean value for each tumor grade, and the error bars show the standard error of the mean. Most of the high-grade tumor cases with low metabolic rates (< 5 mg/100 g per minute) appear on CT with a thin (2–4-mm) viable rim surrounding a necrotic center suggesting an artifactual lowering of the measured rates due to the relatively poor spatial resolution of the tomograph

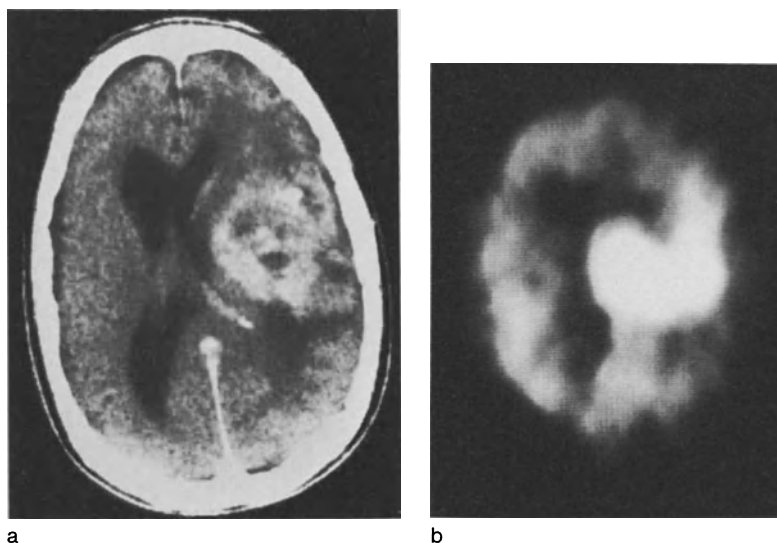


Fig. 2a, b. Here and in the following Figures **a** is a CT scan and **b** (and **c** in Fig. 4) is PET. **a** Right astrocytoma grade IV. **b** Peak tumoral metabolic rate = 8.9 mg/100 g per minute. Glycolytic activity is non-

homogeneous; higher in the autopsy-proven viable posterior tumoral rim, lower in the necrotic anterior portion. Cortical suppression adjacent to white matter edema is shown on CT

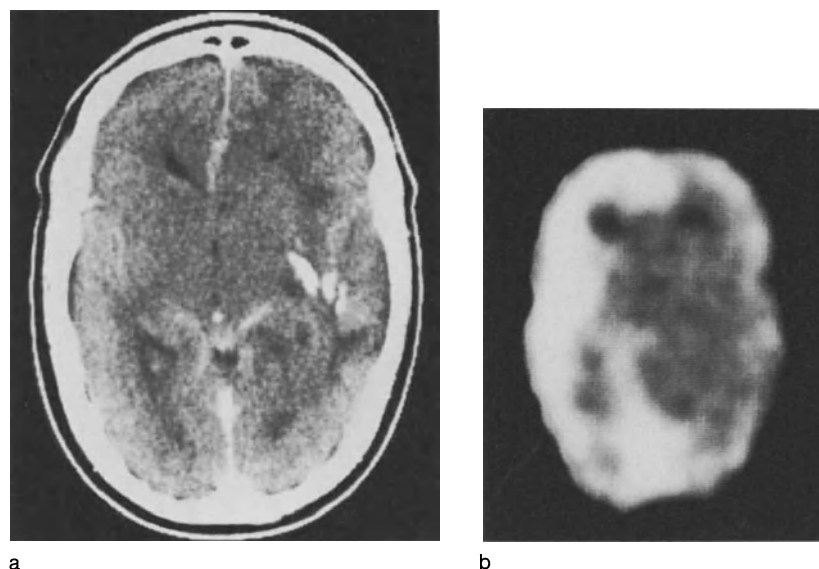


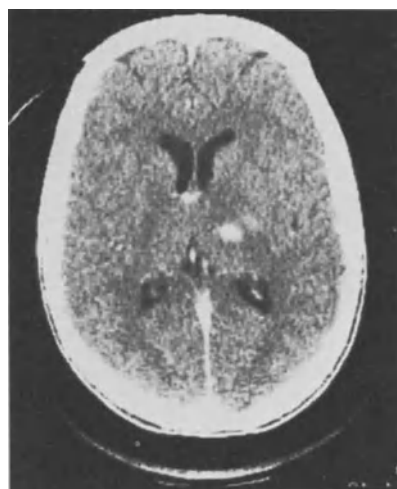
Fig. 3. a Right calcified astrocytoma grade II. **b** Peak tumoral metabolic rate = 3.9 mg/100 g per minute. Adjacent cortical suppression is probably due to

deafferentation, since cortical density is essentially normal on CT

autopsy. The overall cortical metabolic rate is very low (2–3 mg/100 g per minute), probably due to increased intracranial pressure (the patient died 2 days after the scan). An extensive low-grade tumor (Table 1, case 9) with a peak

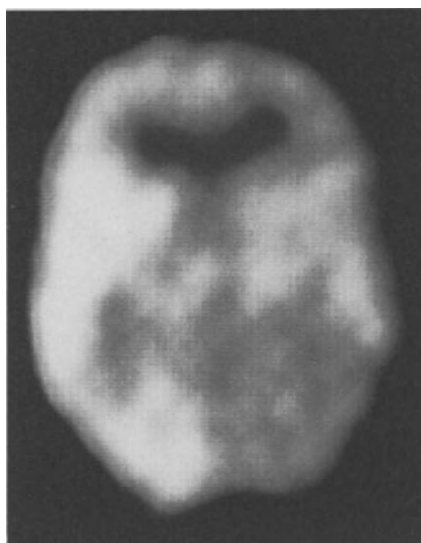
metabolic rate of 3.9 mg/100 g per minute is shown in Fig. 3.

In over half of the studies we noted a localized metabolic suppression in cortical regions either adjacent or remote, but connected neurally to

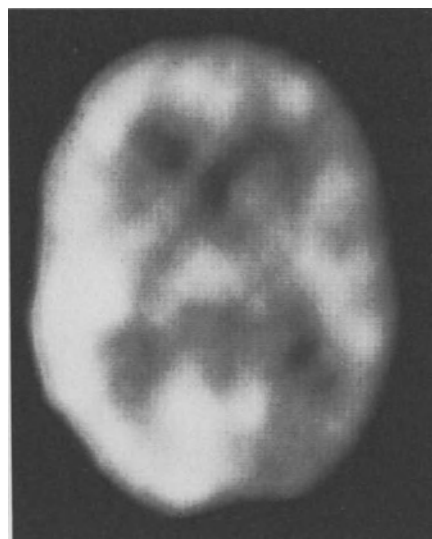


a

Fig. 4a–c. Striking example of parieto-temporo-occipital remote cortical suppression (b, c) in a partially calcified right thalamic astrocytoma grade II (a)



b



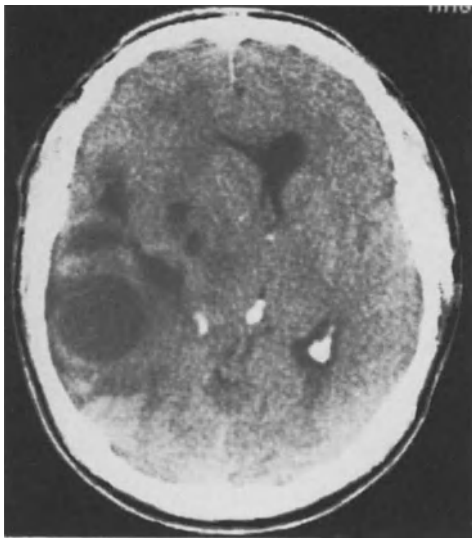
c

the tumor. One such example is seen in Fig. 4 (patient not included in present review), where suppressed activity of the right posterior cortex is associated with a grade II thalamic lesion. Suppressed adjacent cortex is also demonstrated in Figs. 2 and 3. In addition, hypometabolism in the contralateral cerebellar hemisphere was seen in several cases with large unilateral supratentorial lesions. Focal EEG irregular delta waves were associated with suppression in most cases.

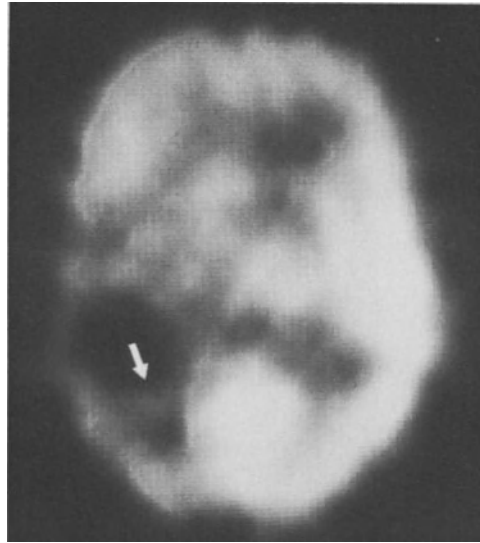
Discussion

Errors in Quantification

The most serious source of inaccuracy in the estimation of metabolic rate by FDG-PET is probably the partial volume effect (Hoffman et al. 1979). The resolution of the ECAT scanner in the mode used for most studies is 1.7 cm FWHM. While higher resolution modes are available, they result in fewer counts being collected and in noisier images. Because the skirts of the line-spread function extend well beyond



a



b

Fig. 5a, b. This left astrocytoma grade III (a) demonstrates well the partial volume artifact seen with a

thin tumoral rim (b; arrow) which outlines the cystic component and is surrounded by edema

the FWHM level, the partial volume effect causes errors even if the active area is uniform over a 1.7 cm diameter. If the tumor shows up on the PET scan with a hot area that is large compared to 1.7 cm (Fig. 2), the partial volume effect is not a problem. Aggressive gliomas, however, often present with a sizable cavitation component surrounded by a thin rim of solid tumor, generally the viable part, at the periphery. In these cases, the metabolic activity of the tumor will be underestimated because the reading will be influenced by the partial volume of necrotic component as well as by surrounding edema and normal white matter, which are necessarily included in the measurement due to the limited resolution of the scanner (Fig. 5). In contrast, in the low-grade gliomas with low metabolic activity, if the tumor is embedded in the gray matter, the resulting values are apt to be artifactually high. This is because of the partial volume of gray matter that is necessarily included in the measurement.

A second source of error is the self-absorption of the emitted gamma rays, which tends to depress the estimate of the concentrations toward the center of the image. While there is a correction program provided with the ECAT scanner based on an elliptical approximation to the

head, an error of 10% or greater may still remain (Huang et al. 1979). Additional errors are also present, including of course statistical fluctuations due to the finite number of counts available. For a small measurement region, the statistical error alone may be $\pm 10\%$.

Another cause of error is the assumption of nominal values for the rate constants ($k_1 - k_4$) and the lumped constant (LC). In fact the values for tumor tissue may be different from those in normal gray matter, because of possible differences in blood flow, transport distribution spaces, phosphatase activity, etc. Variations in the lumped constant directly affect R , while variations in the rate constant affect only the correction terms of Eq. (1), i.e., the term $B \int_0^t \exp[-\alpha_2(t-t')] c_p(t') dt'$ in the numerator and the factor $\beta \exp[-\alpha_1(t-t')]$ in the denominator. We found that even if white matter rate constants are used to calculate gray matter metabolic rates, and vice versa, the average error in R is only 4% (Brooks 1982), indicating that this source of error may be small. However, whatever the error introduced by pathological shifts in the constants, it does not detract from the clinical significance of the results. Even if the apparent increase in R for high-grade tumors were due entirely to shifts in the constants, its

usefulness for tumor grading would be unaltered.

Two pathological features merit separate discussion. One is the possibility that the tumors could be so hypovascular that their metabolic rates are limited by the undersupply of glucose. While cystic and calcified areas are obviously hypovascular, these regions were not included in our measurements. For the regions measured, the angiographic evidence, when available, indicated no severe hypovascularity, but in several of the low-grade tumors and most of the high-grade tumors, there was noticeable hypervascularity. In addition, the fact that the measured rates were comparable to or greater than that of normal white matter suggests that there is no significant limitation caused by hypovascularity. In any event the accuracy of our results is not affected by this possibility, since the reported rates represent actual glucose utilization, whether determined by tissue demand or blood flow limitations.

A second point to discuss is the possibility of increased phosphatase activity within the tumors, which would artifactually depress the calculated metabolic rates, particularly in high-grade tumors where one would expect the increased phosphatase to be greatest. If this possibility were true, then the difference in metabolic rates between the two tumor grades would be even greater than that reported here. However, preliminary results of a study of phosphatase in brain tumors now under way indicate that this is not the case. The study consists of comparing scans taken 2–3 h after injection with scans taken about 1 h after injection. If the phosphatase activity in the tumors were elevated, the apparent metabolic rates calculated from the later scans would be lower than those calculated from the earlier scans, but this has not been observed.

Finally, the effect of medications (anticonvulsants, steroids) on the cerebral glucose metabolic rate of our patients remains an imponderable element. The relative uniformity of the findings in each of the various tumor grades in the presence of different medication regimens would militate against a drug-limiting factor. Also, in no case in this series did we notice generalized hypometabolism which could be related to medications, as was occasionally observed in other PET studies when higher dos-

ages were administered (epilepsy, movement disorders).

One way to evaluate the overall error is to examine the deviations in normal tissue in all subjects. The standard deviation for normal white and gray matter in patients with low-grade tumors was 25%, about the same as reported in normal volunteers (Huang et al. 1980). Part of this variation is doubtless due to real metabolic differences and partly to the errors just described. The elevation of metabolic rate in the high-grade tumors exceeded the standard deviation.

Depressed Cortex

The tumor-associated cortical suppression of glucose utilization seen in our cases has not previously been described. This phenomenon, which was seen in over half of our cases, does not appear to have a clear relationship to tumor grade, tumor infiltration, satellitosis, or degree of cortical edema. Studies of surgical specimens of peritumoral tissue in human glioma have demonstrated an 8%–10% increase in gray matter volume and a 20%–25% increase in white matter volume (Reulen et al. 1969). The volume change alone would not account for the 20%–60% decreased cortical metabolic rate we have observed. Electrolyte changes as well as decreased pH with increased lactate in the peritumoral tissue may partly explain the decreased glycolytic rate (Schmiedek et al. 1974). Lowered peritumoral local cerebral blood flow may also contribute to this finding (Hossmann and Blöink 1981). The fact that there is an association between white matter involvement by tumor or edema and suppression in remote cortex linked by white matter tracts with the pathological site raises the question of deafferentation as a mechanism. It is of some interest that cortical suppression has also been observed soon after completion of radiation therapy, as well as in two cases of radiation necrosis. Further, ipsilateral thalamic suppression has been noted in several of our cases in association with tumors involving the cortex and craniotomy cortical defects. A detailed analysis of the subject of cortical suppression will be dealt with in a separate publication.

Relationship of FDG-PET Results to Other Studies

The positive correlation between tumor grade and glycolytic activity as determined by FDG-PET represents, to our knowledge, the first in vivo human verification of this relationship in man. It has been well established, however, in animal models (Warburg 1930; Dickens and Simer 1930; Weinhouse 1972) and in in vitro tissue culture studies (Dickens and Simer 1930; Macbeth and Bekesi 1962) of non-CNS neoplasms that the transformation from slowly to rapidly growing, poorly differentiated tumor tissue is accompanied by a progressive increase of glycolytic rate. Enzymatic changes in tumor tissue support these observations. For example, in rapidly growing experimental hepatomas, the activity ratio of hexokinase, which mediates the first step of glycolysis, to glucose-6-phosphatase, which reverses this step, is 88 times greater than in normal lines (Weber 1977). Similar results have been found in glioma cultures (Heller and Elliott 1955; Perria et al. 1964; Wolleman 1972; Kirsch et al. 1978; Timperley 1980).

Recently, observations in non-CNS animal tumors, both experimental and spontaneous, have shown high uptake of ^{14}C -DG (Larson et al. 1980) and ^{18}F -DG (Som et al. 1980). A similar positive correlation between glucose utilization and histologic grade has been demonstrated in experimental CNS neoplasms by ^{14}C -DG autoradiography (Blasberg et al. 1981; Yamada et al. 1981; Hossmann et al. 1982). In an ongoing study on a monkey model of primitive anaplastic gliomas, in which the glucose metabolic rate measured by FDG-PET is compared to ^{14}C -DG autoradiography, we are attempting to further validate the FDG-PET assessment of tumor grading. Also, glucose utilization and related enzymatic changes are being measured in vitro in tissue cultures of biopsy and autopsy material taken from cases in the present series. Results of these studies will be reported in subsequent publications.

Other biochemical parameters have been studied with PET, notably oxidative metabolism with $^{15}\text{O}_2$. In a few instances, low oxidative metabolism has been observed in high-grade gliomas (D.G.T. Thomas, personal communication). A combined PET analysis using both

FDG and $^{15}\text{O}_2$ may prove helpful in differentiating the high glycolytic activity in tumoral tissue from that of normal gray matter.

Acknowledgments. The services of Carlo Buonomo, Daniel C. Flynn, Juan J. Galarraga, Howard Golden, Christopher M. O'Connor, Anthony A. Waddock, and Wayne J. Yee were invaluable in conducting the scan procedures and data analysis. We thank Drs. A. B. Brill, Michael A. Channing, J. L. Doppman, R. M. Flynn, J. S. Fowler, E. George, Gerald S. Johnston, Robert M. Kessler, Conrad V. Kufita, A. J. Luessenhop, Ronald G. Manning, P. E. McKeever, E. A. Quindlen, K. G. Rieth, H. V. Rizzoli, and Alfred P. Wolf and Ms. C. S. Redvanly, Mr. R. W. Broseus, and Mr. R. J. Farkas for their support and cooperation. Dr. Victor J. Sank of the Neuroradiology and Computed Tomography Section was helpful in a number of technical aspects. The nuclear medicine technicians Janice Bird and Christine Straus showed their dedication by working many hours overtime. Mr. Cole Stathes handled and supervised a number of the organizational aspects.

References

1. Blasberg RG, Molnar P, Groothuis D, Patlak C, Fenstermacher J (1981) Simultaneous measurements of blood flow and metabolism. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S 68-S 69
2. Brooks RA (1982) Alternate formula for glucose utilization using labeled deoxyglucose. *J Nucl Med* 23: 538-539
3. Di Chiro G, DeLaPaz RL, Brooks RA, Sokoloff L, Kornblith PL, Smith BH, Patronas NJ, Kufta CV, Kessler RM, Johnston GS, Manning RG, Wolf AP (1982) Glucose utilization of cerebral gliomas measured by [^{18}F]fluorodeoxyglucose and positron emission tomography. *Neurology* 32: 1323-1329
4. Dickens F, Simer F (1930) The metabolism of the normal and tumor tissue. *Biochem J* 24: 1301-1326
5. Gooche C, Rasband W, Sokoloff L (1980) Computerized densitometry and color coding of ^{14}C -deoxyglucose autoradiography. *Ann Neurol* 7: 359-370
6. Heller IH, Elliott KHC (1955) The metabolism of normal brain and human gliomas in relation to cell type and density. *Can J Biochem Physiol* 33: 395-403

7. Hoffman EJ, Huang SC, Phelps ME (1979) Quantitation in positron emission computed tomography: 1. Effect of object size. *J Comput Assist Tomogr* 3: 299–308
8. Hossman KA, Blöink M (1981) Blood flow and regulation of blood flow in experimental peritumoral edema. *Stroke* 12: 211–217
9. Hossman KA, Niebuhr I, Tamura M (1982) Local cerebral blood flow and glucose consumption of rats with experimental gliomas. *J Cereb Blood Flow Metabol* 2: 25–32
10. Huang SC, Hoffman EJ, Phelps ME, Kuhl DE (1979) Quantitation in positron emission computed tomography: 2. Effects of inaccurate attenuation correction. *J Comp Assist Tomogr* 3: 804–814
11. Huang SC, Phelps ME, Hoffman EF et al. (1980) Noninvasive determination of local cerebral metabolic rate of glucose in man. *Am J Physiol* 238: E69–E82
12. Ido T, Wan CN, Cassela V et al. (1978) Labeled 2-deoxy-D-glucose analogs. ^{18}F -labeled 2-deoxy-2-fluoro-D-glucose, 2-deoxy-2-fluoro-D-mannose and ^{14}C -2-deoxy-12-fluoro-D-glucose. *J Labelled Compd Radiopharm* 24: 174–183
13. Joyce P, Bentson J, Takahashi M, Winter J, Wilson G, Byrd S (1978) The accuracy of predicting histologic grades of supratentorial astrocytomas on the basis of computerized tomography and cerebral angiography. *Neuroradiol* 16: 346–48
14. Kendall BE, Jakubowski J, Pullicino P et al. (1979) Difficulties in diagnosis of supratentorial gliomas by CAT scan. *J Neurol Neurosurg Psychiatry* 42: 485–92
15. Kernohan JW, Sayre GO (1952) Tumors of the central nervous system. In: *Atlas of tumor pathology*. A. F. I. P., Washington, sect 10, fasc 35
16. Kirsch WM, Tucker WS, Tabuchi K, Fink LM, Van Buskirk JJ, Low M (1978) The metabolism of the glioblastoma: pathological correlate. *Clin Neurosurg* 25: 310–325
17. Larson SM, Grunbaum Z, Rasey JS (1980) Positron imaging feasibility studies: selective tumor concentration of ^3H -thymidine, ^3H -uridine, and ^{14}C -2-deoxyglucose. *Radiology* 134: 771–773
18. Lilja A, Bergström K, Spännare B, Olsson Y (1981) Reliability of computed tomography in assessing histopathological features of malignant supratentorial gliomas. *J Comput Assist Tomogr* 5: 625–636
19. Macbeth RAL, Bekesi JG (1962) Oxygen consumption and anaerobic glycolysis of human malignant and normal tissue. *Cancer Res* 22: 244–248
20. Perria L, Viale G, Ibba F, Anreussi L, Viale E (1964) Istocitochimica dei tumori endocranici. *Neuropsychobiology* 20: 419–532
21. Phelps ME, Huang SC, Hoffman EF et al. (1979) Tomographic measurement of local cerebral glucose metabolic rate in humans with (^{18}F) 2-fluoro-2-deoxy-D-glucose: validation of method. *Ann Neurol* 6: 371–388
22. Reivich M, Kuhl D, Wolf A et al. (1979) The (^{18}F)fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
23. Reulen HJ, Medzihradsky F, Enzenbach R, Marguth F, Brendel W (1969) Electrolytes, fluids, and energy metabolism in human cerebral edema. *Arch Neurol* 21: 517–525
24. Schmiedek P, Baethmann A, Sippel G, Oettinger W, Enzenbach R, Marguth F, Brendel W (1974) Energy state and glycolysis in human cerebral edema. *J Neurosurg* 40: 351–364
25. Sokoloff L, Reivich M, Kennedy C et al. (1977) The (^{14}C) deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
26. Som P, Atkins HL, Bandoypadhyay D et al. (1980) Early detection of neoplasms with a radio-labeled sugar analog. *J Nucl Med* 21: 670–675
27. Tchang S, Scotti G, Terbrugge K, Melancon D, Belanger G, Milner C, Ethier R (1977) Computerized tomography as a possible aid to histological grading of supratentorial gliomas. *J Neurosurg* 46: 735–39
28. Timperley WR (1980) Glycolysis in neuroectodermal tumours. In: Thomas BT, Graham DI (eds) *Brain tumours: scientific basis, clinical investigation and current therapy*. Butterworth, London, pp 145–167
29. Warburg O (1930) *The metabolism of tumors*. Constable, London, pp 75–327
30. Warburg O (1956) On the origin of cancer cells. *Science* 123: 309–314
31. Weber G (1977) Enzymology of cancer cells. *N Engl J Med* 296: 486–493, 541–551
32. Weinhouse S (1972) Glycolysis, respiration, and anomalous gene expression in experimental hepatomas. *Cancer Res* 32: 2007–2016
33. Williams CW, Crabtree MC, Burgiss SG (1979) Design and performance characteristics of a positron emission computed axial tomograph – ECAT-II. *IEEE Trans Nucl Sci NS-26*: 619–627
34. Wolleman M (1972) Biochemistry of brain tumors. In: Lajtha A (ed) *Handbook of neurochemistry*. Plenum, New York pp 503–542
35. Yamada K, Hayakawa T, Ushio Y, Kato A, Arita N, Mogami H (1981) Regional blood flow, capillary permeability, and glucose metabolism in the rate with ethylnitrosourea induced rate gliomas. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: S571–572

Positron Emission Tomography Studies of Brain Energy Metabolism in Schizophrenia

By L. Widén, M. Bergström, G. Blomqvist, C. Bohm, E. Ehrin, S. Elander, K. Ericson, L. Eriksson, T. Greitz, D. H. Ingvar, J.-E. Litton, L. Nilsson, G. Sedvall, and F. Wiesel

Studies with regional cerebral blood flow (CBF) technique (Ingvar 1980; Ingvar and Franzén 1974) suggest differences in distribution between schizophrenic patients and healthy control subjects. The former were reported to show lower blood flow in frontal cortical regions.

A few recent PET studies on the metabolic pattern in schizophrenia (Buchsbaum et al. 1981; Brodie et al., this volume) support the above-mentioned CBF findings, i.e., they show a greater reduction in glucose utilization (CMR-Gl) in the frontal lobes relative to the more posterior regions of the brain than is seen in control cases.

In our own previous study (Widén et al. 1981), eight schizophrenic patients satisfying the research diagnostic criteria (RDC) for schizophrenia (Spitzer et al. 1977) with type I symptoms (Crow 1980) were examined. All except one were on neuroleptic drugs at the time of examination. A one-ring positron emission tomograph (Eriksson et al. 1978) was used for the measurements of the regional CMRGl in a brain slice at the level of the basal ganglia and approximately parallel to the orbitomeatal line. The ratio between the CMRGl values in the frontal and temporal cortices was calculated for each schizophrenic patient and two control subjects and a small but statistically significant difference between the two groups was found, the schizophrenic patients having a lower ratio. In the present study a further four patients and two controls are added to the previous material and a modification of the compartment model for the ^{11}C -glucose tracer is presented. The improvement of the preparation, purification, and analysis of the tracer is briefly commented upon.

Material and Methods

All four patients were suffering from their first psychotic episode and had never been treated with neuroleptic drugs. Only low doses of oxazepam and nitrazepam had occasionally been given before the time of examination. According to RDC, two of them (males, 18 and 35 years old) had Crow's type I schizophrenia, the other two (females, 23 and 28 years old) suffering from a schizo-affective disorder. In the latter two cases the duration of symptoms was 4 weeks, in the former 6 and 12 months respectively. The two control subjects were healthy 40-year-old males.

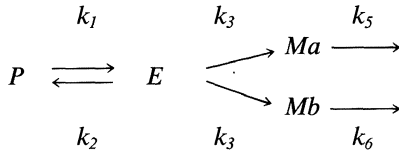
The method of preparation of ^{11}C -labeled glucose has been described previously (Ehrin et al. 1980). Briefly, it was as follows: The tracer was prepared photosynthetically using the green algae *Scenedesmus obtusiusculus* Chod. The carbohydrates were extracted from the cells using dilute HCl and the glucose isolated and purified using high-performance liquid chromatography. Further analysis of the material (gas-liquid and high-performance liquid chromatography) showed that the glucose obtained was radiochemically pure. The radiochemical yield of pure glucose was approximately 25%, based on the amount of $^{11}\text{CO}_2$ introduced to the algae. The complete procedure from the introduction of the radioactivity into the algae suspension to the final sterile injection was carried out in about 40 min.

During the scanning procedure the subjects lay with their eyes closed. The scanning room was quiet and the subjects were not spoken to, though a few of the patients had occasional brief comments or questions. The patients were continuously supervised and an EEG was recorded during each scan to check the level of wakefulness. Room lighting was very much reduced. Between 2 and 4 mCi of the tracer was injected intravenously and blood samples repeatedly taken from the contralateral, heated

arm vein. The "cold" glucose content as well as the activity of total ^{11}C and $^{11}\text{CO}_2$ were determined.

A four-ring positron emission tomograph giving seven tomographic slices (Eriksson et al., to be published) was used. Brain regions of interest were selected and marked with the aid of a cursor on a TV screen, showing the X-ray CT images of the same brain slice as the one examined with PET. The stereotactic coordinates of the CT picture were then transferred to the PET image by the computer. To allow accurate position transfer between CT and PET a special head-positioning device was used (Greitz et al. 1981).

To obtain local cerebral metabolic rates we apply a model with four compartments; tracer in the plasma (P), free glucose in the tissue (E), and two "parallel" compartments of metabolites (Ma and Mb):



The rate constants k_1 and k_2 describe the rate of transport of glucose over the blood-brain barrier and k_3 describes the rate of glucose-6-phosphate production. The free glucose is assumed to enter the compartments Ma and Mb in the ratio a : $(1-a)$ where a has a value between 0 and 1. The production rate of glucose-6-phosphate is thus $k_3 C_e$ irrespective of the value of the parameter a . The constants k_5 and k_6 describe the rate of CO_2 outflow from the brain. The reason for breaking up the compartment of metabolites into two parallel branches is the experimental observation that glucose labelled with ^{14}C in different positions produces $^{14}\text{CO}_2$ with different rates. Further, the metabolic chain consists of a large number of steps, but it is only the concentrations in the stages from which $^{11}\text{CO}_2$ is produced that affect the rate of $^{11}\text{CO}_2$ production. A single term $k_5 C_m^*(t)$ can never describe this situation adequately unless k_5 is made time-dependent. The introduction of two parameters k_5 and k_6 and two concentrations of metabolites gives a more flexible model.

The changes of the tracer concentrations in the different compartments are described by the following series of equations:

$$dC_e^*(t)/dt = k_1 dC_p^*(t) - (k_2 + k_3) dC_e^*(t) \quad (1)$$

$$dC_{Ma}^*(t)/dt = k_3 a dC_e^*(t) - k_5 dC_{Ma}^*(t) \quad (2)$$

$$dC_{Mb}^*(t)/dt = k_3 (1-a) dC_e^*(t) - k_6 dC_{Mb}^*(t) \quad (3)$$

The CMR of glucose is related to the rate constants and the amount of cold glucose in the plasma through the equation:

$$\text{CMR} = C_p k_1 k_3 / (k_2 + k_3) \quad (4)$$

The concentrations of tracer in the compartments E , Ma and Mb are related to the concentration of tracer in blood $C_b^*(T)$ and the regional tracer concentration $C_{iof}^*(T)$ (observed by the positron camera) through:

$$C_{iof}^*(T) = C_b^*(T) + C_e^*(T) + C_{Ma}^*(T) + C_{Mb}^*(T) \quad (5)$$

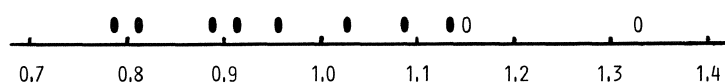
To determine the CMR-GI according to Eq. (4), the rate constants must be estimated. This is achieved by adjusting the rate constants so that Eq. (5) is fulfilled in the least square sense. The obtained metabolic values depend critically on the parameter a . Using $a = 0.5$ gives reasonable values (4–6 mg/100 g per minute) for the whole brain metabolic rate. The values $a = 0$ and $a = 1$ correspond to the previously used one-branch model [Eq. (5)], which gives considerably lower average metabolic rates of around 2.5 mg/100 g per minute.

In practice the parameter k_6 (forced to take large values) has only been fitted for the whole brain volume and the obtained value has then been used as a fixed parameter for the regional measurements.

Results

The ratio of frontal to temporal cortex CMRGI was measured. In our previously published study of eight patients and two controls (Widén et al. 1981), this ratio was significantly lower in the patient group. Fig. 1 illustrates the results obtained. With small regions of interest in the gray matter, corresponding to parts of the superior frontal gyrus and the superior and medial

SMALL CORTICAL REGIONS



LARGE CORTICAL REGIONS

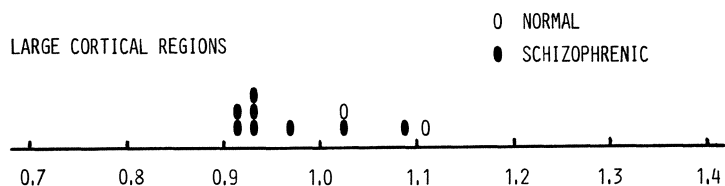


Fig. 1. Frontal to temporal ratio of CMRGI

temporal gyri, the level of statistical significance was higher ($p < 0.01$) than when large areas comprising the main parts of the frontal and temporal cortex were used ($p < 0.05$).

Within our new group of four patients and two controls there was no trend towards a reduced frontal/temporal metabolic ratio in the patients. Furthermore, when the old and the new groups were put together there was no significant difference in metabolic pattern between the 12 patients and the four controls.

Discussion

The results obtained in our recent study – admittedly based on a very small sample – differ from those of our previous study, as well as from those of Buchsbaum et al. (1981) and Brodie et al. (this volume). There are several possible explanations for this. First, the inter-individual variability may be so great that larger study groups are needed to demonstrate constant, significant differences between patients and controls. There are, however, systematic differences between our two groups of patients which may influence the results: The average duration of the disease was much longer in our first group (mean duration 7.5 years), these mainly chronic patients having either type I symptoms or type I and type II symptoms combined. The patients in the second group all had a short history and were considered to be “acute” cases, and two had a schizo-affective disorder. Furthermore, all but one of the pa-

tients in the first group were on neuroleptic drugs and in most cases the medication had been taken for a long time. In contrast, none of the four new patients had ever been on neuroleptic drugs.

Comparing our findings with those of Brodie et al. (this volume) it is noteworthy that all except two of their patients belonged to Crow's type II, yet the “hypofrontality” found was similar to that in our first group. Some of Brodie's patients had apparently never received any neuroleptic treatment.

Trying to find a common denominator to these rather disparate observations, one is struck by the fact that acute, untreated cases of schizophrenia do not seem to show signs of hypofrontality, whereas schizophrenia of a more chronic kind may display an altered cerebral metabolic pattern with relatively decreased energy metabolism in the frontal areas, regardless of whether they are labeled type I or II according to Crow's classification.

Against the above hypothesis is the fact that five of the eight patients studied by Buchsbaum et al. (1981) – all Crow's type I – had a duration of symptoms of a year or less, and therefore could hardly be called chronic, yet showed lower ratios in the frontal cortex, indicating lower glucose use than normal controls. They had all been on neuroleptic treatment, but the medication was discontinued 2 weeks prior to the PET examination. It cannot, however, be excluded that some effects of the neuroleptic treatment on brain function remain longer than 2 weeks.

Hypothetically, therefore, it may be assumed

that the hypofrontal pattern develops in long-standing cases of schizophrenia and in cases involving current treatment or recent prolonged treatment with neuroleptic drugs.

Obviously the samples are too small and heterogeneous to allow anything but speculation concerning the mechanisms and significance of the hypofrontal pattern. Continued research with the PET technique is necessary, selecting the schizophrenic patients carefully on the basis of strictly defined criteria. Comparative studies should be made of patients before, during, and after treatment with neuroleptic drugs. It is to be hoped that significant relationships will be found between changes in metabolic patterns on one hand and subjective symptoms, clinical signs, and psychometric findings on the other.

In our own study the analysis of the cerebral energy metabolic patterns shown in the PET images is still at a preliminary stage. A systematic search for patterns of correlation between various precisely defined brain areas with regard to their CMRgl will be performed in the future.

References

1. Buchsbaum MS, Kessler R, Bunney WE, Cappelletti J, Coppola R, van Kammen DP, Rigal F, Waters R, Sokoloff L, Ingvar DH (1981) Simultaneous electroencephalography and cerebral glucose metabolism with positron emission tomography (PET) in normals and patients with schizophrenia. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 457-458
2. Crow TJ (1980) Molecular pathology of schizophrenia: more than one disease process? *Br Med J* 260: 66-68
3. Ehrin E, Westman E, Nilsson SO, Nilsson JLG, Widén L, Greitz T, Larsson CM, Tillberg JE, Malmberg P (1980) A convenient method for production of 11-C-labelled glucose. *J Labelled Compd Radiopharm* 17: 453-461
4. Eriksson L, Widén L, Bergström M, Bohm C, Floom G, Greitz T, Hovander B, Litton J (1978) Evaluation of a high resolution ring detector positron camera system. *J Comput Assist Tomogr* 2: 649
5. Eriksson L, Bohm Ch, Kesselberg M, Blomqvist G, Litton J, Widén L, Bergström M, Ericson K, Greitz T (to be published) A four ring positron camera system for emission tomography of the brain. *IEEE Trans Nucl Sci*
6. Greitz T, Bergström M, Boethius J, Eriksson L, Nilsson L, Ribbe T, Uggla M, Widén L (1981) A method for reproducible position alignment in transmission CT and positron emission tomography. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 48-49
7. Ingvar DH (1980) Abnormal distribution of cerebral activity in chronic schizophrenia: a neurophysiological interpretation. In: Baxter C, Melnechuk T (eds) *Perspectives in schizophrenia research*. Raven, New York pp 107-130
8. Ingvar DH, Franzén T (1974) Abnormalities of cerebral blood flow distribution in patients with chronic schizophrenia. *Acta Psychiatr Scand* 50: 425-462
9. Spitzer R, Endicott J, Robin E (1977) Research diagnostic criteria: rationale and reliability. *Arch Gen Psychiatry* 35: 773-782
10. Widén L, Bergström M, Blomqvist G, Brismar T, Ehrin E, Elander S, Eriksson K, Eriksson L, Greitz T, Litton J-E, Malmberg P, Nilsson L, Sedvall G, Uggla M (1981) Glucose metabolism in patients with schizophrenia; emission computed tomography measurements with 11-C-glucose. *J Cereb Blood Flow Metabol* 1 [Suppl 1]: 455-456

Regional Correlations in Patterns of Glucose Use in Patients with Schizophrenia and Normal Subjects During Mild Pain Stimulation

By R. M. Kessler, C. M. Clark, M. S. Buchsbaum, H. Holcomb, R. A. Margolin, J. Cappeletti, M. Channing, R. G. Manning, D. P. van Kammen, A. C. King, and J. Johnson

Abbreviations in Tables

AMF	Anterior medial frontal
RAF	Right anterior frontal
LAF	Left anterior frontal
RPF	Right posterior frontal
LPF	Left posterior frontal
LRol	Left rolandic
RRol	Right rolandic
RAP	Right anterior parietal
LAP	Left anterior parietal
LPPO	Left posterior parieto-occipital
RPPO	Right posterior parieto-occipital
MMC	Middle medial cortex
MOP	Medial occipitoparietal
LP	Left parietal
RP	Right parietal
LO	Left occipital
RO	Right occipital
O	Occipital
RAT	Right anterior temporal
LAT	Left anterior temporal
RMT	Right middle temporal
LMT	Left middle temporal
RBG	Right basal ganglia
LBG	Left basal ganglia
RTh	Right thalamus
LTh	Left thalamus
LSyl	Left sylvian
RSyl	Right sylvian
LPT	Left parietotemporal
RPT	Right parietotemporal

Introduction

The ability to study patterns of cerebral metabolism in humans noninvasively is clearly one of the great advantages of positron tomography. The development of the ^{18}F -labeled 2-fluorodeoxyglucose (^{18}F FDG) method has produced a powerful tool for studying cerebral energy metabolism with positron tomography (Huang

et al. 1980). Given the tight coupling normally existing between cerebral function and energy metabolism (Sokoloff 1977, 1981), ^{18}F FDG has been used to study resting patterns of metabolism in resting subjects (Mazziotta et al. 1981), subjects undergoing neuropsychological activations (Greenberg et al. 1981), and schizophrenic subjects at rest and off medication (Buchsbaum et al. 1982).

In the present study, scans with ^{18}F FDG were performed following a cerebral activation procedure – mildly painful electric shock applied to the right forearm – on seven schizophrenic and nine normal subjects. Shock stimulation was chosen because of the relative pain insensitivity in schizophrenics (Davis et al. 1979) and reports that pain stimulation failed to activate frontal blood flow increases in schizophrenics (Ingvar et al. 1975; Ingvar 1980). Regional glucose metabolism in normal subjects and schizophrenics was compared region by region. The interrelations of brain regions in terms of glucose metabolism were then examined by means of correlational analysis.

Methods

Seven schizophrenics (off medication for at least 2 weeks) and nine age-appropriate normal subjects were studied. All subjects were right-handed and the normals had no history of personal or first-degree familial mental illness. The initial portion of the study was performed with the subjects seated in a quiet, darkened room, with the left forearm heated for arterialized venous sampling. Intravenous lines were placed in both arms. Brief electric shocks of a mildly painful nature (Buchsbaum et al. 1981), were administered at 1-s intervals, beginning after the injection of 3–5 mCi ^{18}F FDG and continuing for 30–35 min following injection. The subject was then taken to the scanning room. Scans

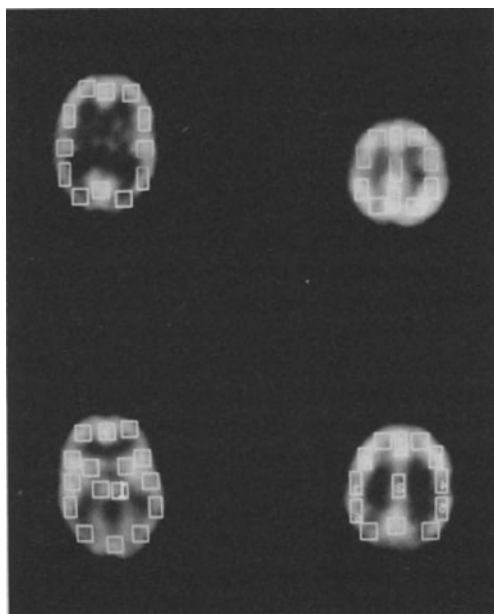


Fig. 1. Template of regions

were performed with an ECAT II scanner (E. G. and G. ORTEC) with medium resolution shadow shields, scan mode, and reconstruction filters to give an in-slice resolution of 1.75 cm and an axial resolution of 1.78 cm. Seven to eight planes were scanned parallel to the canthomeatal (CM) line over a 50- to 80-min period. Glucose metabolic rates were calculated using the Sokoloff equation (Sokoloff et al. 1977) and a ratio of regional to whole plane metabolism was calculated. This ratio was used for the statistical analysis.

Four planes were selected from each study at average heights above the CM line of 91, 78, 65, and 52 mm. A total of 49 regions of interest were selected from each subject, as shown in the standard templates (Fig. 1). The rectangular regions were of varying size (1.71×1.71 cm, 1.14×2.57 cm, or 1.29×2.29 cm) but of constant area (2.9 cm^2).

The data obtained from these regions were then analyzed statistically by two procedures. First, the means of the individual regions, in controls and schizophrenics, were compared by means of two-tailed *t* tests with alpha set at $p < 0.05$. Second, the glucose metabolic rates among regions within slices were correlated. Therefore for each slice the relations among

areas could be estimated. The correlations were calculated for both groups combined and the correlations for the respective groups were compared by using *t* tests on the Fisher Z transformations of the correlation coefficients.

Results and Discussion

Comparison of Group Means

The means of each of the forty-nine regions were compared between normals and schizophrenics by means of *t* tests. Glucose utilization in the left basal ganglia was significantly lower in schizophrenics (left basal ganglia to whole slice ratio of 1.15) than in normals (left basal ganglia to whole slice ratio of 1.25). This same region was found to be lower in schizophrenics than normals on resting studies (Buchsbaum et al. 1982). Comparisons of anteroposterior ratios and ANOVA on log normalized glucose values are reported elsewhere (Buchsbaum et al. to be published).

Correlational Analyses

Correlational analysis was initially performed on a matrix which combined both schizophrenic and normal subjects. The combined correlation matrices yielded several significant correlations. On the 91-, 78-, and 65-mm planes all five frontal lobe regions were positively and/or significantly intercorrelated, suggesting that the frontal regions tend to function in synchrony (Tables 1–3). It is also interesting to note that on both 78- and 65-mm planes, changes in the left posterior frontal cortex were significantly and positively correlated with changes in the parietal regions bilaterally. On all three planes there were negative correlations between frontal metabolism and the posterior parieto-occipital and lateral occipital regions. Five of the frontal/posterior correlations reached significance over the three planes. This finding is consistent with previous studies showing that rates of metabolism (Buchsbaum et al. 1982) and blood flow are higher in the anterior frontal region than in the posterior parietal and occipital regions and that the difference is greater

Table 1. Correlation coefficients between regions on the 91-mm plane for normal and schizophrenic subjects combined

	AMF	LAF	RAF	LPF	RPF	LAP	RAP	LPPO	RPPO	MMC	MOP
AMF	1.0	0.42	0.64*	0.48	0.43	-0.16	-0.38	-0.30	-0.39	0.45	0.40
LAF		1.0	0.73*	0.45	0.62*	-0.16	0.24	-0.55*	-0.42	0.01	0.08
RAF			1.0	0.30	0.46	-0.32	0.18	-0.32	-0.37	0.38	0.22
LPF				1.0	0.64*	0.11	-0.44	-0.37	-0.46	-0.04	0.01
RPF					1.0	0.10	0.08	-0.50*	-0.34	-0.27	-0.03
LAP						1.0	-0.25	0.20	-0.04	-0.13	-0.37
RAP							1.0	-0.18	0.04	-0.40	0.15
LPPO								1.0	0.57*	0.29	-0.22
RPPO									1.0	0.10	0.18
MMC										1.0	0.28
MOP											1.0

Correlations reaching significance: 7/55

Table 2. Correlation coefficients between regions on the 78-mm plane for normal and schizophrenic subjects combined

	AMF	LAF	RAF	LPF	RPF	LRol	RRol	LP	RP	LO	RO	MMC	O
AMF	1.0	0.69*	0.38	0.50*	0.56*	0.01	0.19	0.31	0.06	-0.57*	-0.28	0.03	0.37
LAF		1.0	0.64*	0.57*	0.44	0.31	0.23	0.62*	0.23	-0.47	-0.44	-0.32	0.25
RAF			1.0	0.44	0.43	0.50*	0.57*	0.35	-0.03	-0.61*	-0.56*	-0.11	-0.20
LPF				1.0	0.55*	0.63*	0.55*	0.57*	0.54*	-0.24	-0.20	-0.28	0.13
RPF					1.0	0.49	0.63*	0.38	0.47	-0.12	-0.34	0.05	0.30
LRol						1.0	0.66*	0.43	0.43	-0.08	-0.09	-0.41	-0.01
RRol							1.0	0.23	0.47	-0.36	-0.43	-0.25	-0.05
LP								1.0	0.40	0.07	0.03	-0.58*	0.45
RP									1.0	-0.03	-0.14	-0.20	0.26
LO										1.0	0.76*	-0.03	0.22
RO											1.0	-0.29	0.09
MMC												1.0	-0.16
O													1.0

Correlations reaching significance: 20/78

Table 3. Correlation coefficients between regions on the 65-mm plane for normal and schizophrenic subjects combined

	AMF	LAF	RAF	LPF	RPF	LSyl	RSyl	LPT	RPT	LO	RO	O
AMF	1.0	0.71*	0.77*	0.43	0.51*	0.34	0.24	-0.10	0.23	-0.30	-0.38	-0.04
LAF		1.0	0.64*	0.52*	0.46	0.24	-0.13	0.13	0.32	-0.22	-0.45	-0.10
RAF			1.0	0.02	0.42	0.23	0.28	-0.30	0.20	-0.19	-0.23	0.13
LPF				1.0	0.41	0.27	-0.06	0.58*	0.50*	0.14	0.02	0.31
RPF					1.0	0.10	-0.10	0.16	0.21	-0.11	-0.09	-0.06
LSyl						1.0	0.68*	-0.03	-0.08	-0.54*	-0.43	-0.12
RSyl							1.0	-0.34	-0.18	-0.38	-0.16	0.03
LPT								1.0	0.47*	0.64*	0.50*	0.74*
RPT									1.0	0.37	0.21	0.56
LO										1.0	0.89*	0.74*
RO											1.0	0.74*
O												1.0

Correlations reaching significance: 16/66

Table 4. Correlation coefficients between regions on the 52-mm plane for normal and schizophrenic subjects combined

	MF	LAF	RAF	LPF	RPF	LAT	RAT	LMT	RMT	LBG	RBG	LTh	RTh
MF	1.0	0.70*	0.85*	0.37	0.31	-0.28	0.35	-0.17	0.76*	-0.52*	-0.61*	-0.19	0.00
LAF		1.0	0.55*	0.57*	0.08	-0.06	0.35	0.05	0.44	-0.43	-0.60*	-0.10	0.03
RAF			1.0	0.13	0.43	-0.51*	0.34	-0.42	0.70*	-0.27	-0.33	-0.28	-0.43
LPF				1.0	0.08	0.44	0.56*	0.25	0.01	-0.37	-0.38	0.38	0.37
RPF					1.0	-0.27	0.22	-0.05	0.06	0.08	0.20	-0.29	-0.33
LAT						1.0	-0.05	0.72*	-0.37	-0.07	-0.22	0.37	0.64
RAT							1.0	-0.06	0.10	-0.27	0.03	0.19	0.17
LMT								1.0	0.06	-0.19	-0.29	0.56*	0.45
RMT									1.0	-0.45	-0.53	-0.08	-0.30
LBG										1.0	0.80	0.21	0.24
RBG											1.0	-0.29	0.08
LTh												1.0	0.89*
RTh													1.0

Correlations reaching significance: 18/78

(Ingvar 1980) in resting normals than in schizophrenics.

On the 52-mm plane through the basal ganglia and thalamus (Table 4) the frontal regions were again positively and significantly correlated. The anterior medial frontal cortex and the left anterior frontal cortex were negatively correlated with the basal ganglia; three of the four correlations were significant. The right anterior and left posterior frontal regions were also negatively correlated with the basal ganglia, but not significantly so. The clustering of negative correlations suggests an inverse metabolic relation between the anterior frontal lobes and the basal ganglia. Other less clustered but significant correlations can be seen on this plane. The combined correlation matrix demonstrates where schizophrenic and normal subjects may have similar relations between brain regions, although perhaps not to the same extent in both groups. Because of the small numbers of subjects (seven schizophrenic and nine normal), only very large differences in correlation coefficients between groups became significant at the $p < 0.05$ level. Fourteen significant differences were found between groups for the four planes analyzed (Table 5).

On the 91-mm plane, four significantly different correlations were found. Three of the four involved the middle medial cortex. This region appears to correspond to Brodmann's areas 4 and 6. It was negatively coupled with the right posterior frontal region in normals, whereas schizophrenics showed a positive correlation.

Table 5. Correlation coefficients in brain regions showing a significant difference between normals and schizophrenics

Height of plane above CM line (mm)	Regions	Schizophrenics	Normals
91	MMC:RPF	0.25	-0.79
91	MMC:LPP	-0.77	0.68
91	MMC:RPP	-0.68	0.47
91	RAF:MOP	0.77	-0.48
78	LRol:AMF	-0.75	0.55
78	LRol:RAF	-0.16	0.84
78	LRol:LAF	-0.71	0.90
78	LRol:LPF	-0.22	0.88
78	LRol:LP	-0.58	0.67
78	RRol:AMF	-0.59	0.58
78	RP:RPF	-0.69	0.61
78	RPF:LPF	-0.49	0.74
78	RO:LO	0.96	0.42
52	RAF:RAT	0.86	-0.12

Coefficients are shown for which a significant ($p < 0.05$) difference was found between normals and schizophrenics.

Both posterior parietal regions were correlated positively with the middle medial cortex in normals but negatively in schizophrenics. Correlations involving the middle medial cortex and the three anterior frontal regions, which were just below significance, showed a positive correlation in normals and a negative one in schizophrenics. The middle medial cortex has been implicated in aversive and postural movements (Peele 1981). This is suggestive of a difference in the motor response to shock in schi-

zophrenics than in normals. One other significant difference in correlation coefficients was seen between the right anterior frontal cortex and the medial parieto-occipital region. These were positively coupled in schizophrenics and negatively coupled in normals.

On the 78-mm plane, nine significantly different correlations were seen, six of which involved the rolandic regions. The left rolandic region and the anterior medial frontal cortex, right anterior frontal cortex, left anterior frontal cortex, and left posterior frontal cortex were correlated positively in normals and negatively in schizophrenics. The left rolandic area at this level is about where one would expect cerebral activation to occur with right forearm shock. These differences in correlations suggest the relation between the primary sensory region and the frontal lobes is quite different in schizophrenics and normals.

Differences in correlation coefficients – positive in normals and negative in schizophrenics – were seen between left rolandic region and left parietal cortex, between right rolandic region and anterior medial cortex, between right and left posterior frontal regions, and between right parietal and right posterior frontal regions. The left and right lateral occipital regions were highly positively correlated in schizophrenics but much less so in normals.

No significantly different correlations were seen on the 65-mm plane. Only one significantly different correlation was apparent on the 52-mm plane, the right anterior frontal region being positively coupled with the right anterior temporal region in schizophrenics but not in normals. These results must be considered preliminary as we have only a small study sample and the data has been only partly analyzed. Nonetheless, there is a strong suggestion that differences between normals and schizophrenics may be found by examining relations between regions, rather than only evaluating average metabolic rates.

References

1. Buchsbaum MS, Davis GC, Coppola R, Naber R (1981) Opiate pharmacology and individual differences II. Somatosensory evoked potentials. *Pain* 10: 367–377
2. Buchsbaum MD, Ingvar DH, Kessler RM et al. (1982) Cerebral glucography with positron tomography in normals and patients with schizophrenia. *Arch Gen Psychiatry* 39: 251–259
3. Buchsbaum MS, Holcomb H, Kessler RM et al. (to be published) Lateralized asymmetries in glucose uptake assessed by positron emission tomography in patients with schizophrenia and normal controls. In: Flor-Henry P, Gruzeller J (eds) *Hemispheric asymmetries of function in psychopathology*, vol II. Elsevier Biomedical, Amsterdam
4. Davis GC, Buchsbaum MS, van Kammer DP, Bunney WE (1979) Analgesia to pain stimuli in schizophrenics and its reversal by naltrexone. *Psychiatry Res* 1: 61–70
5. Greenberg JH, Reivich M, Alavi A et al. (1981) Metabolic mapping of functional activity in human subjects with the ^{18}F fluorodeoxyglucose technique. *Science* 212: 678–680
6. Huang SC, Phelps ME, Hoffman EJ et al. (1980) Noninvasive determination of local cerebral metabolic rate of glucose in man. *Am J Physiol* 238 (Endocrinol Metab 1): E 69–E 82
7. Ingvar DH (1980) Abnormal distribution of cerebral activity in chronic schizophrenia, a neurophysiological interpretation. In: Baxter C, Melnechuk T (eds) *Perspectives in schizophrenia research*. Raven, New York
8. Ingvar DH, Rosen I, Elmquist D (1975) Effects of somatosensory stimulation upon rCBF. In: Harper M, Jennett B, Miller D, Rowan J (eds) *Blood flow and metabolism in the brain*. Churchill Livingstone, New York
9. Mazziotta JC, Phelps ME, Miller J, Kuhl DE (1981) Tomographic mapping of human cerebral metabolisms: normal unstimulated state. *Neurology* 31: 503–516
10. Peele TL (1981) *The Neuroanatomic Basis for Clinical Neurology*, 2nd edn. McGraw-Hill, New York
11. Sokoloff L (1977) Relation between physiological function and energy metabolism in the central nervous system. *J Neurochem* 29: 13–26
12. Sokoloff L (1981) Relationships among local functional activity, energy metabolism, and blood flow in the central nervous system. *Fed Proc* 40: 2311–2316
13. Sokoloff L, Reivich M, Kennedy C et al. (1977) The (^{14}C) deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure, and normal values in the conscious and anaesthetized rat. *J Neurochem* 28: 897–916

Evaluation of Regional Glucose Metabolism with Positron Emission Tomography in Normal and Psychiatric Populations

By J.D. Brodie, A.P. Wolf, N. Volkow, D.R. Christman, P. DeFina, M. DeLeon, T. Farkas, S.H. Ferris, J.S. Fowler, F. Gomez-Mont, J. Jaeger, J.A.G. Russell, R. Stamm, and Y. Yonekura

Introduction

Psychiatry deals with abnormal behavior, but normality has been very difficult to define, given the wide range of variations in normal populations caused by the interaction of biological, cultural, and social factors (Von Bertalanffy 1968). Over the past 25 years, there has been increasing interest in the biological concomitants of mental illness. The primary focus of most studies has been on pathology, although a biochemical framework for nonpathological mental functioning has yet to be established in humans. Several years ago, we initiated a study of regional brain metabolism in a group of rigorously defined normal subjects, designed to provide a reference for other human investigations in the Brookhaven National Laboratory – New York University program. Thus far we have studied ^{18}F -2-deoxy-2-fluoro-D-glucose (^{18}FDG) metabolism in about 150 subjects, including normals and patients with schizophrenia, senile dementia, or primary affective disorders. This chapter presents some of the data analyzed to date.

Material and Methods

The normal population consisted of 20 males aged 19–59 years. Inclusion in the study was determined by the absence of medical, neurological, and psychological pathology. Medical reasons for exclusion were a history of severe head trauma, chronic hypertension, significant vascular disorders, diabetes mellitus, thyroid abnormalities, and a history of psychiatric illness.

Gross psychopathology was identified with the Structured Clinical Interview (SCI), an inventory of 179 yes-or-no items filled out by the examiner during a 20- to 30-min interview. The SCI can also be scored for 13 overlapping

scales: anger, hostility, conceptual dysfunction, fear, worry, incongruous behavior, incongruous ideation, lethargy, dejection, perceptual dysfunction, physical complaints, self depreciation, and sexual problems. Any score significantly beyond the norms on any SCI component automatically excluded the candidate.

Neurological and neurophysiological screening included medical history and testing for intelligence (WAIS), anterograde memory [Randt-NYU Memory Test (Randt 1980)], perceptual-motor function and structure (Bender Visual-Motor Gestalt Test), and handedness [Edinburgh Handedness Inventory (Oldfield 1971)]. Subjects also underwent a comprehensive laboratory work-up, a brief neurological screening examination, CT (to provide scans which could be correlated with PETT), computerized EEG, and testing of visual and auditory evoked potentials (John et al. 1977).

The schizophrenia group included 13 volunteer white males aged 21–54 years, diagnosed jointly by two psychiatrists on the Research Diagnostic Criteria (RDC) for schizophrenic disorders (Spitzer et al. 1973). Schizophrenic candidates were screened for history or presence of significant medical illness and frank neurological disease, for use of drugs other than prescribed psychotropic medication, for laboratory abnormalities (on SMA-12, urinalysis, and thyroid function), and for drug and alcohol abuse. Psychopathology was determined by the Combined Instrument Schedule (CIS) which is a combination of three semistructured interviews: the PSE (Wing et al. 1974), the SADS (Endicott and Spitzer 1978), and CMS (Cooper et al. 1969); by a diverse set of perceptual experiments designed to measure a broad spectrum of attention factors; and by a battery of detailed neuropsychological tests for specific patterns of cognitive, linguistic, and behavioral dysfunction.

In the group of nine patients with affective disorders, five had a diagnosis of unipolar depres-

sion, based on RDC, and were clinically depressed at the time of the run. The other four had a diagnosis of manic-depressive psychosis and were clinically manic when the scans were performed.

The senile dementia group consisted of 15 patients, mean age 73. Diagnosis was based on psychiatric, psychometric (Ferris et al. 1980), and CT evaluation. A group of 14 elderly subjects, mean age 68, who failed to show pathology by the same diagnostic criteria served as the control group.

All subjects underwent a series of contiguous noncontrast CT scans, taken on a GE 8800 instrument at 10-mm intervals at an angle 0° from the canthomeatal (CM) reference line, to correspond to the angle of the positron emission scans. All had visually normal scans, which were a requirement for the normal candidates only.

All PETT scans were performed on the PETT III apparatus at Brookhaven National Laboratory, using ¹⁸FDG as tracer. Patients fasted for 2 h before the procedure. The metabolic images were derived using the extension to humans (Reivich et al. 1979) of the Sokoloff method for deoxyglucose uptake (Sokoloff et al. 1977). Samples for counting and glucose determination were obtained either by arterialization of venous blood through hand-warming to 44° C or by direct radial artery cannulation. A venous catheter in the contralateral arm permitted injection of tracer without disturbing the patient. For the 30-min uptake period, patients were not touched unless necessary and silence was maintained except for the sounds of the computer and the air conditioning fan.

Scanning was started 30 min after injection of tracer, with the time per scan varying from 8 to 14 min. Scans were analyzed at the levels of the "basal ganglia" (BG; CM + 40–50 mm) and the centrum semiovale (CS; CM + 60–70 mm). A histogram was constructed by dividing each metabolic image into 20 levels of glucose metabolic activity in 0.4/100 g per minute increments. The distribution of the pixels within this histogram was then calculated for the different brain slices of the normal subjects. The range of metabolic activity with the highest number of pixels within a slice was obtained to determine the predominant level of activity in the group.

To obtain a preliminary estimate of the effect of hand-warming (for blood arterialization), the location of the regions with highest metabolic activity was determined at the BG level of 15 normal subjects and 20 schizophrenics. The glucose metabolic activity was classified according to the histogram. The highest 0.4-mg range of glucose metabolism was determined for each subject, with individual pixels in this category classified as belonging to the left hemisphere, the right hemisphere, or the midline area where the two hemispheres merge.

The relation between the anterior and the posterior regions of the brain was determined at the BG and CS levels for the first 11 normal and first 13 schizophrenic subjects. In the BG scan, the frontal lobe was defined posteriorly by a line bisecting the head of the caudate nucleus perpendicular to the interhemispheric fissure. The region of interest (ROI) coursed anteriorly to avoid the basal ganglia and the anterior horns of the lateral ventricles by approximately 1–2 mm on CT scan (about 1 PETT pixel or 0.43 mm in real brain). On the CS scan, the posterior border of the frontal lobe was defined by the central sulcus and bypassed the ventricle anteriorly. A nonfrontal region, used for reference purposes in the data analysis, was defined for both levels as the entire area within the skull not included in the frontal region.

For the analysis of patients with senile dementia and their elderly controls, regions of interest in the BG scan included frontal cortex, temporal cortex, and caudate nucleus. Correlations between glucose metabolism and cognitive impairment were determined using standardized tests of cortical functioning (Ferris et al. 1980).

Results

All normal subjects showed a unimodal pattern in distribution of pixels on the histogram. The metabolic range with the maximal number of pixels varied at different brain levels in relation to the amount of gray matter, white matter, and cerebrospinal fluid. Three subjects accounted for most of the dispersion. One had a pixel distribution at lower metabolic levels and two were skewed to higher metabolic levels. These deviations from the group pattern appear

throughout most of the brain slices. A test-retest design would reveal whether the differences are state-dependent or individually characteristic. Given the small number of subjects with deviant patterns, it was not possible to relate the finding to any of the other parameters measured (neurometrics, neuropsychological tests, age).

In 13 of the 20 normal subjects, a left-right asymmetry – reflected as higher glucose metabolism on the right than on the left side – was detected at the cortical area between the frontal part of the midline gray matter and the cortical area overlying the lateral aspect of the basal ganglia. In 15 of the 20 normal subjects, left-right asymmetry was noted at the temporal cortex of the BG level.

The significance of this finding is currently unclear (Mazziotta et al. 1981). It could reflect the effects on the brain of the arterialization procedure, which uses a hand-warmer on the subject's left hand. Table 1 shows the location of the pixels with the highest metabolic activity for both normal and schizophrenic subjects who underwent a hand-warmer procedure and those who did not. Although the number of subjects are small, the higher metabolic activity does seem to occur in the right rather than the left hemisphere.

It has been reported that monaural, nonverbal auditory stimulation, regardless of the side stimulated, results in a diffuse frontotemporal asymmetry as well as bilateral parietotemporal activation (Metter et al. 1981). Thus the mechanical noise of the PETT III could account for the asymmetry. Part of this excess right-sided activity may be due to the metabolic reflection of anxiety as reported by Reivich et al. (this volume). Finally, postmortem brain studies have shown the right frontal hemisphere to be wider than the left (Oldfield 1971).

The global metabolic rate at the BG level is summarized in Table 2 for normal, schizophrenic, depressed, and manic subjects. Preliminary results suggest a greater metabolic heterogeneity in the psychiatric population than in normal controls. This may reflect problems in nosology, severity of illness, state variance, or other factors yet unrecognized. Certainly, these findings warrant cautious examination before pathognomonic features can be ascribed to PETT data. Comparison of the

Table 1. Percentage distribution of highest activity pixels

	<i>n</i>	Left hemisphere	Mid-line	Right hemisphere
Normal				
No hand-warmer	4	36	6	58
Hand-warmer	11	5	23	72
Schizophrenia				
No hand-warmer	5	23	35	42
Hand-warmer	15	17	20	63

Table 2. CMRG (mg/100 g per minute) in the BG plane

Group	<i>n</i>	Mean	SD
Controls	11	3.57	0.50
Schizophrenia	13	3.27	0.86
Depression	5	3.62	1.37
Mania	4	3.98	0.99

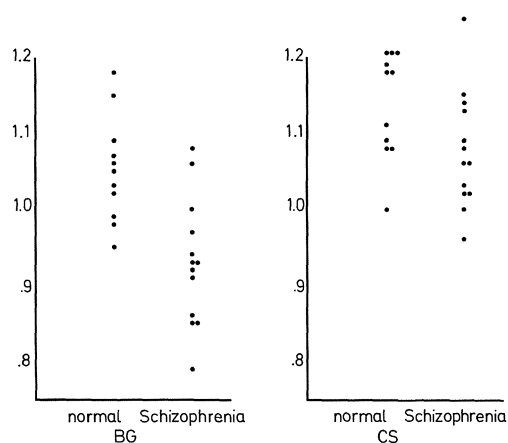


Fig. 1. Frontal to nonfrontal metabolic ratio

frontal to nonfrontal ratio in 11 normal subjects and 13 schizophrenics at the BG and CS levels suggests it is lower in the schizophrenics (Fig. 1). Using covariance analysis with the "nonfrontal slice" as the covariate, this relative difference in frontal activity was significant at the $p < 0.01$ and $p < 0.05$ levels for the BG and CS planes respectively. Although approximately half the patients were on psychotropic medication at the time of the run, they showed no metabolic patterns significantly different from

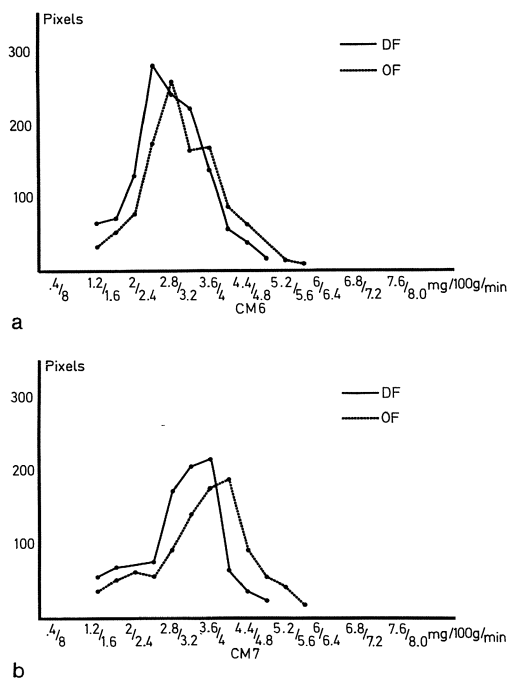


Fig. 2a, b. Pixel distribution in monozygotic twins. **a** high BG level; **b** CS level

those of nonmedicated patients with regard to the whole plane of section or the frontal to non-frontal ratio. There was no significant age difference between the two groups. Regional cerebral metabolism of glucose (RCMRG) was diminished at the BG levels in patients with dementia in all ROIs examined when compared with controls. Significant correlation was found between the quantitative decline in glucose metabolism and the degree of cognitive impairment (Ferris et al. 1981). This change is consistent with the impairment of oxygen metabolism in a similar population (Frackowiak et al. 1981).

A pair of monozygotic twins in the normal group proved to have identical values of neurometric and psychological evaluation. On brain scans, performed 3 weeks apart, global metabolic rates at the BG and CS levels respectively were 3.96 mg/100 g per minute and 3.76 mg/100 g per minute for one twin and 4.78 mg/100 g per minute and 4.67 mg/100 g per minute for the other. Although values were different, distribution of grades of metabolic activity at high BG (CM6) and CS (CM7) levels were very similar (Fig. 2a, b).

Discussion

The metabolic distribution was similar for most of the normal subjects. If this pattern proves to be consistent, it may be a useful parameter for comparison of psychiatric illnesses. Also noteworthy is the finding that similarities in glucose metabolism between the identical twins appeared at the level of their activity distribution rather than on the absolute values. Considering the similarity in all other variables examined, the difference in absolute activity probably represents state dependence but may be attributed to test-retest error.

The unimodal distribution of metabolic activity seen in the normal subjects may be due to an averaging effect secondary to the low resolution of PETT III, or may reflect metabolic heterogeneity in white and gray matter as a whole (John et al. 1977). Comparison of metabolic activity in younger (Kendell 1979) and older (Ingvar and Schwartz 1974) normal populations revealed no significant differences in global absolute values or metabolic activity distribution.

On comparison of the cerebral metabolic activity in normal and psychiatric populations, we observed differences in both absolute values and patterns of activity. The diminution in the frontal-nonfrontal ratio in the schizophrenics seems to be fairly consistent. Future analysis on the PETT VI instrument should help determine whether this hypofrontality is due to diminution of glucose uptake in the gray matter or white matter or is a part of a more widely distributed alteration in the pattern of glucose utilization. Although these findings may reflect a trait in schizophrenics, a provocative test-retest situation will probably be a more sensitive index of regional cerebral metabolic functioning in this illness. On the other hand, in our studies of patients with Alzheimer's disease a high correlation was found between degree of cognitive impairment and regional and subregional CMRG (Ferris et al. 1981).

Because of the small number of subjects with primary affective disorders and their metabolic heterogeneity, no firm conclusions can be drawn at this time. In designing future PETT studies of the affective disorders, however, investigators should keep this heterogeneity in

mind and obtain a reliable quantitative description of psychiatric symptom presentation.

Any attempt to find an anatomical focus for behavior or a pattern of behaviors that presents as a psychiatric syndrome raises a number of methodological and analytical problems. For example, the 30-min glimpse of CMRG afforded by a PETT study may not be representative. Most psychiatric patients, unless they have irreversible impairment, would not be expected always to be pathological, even in the midst of florid disease. It is difficult to verify what is going on in the patient's brain during the study, or the level of anxiety and its effects on the metabolic rate. Moreover, the effects of diet and fasting on stress and on RCMRG itself have not been studied and are difficult to evaluate in the present context. We must depend on verbalization of behavior. A patient may be able to tell the investigator he heard voices while in the machine, but were these voices reflected in his mental activity, did they cause great anxiety, or were they reflected at various times in the uptake of FDG or in metabolic rate changes of different degrees? If we were averaging over a 30-min uptake and the patient had 3 min of intense mental activity early in the procedure, it might produce a different metabolic pattern than would be observed if it occurred later.

A number of other questions wait to be answered. To what extent are these images affected by utilization of alternative energy sources, for example? The amounts of aerobic and anaerobic metabolism have never been assessed. There might even be regional metabolic preferences for energy sources other than glucose. Another tracer might reflect patterns of activity substantially at variance with those determined by the glucose method.

Furthermore, when attempting neuroanatomical localization of metabolic activity we will have to take into account that a defined anatomical area is not necessarily functionally homogeneous and that a given function may not be localized.

Perhaps the major problem in applying PETT technology to human behavior lies in the fact that we are dealing with a greater variance than might be reflected in any single biochemical alteration. We are looking for anatomic loci for feeling states or logical thoughts, factors that are not at issue with lower animals. Our search

may be chimerical; it may be that the *relationships* of various mental processes, perhaps reflected by metabolic activity, will turn out to be far more pertinent than the intrinsic metabolic activity in any given area. This is related to the unresolved issue of the localized vs the distributed fashion in which the human nervous system processes information (Luria 1980) and it also addresses the distinction between functional differences (uncorrelated areas) and anatomical differences.

Acknowledgments. This research was supported in part by the Department of Energy, Office of Health and Environmental Research and Public Health Service Grant NS-15638. The authors thank Dr. A.B. Brill, Dr. B. Jones, Ms. A.A. Farrell, and Ms. K. Karlstrom for their assistance.

References

1. Cooper JE, Kendell RE, Gurland BJ et al. (1969) Combined mental state. Cross-national study of diagnosis of the mental disorders: some results from the first comparative investigation. *Am J Psych [Suppl]* 125: 1-46
2. Endicott J, Spitzer R (1978) A diagnostic interview: the schedule for affective disorders and schizophrenia. *J Gen Psychiat* 35: 837
3. Ferris SH, DeLeon MJ, Wolf A et al. (1980) Positron emission tomography in the study of aging and dementia. *Neurobiol Aging* 1: 127-131
4. Ferris SH, DeLeon MJ, Christman D et al. (1981) Positron emission tomography studies of regional brain metabolism in elderly patients. In: Perris C, Struwe G, Janson B (eds) *Biological psychiatry*. Elsevier/North Holland Biomedical, Amsterdam, pp 280-283
5. Frackowiak RSJ, Pozzilli C, Legg NJ et al. (1981) Regional cerebral oxygen supply and utilization in dementia. *Brain* 104: 753-778
6. Ingvar DH, Schwartz MS (1974) Blood flow patterns induced in the dominant hemisphere by speech and reading. *Brain* 96: 274-288
7. John ER, Karmel BZ, Corning WC et al. (1977) *Neurometrics*. Science 196: 1393-1410
8. Kendell RE (1979) The classification of depressions: a review of contemporary confusion. *Br J Psych* 129: 15-28
9. Luria AR (1980) *Higher cortical functions in man*, 2nd ed. Basic, New York
10. Mazziotta JC, Phelps ME, Miller J et al. (1981) Tomographic mapping of human cerebral metabolism: normal unstimulated state. *Neurology* 31: 503-516

11. Metter EJ, Wasterlain CG, Kuhl DE et al. (1981) FDG positron emission computed tomography in a study of aphasia. *Ann Neurol* 10: 173–183
12. Oldfield RC (1971) The assessment and analysis of handedness. The Edinburgh inventory. *Neuropsychologia* 9: 97–113
13. Randt CT (1980) A memory test for longitudinal measurement of mild to moderate deficits. *Clin Neuropsychol* 2: 184
14. Reivich M, Kuhl DE, Wolf A et al. (1979) The ^{18}F -fluorodeoxyglucose method for the measurement of local cerebral glucose utilization in man. *Circ Res* 44: 127–137
15. Sokoloff L, Reivich M, Kennedy C et al. (1977) The ^{14}C -deoxyglucose utilization: theory, procedure and normal values in conscious and anesthetized albino rat. *J Neurochem* 28: 897–916
16. Spitzer RL, Endicott J, Robins E (1973) Research diagnostic criteria. New York State Department of Mental Hygiene, New York
17. Von Bertalanffy L (1968) General systems theory. Braziller, New York
18. Wing JK, Cooper JE, Sartorius N (1974) The description and classification of psychiatric symptoms: an instructional manual for the P.S.E. and CATEGO system. Cambridge University Press, London

Protein Synthesis and Receptor Studies

(Chairman: L. E. Feinendegen)

Local Brain Protein Metabolism in Dementia and Schizophrenia: In Vivo Studies with ^{11}C -L-Methionine and Positron Emission Tomography

By P. Bustany, J. F. Henry, T. Sargent, E. Zarifian, E. Cabanis, P. Collard, and D. Comar

Introduction

Positron emission tomography (PET) now provides the opportunity for local studies of brain functions such as blood flow and oxygen and glucose, and aminoacid consumption. A reduction in mean value of one of these biochemical parameters is not always followed by the same decrease in the others, and these discrepancies are particularly interesting in diseased nervous tissue.

We have developed a dynamic three-compartment model of protein synthesis in brain (Fig. 1) using P. E. T. with ^{11}C -L-methionine as a tracer for human studies. This model was carefully checked by varied biochemical and physiological procedures in rats and baboons and subjected to autoradiographic examination in animals (Bustany, to be published). We report here its application by PET to 48 patients with or without Alzheimer's dementia and more recently to four young schizophrenics who had never been treated.

Principle and Methods

Briefly, the patient's head is positioned under an ECAT II (ORTEC) camera, with section 5 cm above the orbitomeatal (OM) line. Spatial resolution is 19 mm axially and 16 mm transversely. Special care is given to quantification through daily activity standardization of the camera, transmission data for picture reconstruction, hardware correction of random coincidences, etc.

Pure ^{11}C -L-methionine (600 Ci/mM) (Comar et al. 1976) was rapidly injected intravenously into the patient's arm (20 mCi in 2–3 ml) and blood activity during scanning was sampled in arterialized venous blood. Corrections were introduced for the difference between cerebral

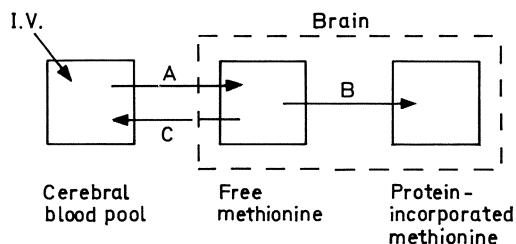


Fig. 1. Three-compartment model of methionine metabolism in brain following intravenous bolus injection

and peripheral hematocrit (0.85) as for red cell vs plasma activity (0.66 in volume).

Twelve scans are recorded (6×1 min, 4×5 min, 2×10 min) with a least 800 000 events per image. The overall duration of metabolic examination is 47 min, during which time the patient is rested in warm and quiet conditions.

For a precise calculation of the most important parameters of protein metabolism in small regions of brain, a correction for residual blood activity in the slice is needed. This is obtained the next day with a 10-min image at the same level using $^{68}\text{GaCl}_3$ -labeled transferrin as a plasmatic marker (Maziere et al. 1982). Blood sampling allows direct calibration of the picture in milliliters of blood per pixel or per milliliter of brain. An iterative program of modelization using the cerebral blood activity curve and corrected brain activity curve yields the three transport constants of the model and so calculates activity in the two brain compartments: free methionine in extracellular space and incorporated into proteins (Fig. 2).

The final images for computer calculation were a series of 12 scans corrected for cerebral blood activity, which was high in early images and still 15%, for example, 5 min after intravenous injection of ^{11}C -L-methionine. For each pixel of a transformed 50×50 image, a 12-point curve is extracted and fitted. Local parameters are

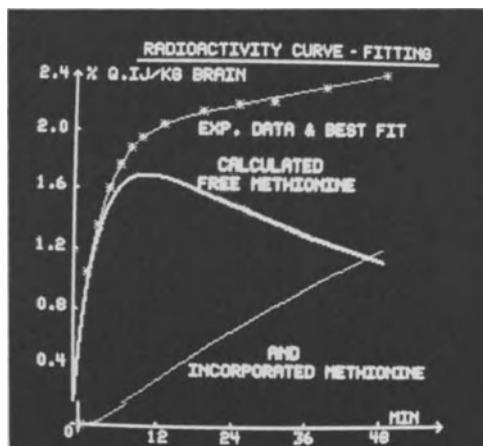


Fig. 2. Computer fitting of experimental data

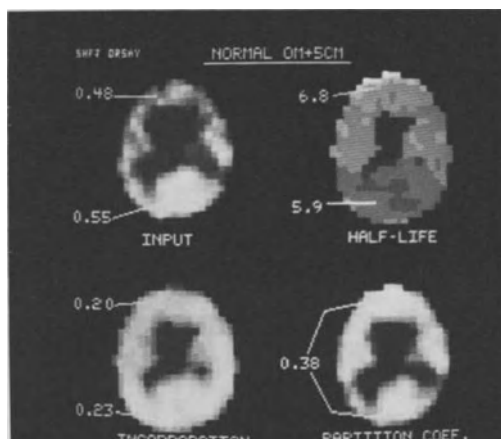


Fig. 3. Metabolic maps at OM + 5 cm level in a normal patient. Input and incorporation are expressed in nM/min per gram brain (see Tables 1-3), and half-life in minutes. The partition coefficient of methionine between brain spaces and blood remains very constant in each case, reflecting the well state of the blood-brain barrier. (The whole color scale is used for each image and is not employed to compare absolute values in the three cases)

given numerically and as real metabolic maps of the slice: methionine input and partition coefficient at the blood-brain barrier level, half-life of free methionine, and protein incorporation rate are the main clinically demonstrative parameters (see Figs. 3-5).

The mean age of our subjects was 86.3 ± 6.1 years for the demented patients ($n=28$), 80.2 ± 9.4 years for the normal group ($n=20$),

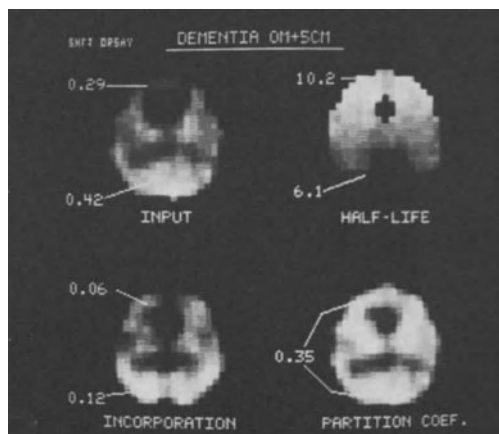


Fig. 4. Metabolic maps at OM + 5 cm level in a patient with Alzheimer's dementia (for units see Fig. 3)

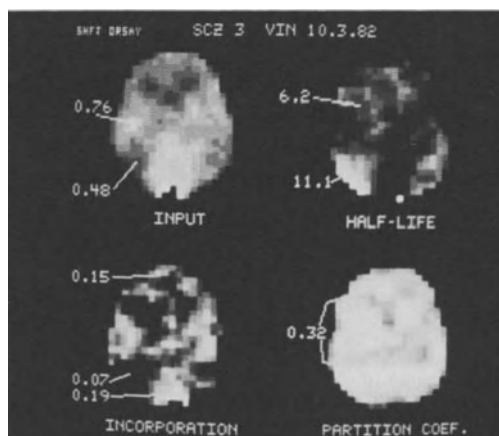


Fig. 5. Metabolic maps at OM + 5 cm level in a schizophrenic subject (for units see Fig. 3)

and 25.2 ± 3.2 years for the schizophrenic patients ($n=4$). All were right-handed.

Clinical, biochemical, and psychometric (WAIS and Rey's test) examinations were made of each patient. Demented patients had not been treated for 3 weeks and the newly detected schizophrenic patients had never had treatment. A CT scan completed the examinations and gave valuable data about the local anatomy of the studied slice. Particular efforts were made to avoid any form of vascular dementia (clinical history, Hachinski's test, CT scan, etc.). Lacking some results at the present time, we have classified demented subjects roughly in two groups according to clinical and

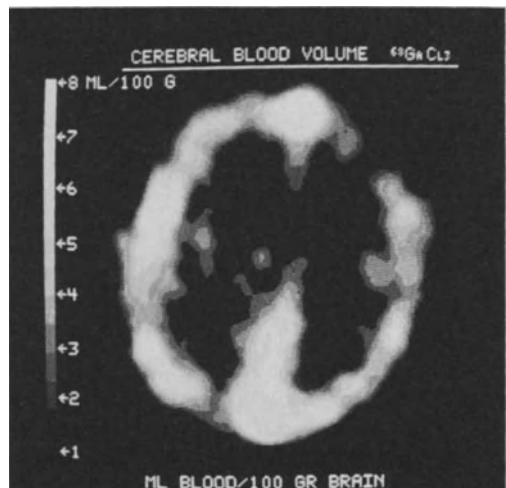


Fig. 6. Cerebral blood volume map with ⁶⁸GaCl₃

psychometric test results. Three of our very demented patients were discarded because they showed brain abnormalities on CT scan compared to the cross-matched normal reference group.

Results and Comments

Figure 6 shows a ⁶⁸Ga image of cerebral blood volume in dementia. Like others (Grubb et al. 1977), we have not found any local or global modification of this volume in demented patients, even in those in a more altered state. Modifications were also absent in young schizophrenic people. On the other hand, important modifications of local protein synthesis were found in these cases.

Besides the absolute values of brain physiological parameters, we calculated an occipital to frontal ratio using large bilateral symmetrical zones on metabolic maps, according to the anatomy of the brain slice studied.

Alzheimer's Dementia

Metabolic maps given in Fig. 4 are compared to normal values in Fig. 3. The results are summarized in Tables 1 and 2. Regarding the clinical status, a net diminution in frontal metabolism can be seen in Alzhei-

Table 1. Whole slice methionine metabolism in dementia

	Normal (n=20)	Dementia (n=25)
Age (years)	80.2 ± 9.4	86.3 ± 6.1
Input (nM/min per gram brain)	0.46 ± 0.16	0.31 ± 0.12
Half-life (min)	8.1 ± 1.9	9.0 ± 2.7
Incorporation (nM/min per gram brain)	0.15 ± 0.05	0.09 ± 0.04
Extraction (%)	24.7 ± 9.4	14.9 ± 5.7

mer's dementia. This is particularly plain on calculation of the fronto-occipital ratio in the studied brain slice. Parietal cortex is also often altered. The reason for a relative apparent stability of occipital tissue in the disease is not known, but the stability was also observed with FDG studies (Frackowiak et al. 1981). A decrease of more than 65% in the protein synthesis rate has been measured in the most severely demented patients.

Schizophrenia

Results shown in Table 3 concerning the whole slice cannot yet be compared to those from a normal age-matched group, but the values are plausible since they come between the lower values of our older reference group and our well-treated phenylketonuric children (Bustany et al. 1981). For example, protein synthesis rates (in nM/min per gram brain) are $0.35 \pm 0.09 < 0.23 \pm 0.07 < 0.15 \pm 0.05$. This must certainly be assigned to the normal aging process of the brain, as was reported in animals. The observed asymmetry in fronto-occipital ratio remains to be confirmed on a greater scale than that of our first patients; antecedent general medical treatment seems to be able to alter it substantially. In one of our schizophrenic subjects bilateral focal parietal lesions appeared at the OM+ 5 cm level (see metabolic maps, Fig. 5), and more thorough medical tests are under way.

Table 2. Local protein synthesis rate: fronto-occipital ratio

Region of interest	Normal (<i>n</i> = 20)	Dementia (<i>n</i> = 25)	Region of interest	Light dementia (<i>n</i> = 8)	Severe dementia (<i>n</i> = 17)
Frontal	0.15 ± 0.06	0.09 ± 0.03	Frontal	0.13 ± 0.03	0.05 ± 0.02
Occipital	0.16 ± 0.06	0.15 ± 0.07	Occipital	0.15 ± 0.05	0.10 ± 0.04
Fronto-occipital ratio	0.96 ± 0.11	0.64 ± 0.21	Fronto-occipital ratio	0.99 ± 0.18	0.52 ± 0.13

Frontal and occipital values expressed in nM of methionine/min per gram brain

Table 3. Results in schizophrenia

Global		Regions of interest	
Input (nM/min per gram brain)	0.83 ± 0.14	Frontal (nM/min per gram brain)	0.21 ± 0.07
Half-life (min)	5.7 ± 0.5	Occipital (nM/min per gram brain)	0.27 ± 0.07
Incorporation (nM/min per gram brain)	0.23 ± 0.07	Fronto-occipital ratio	0.80 ± 0.06
Extraction (%)	16.9 ± 3.6		

Conclusion

Protein metabolism is very stable in the brain and not correlated with any acute stimulation of neurons. Comparison with CT scans shows that a metabolic impairment precedes by some time all signs of death and disappearance of cerebral tissue. Dramatic variations can be found in protein metabolism even in slight dementia, and it is likely that as reported for other kinds of cells, a decrease in protein metabolism is a normal reaction of the nervous cells to biochemical injury. Discrepancy between glucose and amino acid metabolism in focal lesions such as epileptic foci or cortical injuries in animals (Willmore et al. 1981; Wasterlain and Dwyer 1982) confirms the interest and the uniqueness of brain protein synthesis in itself. Much remains to be learned about protein synthesis in the brain, but ¹¹C-L-methionine examination of the human brain with PET can improve our understanding of mental illness.

References

1. Bustany P, Sargent T, Saudubray JM et al. (1981) Regional human brain uptake and protein incorporation of ¹¹C-L-methionine studied in vivo with PET. J Cereb Blood Flow Metabol 1 [Suppl 1]: S 17-S 18

2. Comar D, Carton JC, Mazière M et al. (1976) Labelling and metabolism of methionine - Methyl ¹¹C. Eur J Nucl Med 1: 11-14

3. Frackowiak RSJ, Pozzilli C, Legg NJ et al. (1981) Regional cerebral oxygen supply and utilization in dementia. Brain 104: 753-778

4. Grubb RL, Raichle ME, Gado MH et al. (1977) Cerebral blood flow, oxygen utilization, and blood volume in dementia. Neurology 27: 905-910

5. Mazière B, Loc'h C, Ricordel Y et al. (1982) Regional cerebral blood pool measurements using ⁶⁸Ga labeled proteins. WFNMB Meeting Abstracts, Paris, September 1982, Vol 1. Pergamon Press, pp 666-669

6. Wasterlain CG, Dwyer B (1982) Generalized neonatal seizures in primates: focal inhibition of protein synthesis in vulnerable regions. Neurology: A 169 (abstract)

7. Willmore LJ, Dunn AJ, Rubin JJ (1981) Changes in (³H) lysine incorporation into protein in a seizure focus in rat isocortex. Brain Res Bull 6: 463-466

Brain Regional Pharmacokinetics of ^{11}C -Labeled Diphenylhydantoin and Pimozide in Man

By J. C. Baron, D. Comar, C. Crouzel, D. Roeda, G. Mestelan, E. Zarifian, C. Munari, C. Stoffels, J. Bancaud, J. P. Chodkiewicz, H. Loo, and Y. Agid

Positron emission tomography (PET) offers the new opportunity to study, *in vivo* and non-invasively, the brain regional pharmacokinetics of labeled drugs in man.

Two different approaches can be used. The first is to correlate the clinical condition with the regional concentration of the labeled drug. One possible application is partial epilepsy, where the therapeutic efficacy of anticonvulsants is well correlated to their plasma levels, except in intractable cases, in whom low brain concentration of drugs in spite of adequate plasma levels has long been suspected.

The second and entirely different approach is to use the pharmacologic properties of some drugs to bind specifically to a class of physiologic receptors. The labeled ligand is then used as a probe to study *in vivo* the regional concentration and affinity of specific receptors in both normal and pathologic situations for which there is no satisfactory animal model. Potential applications are numerous; the pathophysiologic study of schizophrenia (i.e., the dopaminergic hypothesis), depression, anxiety, degenerative disorders, etc.

This paper presents our results from both approaches, the first using ^{11}C -diphenylhydantoin has a labeled anticonvulsant in the study of intractable partial epilepsy, the second using ^{11}C -pimozide as a labeled neuroleptic in the study of dopaminergic receptors.

^{11}C -Diphenylhydantoin

We chose 5-5-diphenylhydantoin (DPH) because it is one of the most widely applied anticonvulsant drugs and is well-characterized in terms of plasma kinetics and therapeutic steady-state concentrations, brain penetration and binding, mechanisms of antiepileptic activity, and metabolism (Alvin and Bush 1977). However, the brain pharmacokinetics in rela-

tion to plasma drug levels are less wellknown, and the few human studies that have been reported used brain samples obtained during neurosurgery. Most data on the distribution and kinetics of DPH in brain originate from animal studies, including two ^{11}C -DPH animal studies (Stavchansky et al. 1978; Winstead et al. 1976).

Material and Methods

Patients

Ten patients (age 18–30 years) were studied. Eight of them suffered from medically resistant focal epilepsy and were studied while hospitalized for investigations designed to evaluate the indications for neurosurgical cortectomy. All eight had various anticonvulsants in their daily oral medication (including three patients taking DPH). The epileptogenic area, as assessed by stereoencephalography (performed after the PET study), was of variable topography, and the presumed pathologic process was definite in only five patients. The remaining two patients had no history of epilepsy and served as controls.

^{11}C -DPH Study

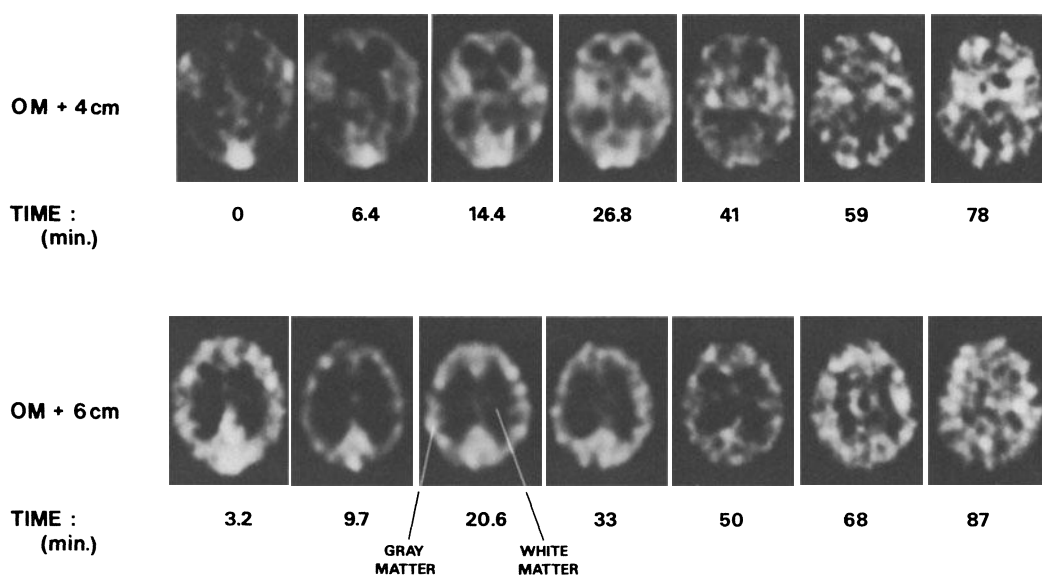
The ^{11}C -DPH was synthesized from ^{11}C -phosgene (Roeda and Westera 1981); it was pyrogen-free, sterile, and radiochemically pure at injection time. Two levels, parallel to the orbitomeatal (OM) line and chosen individually according to the presumed location of the epileptogenic area, were studied. Slow i.v. injection of ^{11}C -DPH (2–26 mCi, specific activity 137–287 mCi/ μmol) in saline was performed after the patient had closed his eyes and the environment had been made as noiseless as possible. While the images were collected, venous blood samples were withdrawn. Blood count-

ing was performed on a scintillation well counter properly calibrated to yield ^{11}C concentrations in $\mu\text{Ci}/\text{ml}$.

The coincidence photons were collected by an ECAT II (ORTEC) positron tomograph (resolution $\sim 16\text{ mm}$ in the lateral plane and 19 mm in the axial plane). Routine ensuring of accurate quantitation entailed (1) automatic subtraction of random coincidences, (2) measured attenuation correction mode, (3) normalization of detector sensitivity, and (4) accurate cross-calibration between the ECAT system and the well counter.

Image acquisition started immediately after ^{11}C -DPH injection, and consisted of repeated alternate scanning of the two study levels. Usually, the same levels were scanned 7 times at progressively increasing acquisition times; the whole study lasted from 40 to 90 min depending on the total radioactivity injected. The

total number of true coincidences was typically $\sim 4 \times 10^5$ in the first image and $\sim 8 \times 10^5$ in the third image, falling progressively to $\sim 2 \times 10^5$ in the last image. On each of the two brain images in every patient, circular (4-cm^2) regions of interest (ROIs) were sited in anatomically meaningful areas distributed as homogeneously as possible over the asymptomatic hemisphere. An automatic software program was used to provide the time course of the ^{11}C ROI concentration across the seven scans (midscan time values), expressed either in $\mu\text{Ci}/\text{ml}$ or as the percentage of the injected ^{11}C -DPH dose per milliliter brain tissue (% ID). From these curves, and from the blood time-activity curve plotted in an identical manner, brain to blood ratios were calculated at various time intervals.



^{11}C -PHENYTOIN NORMAL BRAIN

(S.H.F.J., ORSAY)

Fig. 1. Sequential positron tomographic images representing the distribution of ^{11}C -DPH at two contiguous brain levels parallel to the OM line. Each brain level has been scanned seven times (alternately) after i.v. injection of ^{11}C -DPH (figures shown are midscan times). At early scan times (up to $\sim 10\text{ min}$), high activity is present in blood relative to brain, as indicated by the active spots in the posterior part of the sagittal

sinus and in both sylvian fissures. Later ^{11}C -DPH accumulates in brain, and gray matter structures are well demarcated from white matter by their higher activity (10–30 min). Later still, white matter activity increases to almost the level of gray matter, resulting in nearly uniform images (while noise increases due to ^{11}C physical decay)

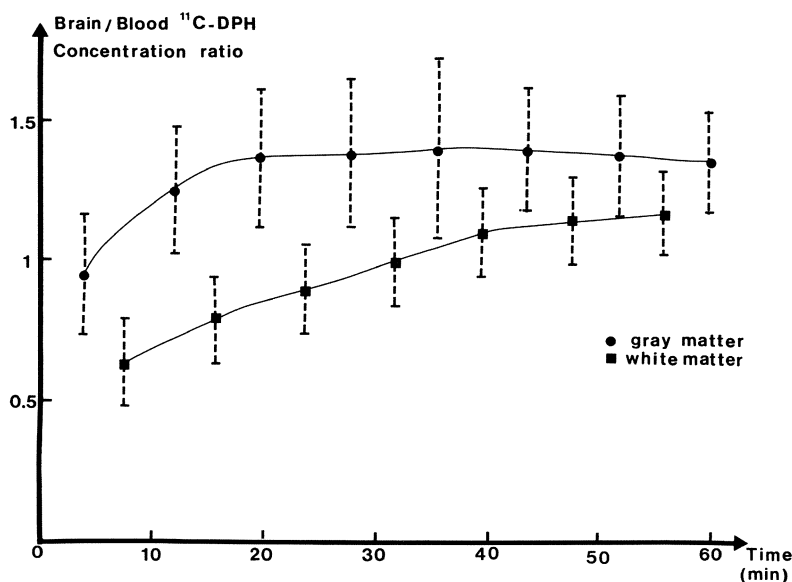


Fig. 2. Mean brain to blood ¹¹C-DPH concentration ratio with standard deviations plotted against time in ten studies

Results

Blood activity time course

The mean % ID per milliliter whole blood showed an immediate peak at 2.6×10^{-3} , followed by a rapid decline to 1.7×10^{-3} at 10 min and a slow washout down to 1.5×10^{-3} at 60 min.

Brain activity time course

Typical sequential images obtained at two adjacent brain levels in a control patient are displayed in Fig. 1. Gray matter structures are well demarcated from white matter structures at early scan times, but thereafter white matter activity approaches that of gray matter, resulting in less gray-white imaging contrast.

The mean % ID per milliliter brain in two typical structures, such as the visual cortex and the corona radiata, confirmed this differing behavior of ¹¹C-DPH in gray and white matter respectively. In gray matter the activity rose during the first 20 min to reach a mean of 2.2×10^{-3} and then slowly declined to 2.1×10^{-3} at 60 min, whereas in white matter it rose continuously, being 1.4×10^{-3} at 20 min and 1.8×10^{-3} at 60 min.

The time course of the brain to blood ratios showed the same features (Fig. 2). Their values

at 50 min ranged from 1.05 to 1.87 for gray matter (mean 1.37 ± 0.23) and from 0.85 to 1.36 in white matter (mean 1.06 ± 0.16); brain to plasma ratios were ~27% lower (four determinations). The highest values were in one control, but those of the other control patient were unremarkable. The brain uptake and kinetics of ¹¹C-DPH were not affected by current medication with DPH.

Blood-Brain ¹¹C Activity Correlation

The ¹¹C concentrations in gray and white matter for all 10 patients studied were plotted against that in whole blood, all at 50 min after injection (Fig. 3). The regression lines were: gray = $1.21 \text{ blood} + 0.04$ ($r = 0.925$, $p < 0.001$); white = $0.92 \text{ blood} + 0.04$ ($r = 0.945$, $p < 0.001$).

Comments

A basic assumption underlying the present work is that the ¹¹C radioactivities recorded in blood and cerebral tissue represent ¹¹C-DPH and not other related or unrelated labeled substances. Radiochemical analysis confirmed that the injected compound was over 99% pure ¹¹C-DPH (Roeda and Westera 1981). Previous work with ¹⁴C-DPH demonstrated that, 1 h after i.v. injection, the radioactivity present in

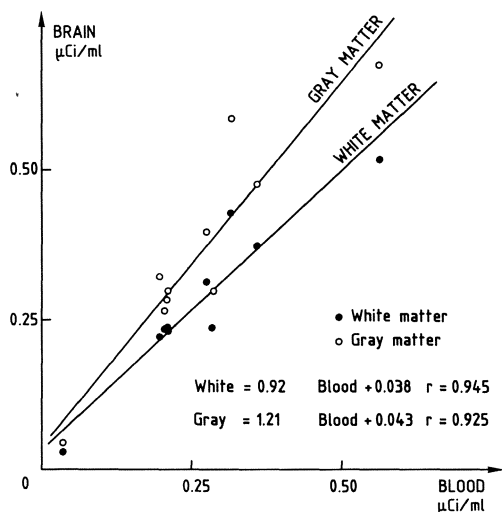


Fig. 3. Gray and white matter ^{11}C radioactive concentrations at 50 min after i. v. injection of ^{11}C -DPH plotted against blood ^{11}C radioactive concentration for all ten patients studied. The calculated regression lines are shown; both correlations are significant at the $p < 0.001$ level

blood and brain was exclusively in the form of DPH (Firemark et al 1963).

In blood, we observed a fast equilibration phase of ^{11}C -DPH, followed by a very slow decline. This is consistent with the blood DPH pharmacokinetics reported in man as well as in animal studies (Stavchansky et al. 1978; Firemark et al. 1963; Wilder et al. 1977; Ramsay et al. 1979; Kutt et al. 1968).

Our study shows that relatively large amounts of ^{11}C -DPH penetrate brain tissue immediately. Because of its high solubility in lipids, DPH is thought to cross the blood-brain barrier readily (Alvin and Bush 1977), although large (>90%) reversible binding to plasma proteins and significant lung retention presumably affect the rate of entry into brain (Alvin and Bush 1977; Winstead et al. 1976). In gray matter, ^{11}C -DPH accumulated during the first 20 min after i. v. injection and then showed a slow decline parallel to that present in blood, indicating that brain-blood equilibrium had essentially been achieved. The behavior observed in white matter was different: ^{11}C -DPH concentration rose slowly over 60 min to approach that present in gray matter, but equilibrium with blood was not reached early enough to be detected within the time frame of our studies. These findings

are consistent with previous animal studies: Firemark et al. (1963) showed that ^{14}C -DPH accumulated in brain after i. v. injection, but that equilibrium was reached much faster in gray than in white matter; activity in the latter equaled that in the former at 1 h after injection. The chief factor responsible for the different rates of accumulation of ^{11}C -DPH in gray and white matter appears to be circulatory, the higher perfusion of gray matter allowing equilibrium to be reached earlier (Firemark et al. 1963). Thus the early ^{11}C -DPH distribution tomographic images (Fig. 2) closely resemble the flow maps obtained in our laboratory using PET and the ^{15}O steady-state technique (Baron et al. 1981). However, additional factors to explain such differences have been suggested, e. g., the "myelin barrier" in white matter (Firemark et al. 1963). The slow accumulation of DPH in brain is thought to be due to nonspecific (reversible) binding to the cell membranes, particularly to their proteidic and phospholipidic components (Goldberg and Crandall 1978).

Most animal and human studies have reported steady-state DPH concentration in white matter equal or superior to that present in gray matter, a difference attributed to a preferential solubility of DPH in the lipidic components of myelin (Alvin and Bush 1977). White to gray ^{14}C -DPH concentration ratios of 1.1 and 1.3 at 1 and 3 h, respectively have been reported in cats (Firemark et al. 1963). In samples of human brain taken at surgery, white to gray DPH ratios ranged from 1.2 to 1.4 (Sherwin et al. 1973; Harvey et al. 1977; Sironi et al. 1980). We could not observe this because ^{11}C -DPH steady state could not be fully achieved in white matter, a consequence of the fast physical decay of ^{11}C . The tendency of white matter ^{11}C -DPH concentration to approach gray matter values was clear, however (Figs. 2, 3). In fact our sequential brain images duplicate in man in vivo the autoradiographic studies by Firemark et al. (1963), who found clear delineation of gray and white structures at 30 min but almost uniform radioactivity at 6 h after i. v. injection.

We found a mean gray matter to blood ^{11}C -DPH equilibrium concentration ratio of 1.37 (range 1.05–1.87), with gray matter to plasma ratios ~27% lower. This is well within the reported ranges obtained by brain sampling in

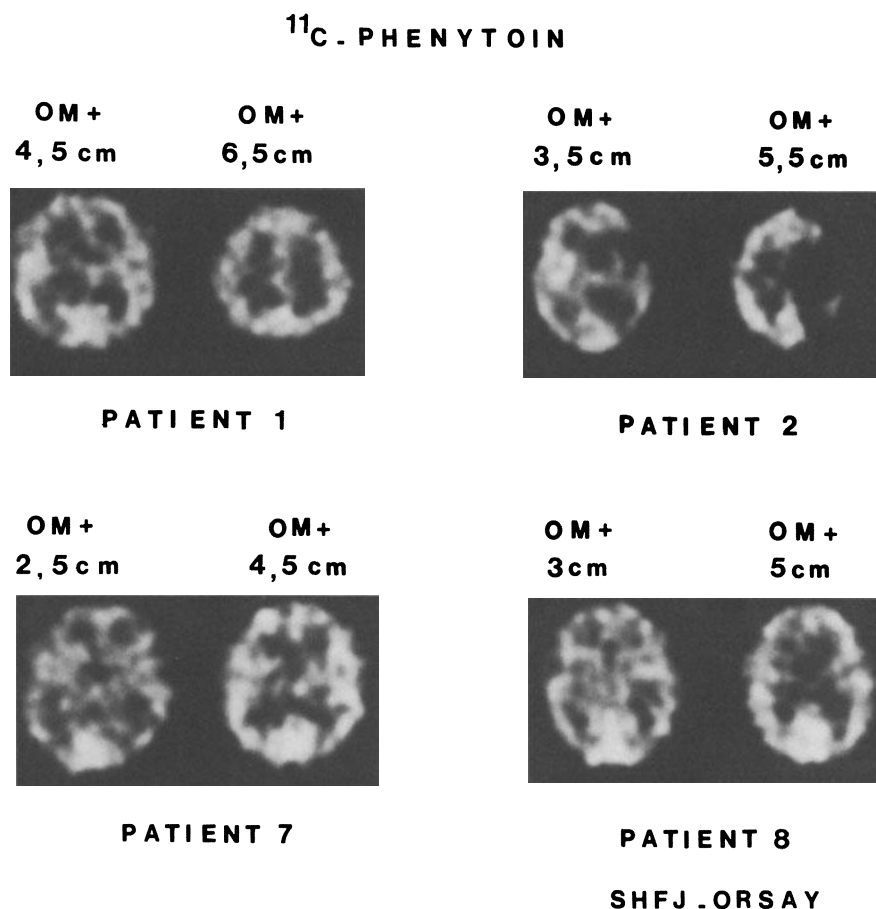


Fig. 4. ^{11}C -DPH brain images obtained from four patients with medically resistant partial epilepsy. Patients 1, 7, and 8 had epileptogenic areas presumably located in the left medial parietal cortex, the right external temporal cortex, and the right central operculum respectively; no ^{11}C -DPH defect is clearly visible

in such areas. Patient 2 had an old porencephalic lesion of the right hemisphere, which is clearly visible as a ^{11}C -DPH defect; assessment of the neighboring epileptogenic area is made difficult by the partial volume effect

neurosurgical patients. In nonepileptic patients, Rapport et al. (1975) reported a gray matter to plasma ratio of 1.74, and Wilder et al. (1977) a brain to plasma ratio of about 1.1. In areas distant from the epileptogenic area per se, brain to plasma ratios of 0.4–1.3 and gray matter to plasma ratios of 1.13–1.44 have been reported (Goldberg and Crandall 1978; Sherwin et al. 1973; Harvey et al. 1977; Sironi et al. 1980; Houghton et al. 1975; Vajda et al. 1974). Our data, in fact, show notably less interindividual variation than did many of the above studies.

The correlation between ^{11}C -DPH concentration in brain and blood at equilibrium was ex-

cellent (Fig. 4). Similar findings have been reported both in rats after i.v. injection of stable DPH (Sherwin et al. 1973) and in neurosurgical patients on chronic DPH medication (Harvey et al. 1977; Sironi et al. 1980; Houghton et al. 1975; Vajda et al. 1974). Our in vivo study therefore confirms that at least in normal brain regions of these epileptic patients, blood concentration of DPH can be regarded as a reliable index of cerebral drug concentration.

We observed the highest brain to blood ^{11}C -DPH ratio in one control patient, but that of the second control patient was unremarkable. Previous surgical assays of stable DPH in "normal" brain regions of intractable epileptics and

of various other diagnostic categories yielded discrepant values: in the latter patient group, Wilder et al. (1977) found a brain to plasma ratio of 1.1, well within the range reported in intractable epileptics, but Rapport et al. (1975) found ratios of 1.14, 1.52, and 2.56 in three controls. Although limited by the small sample, our study suggests that normal brain tissue of intractable epileptics binds DPH as effectively as that of nonepileptics.

The question of whether DPH steady-state concentration is lower *within* the epileptogenic focus is a different matter, and one that has been long debated since the early observations by Rapport et al. (1975) of a fourfold decrease compared to control brain tissue. Recently, Sironi et al. (1980) reported no difference in DPH concentration when tissue showing the maximum intraoperative spiking activity was compared to neighboring brain parenchyma. Another study, however, revealed that the concentration of various anticonvulsants in the epileptogenic area was generally lower than in neighboring samples, but showed large regional differences (Munari et al. 1980). Our data on ^{11}C -DPH within the estimated epileptogenic area, to be reported later, did not disclose any significant difference in ^{11}C -DPH uptake or kinetics when compared to the contralateral homologous brain region (see Fig. 4) (J. C. Baron et al., in preparation). This suggests that, although present at adequate levels within the epileptogenic focus, DPH may still be pharmacologically inefficient in such patients; however, because of the relatively gross spatial resolution of our PET device, highly localized changes may well have been overlooked.

It may be argued that the single i. v. injection of trace amounts of ^{11}C -DPH does not mimic the clinical situation of long-term oral medication with stable DPH. However, the following facts seem to validate the approach used: (1) ^{11}C -labeled DPH does not differ pharmacologically from stable DPH; (2) our equilibrium gray matter to blood ratios are very similar to data reported in long-term DPH-treated patients; (3) the brain uptake and kinetics of ^{11}C -DPH were not affected by the presence of oral DPH in daily medication, consonant with previous evidence that brain binding of DPH is largely nonspecific (Firemark et al. 1963; Goldberg and Crandall 1978). Differences from the long-

term DPH therapeutic regime, such as secondary drug redistribution in the depot tissues and dose-dependent hepatic metabolism of DPH, would presumably affect the brain to blood ratios only slightly. Thus it seems reasonable to assume that acute i. v. injection of trace molecules of ^{11}C -DPH allows clinically relevant conclusions.

^{11}C -Pimozide

The goal using ^{11}C -Pimozide was to investigate whether pharmacokinetic phenomena directly related to specific binding of this labeled neuroleptic to dopaminergic (DA) receptors could be demonstrated in vivo in man.

Pimozide is a dopaminergic antagonist that fulfills most of the necessary conditions for in vivo demonstration of specific binding. It is neither completely trapped in lungs (Crouzel et al. 1980) nor significantly metabolized within 2 h of i. v. injection (Janssen and Allewijn 1968); it crosses the blood-brain barrier well, with a first-pass extraction fraction of about 70% after intracarotid injection in the baboon (unpublished data from our laboratory); and in vitro studies have shown its high affinity ($K_i = 1.2 \text{ nM}$) and high selectivity for the DA receptors (Leysen and Gommeren 1978), although its lipophilicity was probably responsible for its high nonspecific binding (Leysen, to be published). Using i. v. injection of ^3H -labeled pimozide in trace doses in rats and mice, several authors have reported regional uptake and kinetics in brain consistent with specific binding to DA receptor in vivo, a finding supported by inhibition and displacement studies (Baudry et al. 1977; Laduron et al. 1978). Thus ^3H -pimozide accumulates in the DA-rich striatum during the first 2 h, whereas it washes out continuously from the cerebellum (a reference organ because of its lack of DA receptors), this difference in regional pharmacokinetics being abolished by inhibiting or displacing doses of cold competitor. However, these reports indicated that although better than haloperidol, pimozide was not quite as good as spiperone in terms of in vivo nonspecific binding (Laduron et al. 1978). On the other hand, spiperone binds to serotonergic receptors with very high affinity

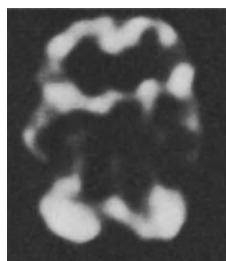
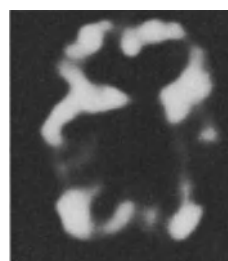
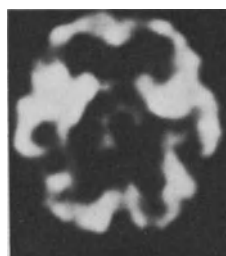
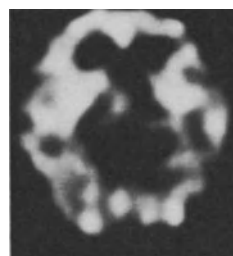
¹¹C - PIMOZIDE**STUDY 3****To + 14,5min****To + 56,5 min**
STUDY 9
HALOPERIDOL
10 mg I.M
**To + 6,5 min****To +43 min****SH FJ . Orsay**

Fig. 5. Typical ¹¹C-pimozide brain images from two studies, showing the redistribution of radioactivity with increasing time in the control study (*top*) but no such redistribution in a haloperidol pretreatment study (*bottom*). In the control study, the striatal area

is hypoactive relative to the cerebellum at early scan times, but has reached or surpassed cerebellar activity at late scan times. In the competition study, the striatal area remains stable with increasing time relative to the cerebellar area

as well, and hence is not a selective DA ligand. The application to in vivo PET study, however, requires that the labeling procedure with ¹¹C be easy, reproducible, and fast, and that pure ligand of high specific radioactivity at injection time be available, as is the case for ¹¹C-pimozide (Crouzel et al. 1980). In addition, the specific receptors studied must be in sufficient concentration in the brain structure studied, and the latter must be adequate in terms of both size and shape to allow imaging with current PET systems, as should be the case for the dopaminergic receptors in the striatum.

Finally, the approach has been shown to be feasible in previous PET in vivo studies of benzodiazepine receptors in baboon brain (Comar et al. 1979) and of muscarinic receptors in ba-

boon heart (Mazière et al. 1981), although some unexpected difficulties inherent to the in vivo approach were stressed (Comar et al. 1981).

Material and Methods**Patients**

Thirteen studies were performed. There were eight naive patients (group A) and five patients studied after prior i.m. injection of a 10-mg therapeutic dose of cold haloperidol (group B). Haloperidol is a widely used neuroleptic with high affinity for the DA receptors, and has been shown to prevent specific binding of ³H-pimozide in the striatum of rats and mice in

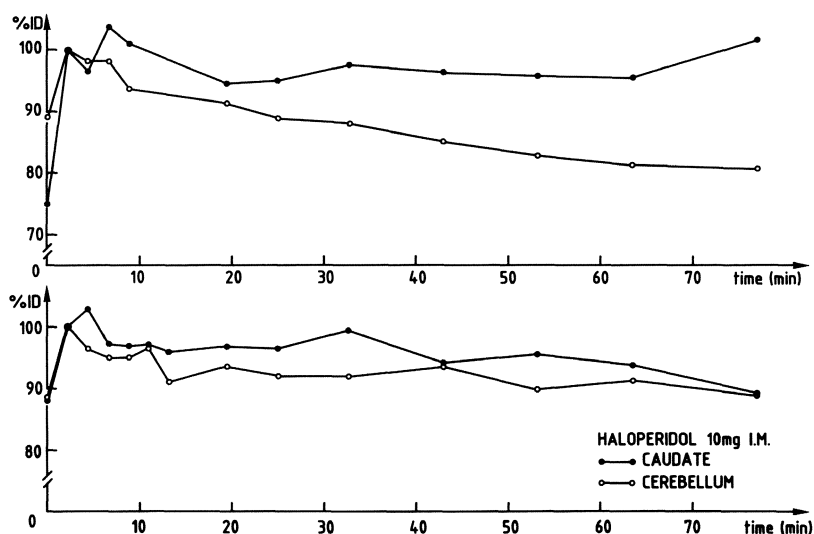


Fig. 6. Caudate and cerebellar time course of ^{11}C -pimozide concentration (expressed in % ID and normalized to 100% at $t = 2$ min) in the same patient, studied first in the control state (*top*) and one week later after i.m. injection of 10 mg haloperidol. Despite

the statistical fluctuations, retention is seen in the caudate but not in the cerebellum in the control state, whereas competition with cold haloperidol abolishes this difference

experiments in vivo (Baudry et al. 1977). The eight naive patients comprised six males and two females and ranged from 19 to 56 years of age. Six suffered from various disorders (two chronic alcoholism, one conversion neurosis, one transverse myelitis, one possible multiple sclerosis, one depression) and two were schizophrenics (one never treated before, one treated with neuroleptics which were withdrawn 8 days before study). For ethical reasons, the five group B patients were all chronic psychotics of the schizophrenic type (as was the untreated schizophrenic from group A, who was studied twice with a 1-week interval). Oral treatment with neuroleptics had usually been withdrawn 2–10 days before study.

Methods

The ^{11}C -pimozide was synthesized as published previously (Crouzel et al. 1980) and 7.7–25.6 mCi was injected intravenously. Specific radioactivity at the time of injection ranged from 20 to 295 mCi/ μmol (mean injected mass 142 nmol). Venous blood samples were collected and counted as indicated above.

A single brain level was scanned repeatedly for up to 73 min (10–14 scans) after i.v. injection.

General details concerning the detection system, quantitation procedure, and image reconstruction did not differ from the ^{11}C -DPH study. The brain level to study was chosen in order to obtain on the same image (see Fig. 5) a clear delineation of both the striatum and the cerebellum; for this purpose, the level was set at $+20^\circ$ rostral to the OM line. Total image counts were typically in the range $1.5\text{--}5 \times 10^5$.

Data Analysis. On the image with the highest number of counts, circular ROIs were sited visually over each cerebellar hemisphere and over each striatal area. The latter was usually clearly visible as an active zone just posterior to the frontal white matter – ventricular horn area (Fig. 5). The relative radioactive concentration (expressed as % ID) in each of these four ROIs was automatically decay-corrected and printed out for each of the 10–14 sequential scans. The time courses of the mean striatal and cerebellar values were then calculated and plotted on regular graph paper. Starting from the second data point (i.e., at $t = 2$ min), all the curves displayed a linear shape (see Fig. 6), and were thus processed by the least squares linear regression fitting procedure, expressing the % ID as a function of time for both striatum and cerebellum in

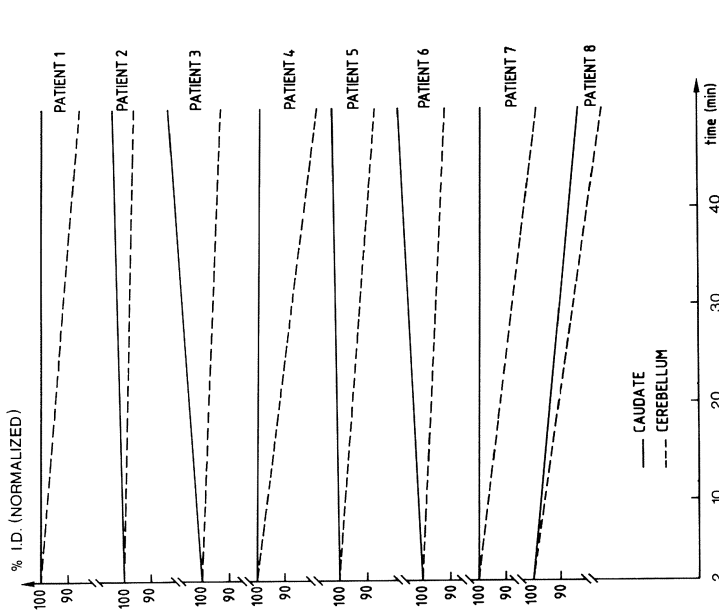
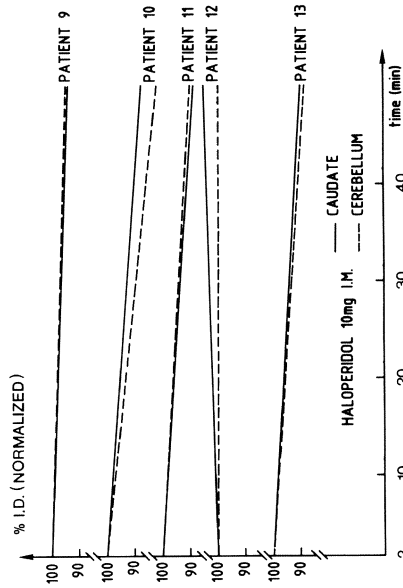


Fig. 7. Fitted regression lines of the caudate and cerebellar radioactive time course (normalized to 100% at $t = 2$ min) in the eight group A studies (*left*) and the five group B studies (*right*).

each subject. The same procedure was also applied to the anteromedial (cingulate) frontal cortex. Algebraic differences between the slopes obtained were calculated individually, taking the cerebellar slope as the reference (i.e., striatal slope to cerebellar slope).

Results

The time course of the whole blood ^{11}C -pimozide concentration showed a fast equilibration phase, followed by a nearplateau after 20 min. On the brain images of group A subjects, the time course of ^{11}C in striatum appeared to differ slightly from that in cerebellum, with the former structure becoming more and more in-



The curves are seen to diverge in all group A studies, but are essentially parallel in group B studies

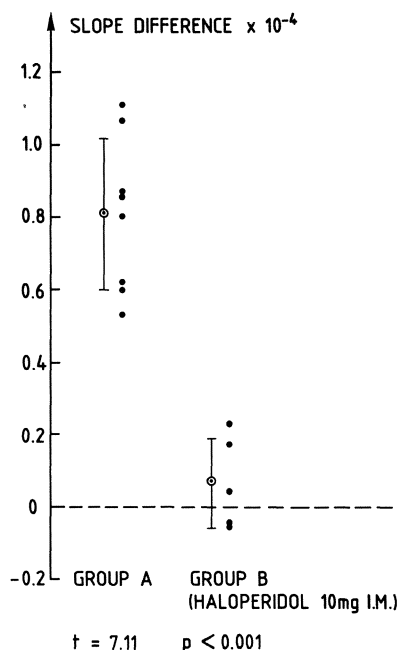


Fig. 8. Caudate-cerebellar slope difference in group A and group B studies (see text); mean and SD are also shown. There is no overlap between groups, and their difference is highly significant

tensely radioactive relative to the latter as time progressed. No such difference was visible in group B subjects (Fig. 5).

This regional difference in ^{11}C -pimozide kinetics was confirmed when the individual time-activity curves were inspected. In group A patients, the striatal curves exhibited a slow rise to plateaus in seven of eight studies, but the cerebellar curves decreased in all studies. The two structures displayed a parallel decrease (in four cases) or slight increase (in one case) in group B studies. An example is shown in Fig. 6.

In group A studies, the means ($\pm$ SD) of the calculated slopes (expressed in $\% \text{ID} \times 10^{-4} / 10^3 \text{ s}$) were -0.09 ± 0.58 (range $+0.43$ to -1.45) and -0.90 ± 0.67 (range -0.35 to -2.31) in striatum and cerebellum respectively ($p < 0.05$). The mean slope difference was $+0.81 \pm 0.21$ (range $+0.56$ to $+1.11$), which is significantly different from zero ($p < 0.001$). In group B studies, the results were -0.34 ± 0.30 (range $+0.19$ to -0.58) and -0.41 ± 0.26 (range -0.04 to -0.75) for striatum and cerebellum respectively (not significantly different). The mean slope difference was $+0.07 \pm 0.12$

(range $+0.23$ to -0.04), which is not different from zero but significantly different from group A ($p < 0.001$) (Figs. 7, 8).

The slopes obtained from the frontal cortex showed no significant differences from the cerebellar slopes in any of the above tests.

In group A, the calculated concentrations of ^{11}C -pimozide at $t = 57 \text{ min}$ were 2.65 nM (range 1.2 – 8.8) in caudate and 2.45 nM (range 1.1 – 7.5) in cerebellum. In group B studies, they were 0.99 nM (range 0.64 – 1.5) in caudate and 0.99 nM (range 0.6 – 1.7) in cerebellum. These values are not significantly different from one another.

Neither the caudate nor the cerebellar slopes (or their difference) were correlated to the amount of pimozide injected or to its local concentration in brain in group A studies. Both the caudate and cerebellar concentrations of ^{11}C -pimozide at $t = 57 \text{ min}$ were correlated linearly to the amount of pimozide injected ($r = 0.993$ and 0.997 respectively), but the regression lines were not different.

Comments

This PET study is the first to demonstrate in vivo in man phenomena that are presumably directly related to the presence of specific dopaminergic receptors in the striatum. Compelling evidence is provided by the following observations. First, there was a small but highly significant difference in the regional kinetics of ^{11}C -pimozide – a neuroleptic known to bind with high affinity to the DA receptors (Leysen and Gommeren 1978) – between striatum and cerebellum, two extreme brain structures in terms of their DA receptor concentration (Murrin 1981). In all naive patients studied, there was a retention of ^{11}C -pimozide in striatum and a decrease in cerebellum (Figs. 7, 8), similar to although not as prominent as that observed in rats or mice injected i.v. with trace amounts of ^3H -pimozide (Baudry et al. 1977; Laduron et al. 1978). Second, in keeping with the latter animal experiments, no such differences in the kinetics of ^{11}C -pimozide were seen in the frontal cortex when compared to cerebellum, a finding consistent with the relatively low density of DA terminals in this area (Berger et al. 1976). Third, in patients pretreated with unlabeled

beled haloperidol – a potent competitor of pimozone for binding to the DA receptors – in amounts presumably large enough to cause near complete occupation of the DA receptor sites, the regional kinetics of trace molecules of ^{11}C -pimozone this time showed no significant retention in striatum when compared to cerebellum (Figs. 7, 8), without affecting the kinetics in the frontal cortex. These results are consistent with the *in vivo* inhibition studies by Baudry et al. (1977) in mice (using pretreatment with haloperidol), which showed a dose-related effect on the kinetics of ^3H -pimozone but no effect on frontal or cerebellar kinetics. The displacement studies by Laduron et al. (1978) in rats showed similar results. The inference from the above and other *in vivo* studies was that the retention and accumulation of the labeled ligand in the striatum of naive animals is a direct expression of its specific, high-affinity binding to the DA receptors, although not all the criteria necessary to prove this hypothesis could be seen to exist (Laduron 1980); occupation of the DA specific sites by large amounts of unlabeled ligand would then allow the labeled ligand to wash out nonspecifically from brain at the same rate as that seen in areas devoid of DA receptors.

One could argue that since the rate of washout from brain must be in some way governed by the local blood flow, a differential effect of haloperidol on caudate flow could explain our findings. This seems unlikely, since it has been shown that neuroleptics in therapeutic doses do not affect cerebral blood flow to any great degree (Mac Culloch and Harper 1977). Another criticism would be that the difference in kinetics observed in group B patients is related not to pretreatment with haloperidol, but rather to the disease state of the subjects, namely schizophrenic-like psychosis, or alternatively to their chronic neuroleptic medication, the discontinuation of which can lead to DA receptor supersensitivity (Owen et al. 1980). However, the two schizophrenics who were included in group A did not differ from the remaining six controls, and the one unmedicated schizophrenic studied twice did show typical ^{11}C -pimozone kinetics on both occasions (Fig. 6).

Although this study showed convincingly that the nature of the observed kinetics was directly

related to specific binding of ^{11}C -pimozone to striatal DA receptors, it is true that the magnitude of the phenomena observed, although highly significant, was relatively small.

Particularly, in contrast to the findings reported in animals (Baudry et al. 1977; Laduron et al. 1978), we could not observe any reliable difference in ^{11}C -pimozone concentrations in striatum and cerebellum at 60 min after *i.v.* injection in naive patients. In rats and mice, striatal ^3H -pimozone concentrations were found to be 60% and 100% higher than in cerebellum at 1 and 2 h respectively (Baudry et al. 1977; Laduron et al. 1978), a difference taken as representing specific binding to DA receptors in striatum. Many facts partly explain this discrepancy. First, as a combined result of the constraints of ^{11}C -radiochemistry, acceptable statistics, and radiation exposure to the patient, the mass of pimozone injected resulted in striatal concentrations ranging from 1.2 to 8.8 nM, *i.e.* levels at which near saturation of the DA receptor sites is expected to occur, with the consequent masking of specific binding by nonspecific binding. Second, the human striatum is of a size and shape such that faithful imaging with our current PET device is not expected (Mazziotta et al. 1981): the partial volume effect results in dilution of its radioactive concentration by that of neighboring structures exhibiting low, exclusively nonspecific binding, *e.g.*, white matter and ventricles. Third, because of the rapid physical decay of ^{11}C ($T_{1/2} = 20$ min), brain imaging could not be pursued for more than an hour or so, thus minimizing the difference in uptake between striatum and cerebellum. Finally, dopaminergic ligands with higher affinity and lower nonspecific binding than pimozone, *e.g.*, spiperone (Laduron et al. 1978), would probably have provided more clear-cut results, but radiochemical labeling of spiperone with ^{11}C is difficult.

The kinetic approach used here, although limited, nevertheless provided highly significant and reproducible results. By showing regional differences in pharmacokinetics consistent with known *in vitro* distribution of DA receptors, and abolished by a competition procedure, three of the five criteria necessary for *in vivo* demonstration of specific binding have been fulfilled (Laduron 1980). Pathophysiological studies investigating the striatal DA recep-

tors population in various disorders are now in the planning stage.

In this work, substantial steps forward are likely to come from simultaneous improvements in the three fields of radiochemistry (better ligands, higher specific radioactivity, ^{18}F labeling), detection (improved spatial resolution and sensitivity), and neuroscience (modelling for in vivo quantitative estimation of receptor concentration and affinity).

However, the labeled ligand will still have to be lipophilic in order to penetrate the blood-brain barrier, its uptake by the brain therefore being governed mainly by nonspecific binding. Only displacement or competition experiments will then enable differentiation of specific and nonspecific components.

References

^{11}C -Diphenylhydantoin

1. Alvin JD, Bush MT (1977) Diphenylhydantoin and other hydantoins In: Vida JA (ed) *Anticonvulsants* Academic, New York p116
2. Baron JC, Bousser MG, Comar D, Soussaline F, Castaigne P (1981) Non-invasive tomographic study of cerebral blood flow and oxygen metabolism in vivo: potentials, limitations and clinical applications in cerebral ischemic disorders. *Eur Neurol* 20: 273–284
3. Firemark H, Barlow CF, Roth LJ (1963) The entry accumulation and binding of diphenylhydantoin-2- C^{14} in brain – studies on adult, immature and hypercapnic cats. *Int J Neuropharmacol* 2: 25–38
4. Goldberg MA, Crandall PH (1978) Human brain binding of phenytoin *Neurology* 28: 881–885
5. Harvey CD, Sherwin AL, Van der Kleyn E (1977) Distribution of anticonvulsant drugs in gray and white matter of human brain. *Can J Neurol Sci* 4: 89–92
6. Houghton GW, Richens A, Toseland PA et al. (1975) Brain concentration of phenytoin, phenobarbitone and primidone in epileptic patients. *Eur J Clin Pharmacol* 9: 73–78
7. Kutt H, Louis S, McDowell F (1968) Intravenous diphenylhydantoin in experimental seizures. *Arch Neurol* 18: 465–471
8. Munari C, Talairach J, Rovei V et al. (1980) Brain levels of antiepileptic drugs in man. *Epilepsy: a clinical and experimental research*. *Monogr Neural Sci* 5: 213–220
9. Ramsay RE, Hammond EJ, Perchalsky RJ et al. (1979) Brain uptake of phenytoin phenobarbital and diazepam. *Arch Neurol* 36: 535–539
10. Rapport RL, Harris AB, Fried PN et al. (1975)

Human epileptic brain. Na, K, ATP-ase activity and phenytoin concentrations. *Arch Neurol* 32: 549–554

11. Roeda D, Westera G (1981) The synthesis of some ^{11}C -labelled antiepileptic drugs with potential utility as radiopharmaceuticals: hydantoins and barbiturate. *Int J Appl Radiat Isot* 32: 843–845
12. Sherwin AL, Eisen AA, Sokolowski CD (1973) Anticonvulsant drugs in human epileptogenic brain. Correlation of phenobarbital and diphenylhydantoin levels with plasma. *Arch Neurol* 29: 73–77
13. Sironi VA, Gabrini G, Porro MG et al. (1980) Antiepileptic drug distribution in cerebral cortex, Ammon's horn, and amygdala in man. *J Neurosurg* 52: 686–692
14. Stavchansky SA, Tilbury RS, McDonald JM et al. (1978) In vivo distribution of carbon-11 phenytoin and its major metabolite and their use in scintigraphic imaging. *J Nucl Med* 19: 936–941
15. Vajda F, Williams FM, Davidson S et al. (1974) Human brain, cerebrospinal fluid and plasma concentrations of diphenylhydantoin and phenobarbital. *Clin Pharmacol Ther* 15: 597–603
16. Wilder BJ, Ramsay RE, Willmore LJ et al. (1977) Efficacy of intravenous phenytoin in the treatment of status epilepticus: kinetics of central nervous system penetration. *Ann Neurol* 1: 511–518
17. Winstead MB, Parr SJ, Royal MJ (1976) Relationship of molecular structure to in vivo scintigraphic distribution patterns of carbon-11 labeled compounds -3-(^{11}C) hydantoins. *J Med Chem* 19: 279–286

^{11}C -Pimozide

18. Baudry M, Martres MP, Schwartz JC (1977) In vivo binding of ^3H -pimozide in mouse striatum. Effects of dopamine agonists and antagonists. *Life Sci* 21: 1163–1170
19. Berger B, Thierry AM, Tassin JP, Moyne MA (1976) Dopaminergic innervation of the rat prefrontal cortex: a fluorescence histochemical study. *Brain Res* 106: 133–145
20. Comar D, Mazière M, Godot JM, Berger G, Soussaline F, Menini C, Arfel G, Naquet R (1979) Visualisation of ^{11}C -flunitrazepam displacement in the brain of the live baboon. *Nature* 280: 329–331
21. Comar D, Mazière MA, Cepeda C, Godot JM, Menini C, Naquet R (1981) The kinetics and displacement of ^{11}C -flunitrazepam in the brain of the living baboon. *Eur J Pharmacol* 75: 21–26
22. Crouzel C, Mestelan G, Kraus E, Lecomte JM, Comar D (1980) Synthesis of a ^{11}C -labelled neuroleptic drug: pimozide. *Int J Appl Radiat Isot* 31: 545–548
23. Janssen PAJ, Allewijn FTN (1968) Pimozide, a clinically novel, highly potent and orally long-act-

- ing neuroleptic drug. II. Kinetic study of the distribution of pimozide and metabolites in brain, liver, and blood of the Wistar rat. *Arzneim Forsch* 18: 282–287
24. Laduron PM (1980) Dopamine and muscarinic receptors: in vivo identification, axonal transport and solubilization. In: Yamamura, Olsen, Usdin (eds) *Psychopharmacology and biochemistry of neurotransmitter receptors*. Elsevier, Amsterdam pp 115–132
 25. Laduron PM, Jansen PFM, Leysen JE (1978) Spiperone, a ligand of choice for neuroleptic receptors. 2. Regional distribution and in vivo displacement by neuroleptic drugs. *Biochem Pharmacol* 27: 371–321
 26. Leysen JE (to be published) Receptors for neuroleptic drugs. In: Burrows GD, Weney JS (eds) *Advances in human psychopharmacology*, vol 3. JAI Press, Greenwich
 27. Leysen JE, Gommeren W (1978) Different kinetic properties of neuroleptic receptor binding in the rat striatum and frontal cortex. *Life Sci* 23: 447–452
 28. Mac Culloch JM, Harper AM (1977) Cerebral circulation: effect of stimulation and blockade of dopamine receptors. *Am J Physiol* 233: H222–H227
 29. Mazière M, Comar D, Godot JM, Collard P, Cepeda C, Naquet R (1981) In vivo characterization of myocardium muscarinic receptors by positron emission tomography. *Life Sci* 29: 2391–2397
 30. Mazziotta JC, Phelps ME, Plummer D, Kuhl DE (1981) Quantitation in positron emission computed tomography: 5. Physical anatomical effects. *J Comput Assist Tomog* 5: 734–743
 31. Murrin LC (1981) Neurotransmitter receptors, neuroanatomical localization through autoradiography. *Int Rev Neurobiol* 22: 111–171
 32. Owen F, Cross AJ, Waddington JL, Poulter M, Gamble SJ, Crow TJ (1980) Dopamine-mediated behaviour and ³H-spiperone binding to striatal membranes in rats after nine months haloperidol administration. *Life Sci* 26: 55–59

Mini-Papers

(Chairman: L. Sokoloff)

Potential Bromine-75-Labeled Brain Radiopharmaceuticals

By G. Kloster, H. Scholl, P. Laufer, W. Wutz, and G. Stöcklin

Bromine-75 offers interesting perspectives for positron-emitting radiopharmaceuticals. At Jülich, we have been investigating ^{75}Br production methods (Blessing et al. 1982; Weinreich et al. 1980) and labeling procedures for some years (Coenen et al., in preparation). ^{75}Br is a predominant (75.5%) positron-emitting nuclide with a 1.6-h half-life. Besides the 511-keV annihilation radiation, its principal γ -line is at 286 keV (abundance 92%). It decays to the long-lived radioactive daughter ^{75}Se ($T_{1/2} = 120$ days), which does not contribute significantly to the radiation dose, at worst amounting to one-third of the total dose.

The advantages of ^{75}Br over other positron emitters like ^{11}C or ^{18}F are: (a) large amounts (up to 200 mCi) can be produced (Blessing et al. 1982) at the no-carrier-added (nca) level, resulting in very high specific activities that cannot be attained with ^{11}C ; (b) the chemistry of bromine is much simpler than that of fluorine and many high-yield bromination reactions are known; so large activities of ^{75}Br radiopharmaceuticals can be produced. The disadvantage lies mainly in the higher rate of random coincidences due to the 286-keV γ -line, leading to

noisy pictures; however, several correction methods can be applied to avoid the higher background.

Using ^{75}Br , we have investigated two compounds as potential brain radiopharmaceuticals, namely a sugar derivative labeled in the 2-position (Fig. 1) and a 1,4-benzodiazepine labeled in the 7-position (Fig. 2).

During an unsuccessful attempt to synthesize 2-deoxy-2-bromo-D-glucose (Kloster et al. 1982a), we observed that one of the intermediate products of that synthetic sequence, namely 1-methyl-3,4,6-triacetyl-2-deoxy-2-bromo-D-glucoside (MTBG), rapidly entered mouse brain. MTBG was synthesized from the commercially available triacetyl-D-glucal and $^{75}\text{Br}^-$ (nca) in methanol using dichloramine T as oxidizing agent (see Fig. 1). After chromatographic separation of MTBG from the corresponding manno derivative, a radiochemical yield of 40% was obtained; the reaction time was less than 1.5 h.

Biodistribution data in mice (Fig. 3) show that MTBG rapidly enters the brain after i.v. injection, reaching a maximum of 160% mean body concentration (% MBC) 0.25 min after administration. At the same time, the blood concentration reaches 188% MBC. The brain to blood concentration ratio stays fairly constant at about 0.8 during the first 10 min after administration, while the absolute concentration in brain falls to 106% MBC at 10 min. Considerable activity is observed in the kidneys, indicating possible renal excretion.

Nothing is known about the mechanism by

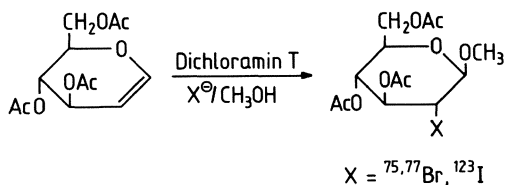


Fig. 1. Synthesis of MTBG

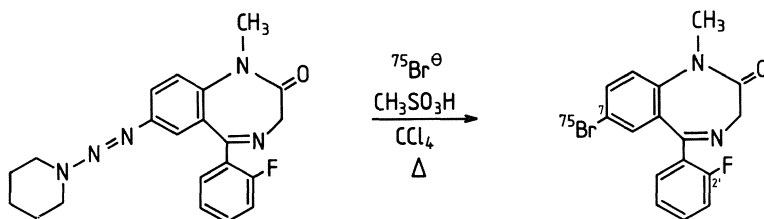


Fig. 2. Synthesis of BFB

which MTBG enters the brain. If it were a substrate for the hexose carrier of the blood-brain barrier, MTBG would be a longer-lived alternative to 3- ^{11}C -methyl-D-glucose (Kloster et al. 1981; 1982b) as a tracer for hexose transport. It was shown, however, that MTBG remains unchanged in the mouse brain at 5 min after application, indicating that hydrolysis or dehalogenation does not take place.

A different approach was used in the development of the second radiopharmaceutical. 1,4-Benzodiazepines that contain a bromine atom in the 7-position are known to have high affinity for the benzodiazepine receptor of rat brain; so a compound having a ^{75}Br label in that position should be useful for in vivo mapping of brain benzodiazepine receptor areas.

We prepared 7- ^{75}Br -5-(2'-fluorophenyl)-1-methyl-1,3-dihydro-2H-1,4-benzodiazepin-2-one (BFB) from $^{75}\text{Br}^-$ by acid-catalyzed decomposition of a triazene precursor (see Fig. 2) (Scholl et al. 1982). Up to 14 mCi BFB was prepared in 1 h, resulting in 20% radiochemical yield. The specific activity was greater than 17000 Ci/mmol. Compared to ^{11}C -flunitrazepam prepared for the same purpose by the Orsay group (Comar et al. 1979), the specific activity is higher by a factor of at least 40; furthermore, due to the longer half-life of ^{75}Br , the time available for in vivo investigation is longer by a factor of 5.

Biodistribution data in mice show that much like ^{11}C -flunitrazepam (Comar et al. 1979), BFB enters the brain rapidly after i.v. injection, reaching a maximum of 188% MBC 0.25 min

after administration, followed by a decrease to about 100% MBC after 10 min (see Fig. 4). Over this period the relatively low blood level of 45% MBC is nearly constant. If these data can be extrapolated to man, BFB is expected to be a useful radiopharmaceutical for in vivo mapping of brain benzodiazepine receptor areas. BFB is bound to the benzodiazepine receptor rather specifically, having an inhibition constant (Möhler and Okada 1977) of 1.7 nM (Möhler, personal communication). Thus, we expect BFB to have a high degree of specific binding.

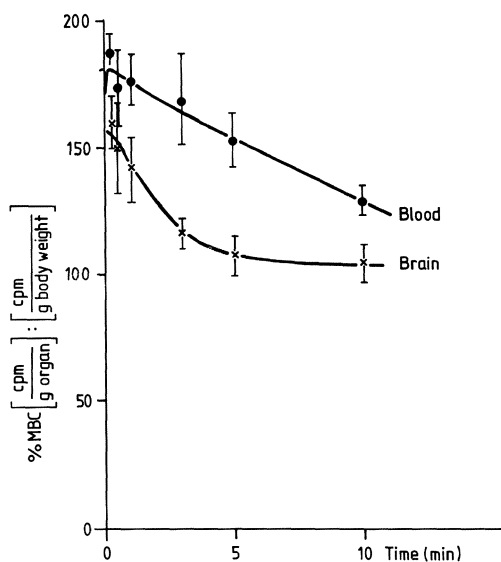


Fig. 3. Biodistribution of MTBG in mice (i.v. injection)

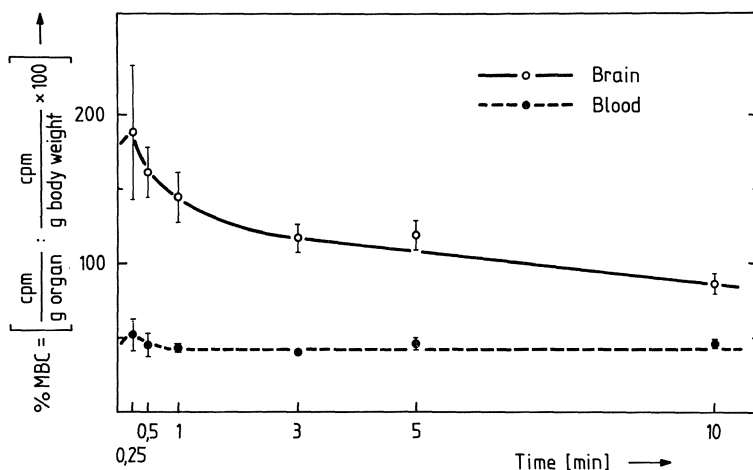


Fig. 4. Biodistribution of BFB in mice (i.v. injection)

At present we are evaluating the clinical relevance of BFB in collaboration with the Max Planck Institute for Neurological Research, Cologne (Prof. W.-D. Heiss).

Acknowledgements. We thank Dr. H. Möhler, Hoffmann-La Roche, Basel, for determining the inhibition constant of BFB. We further thank Hoffmann-La Roche for the generous gift of various benzodiazepines.

References

- Blessing G, Weinreich R, Qaim SM, Stöcklin G (1982) Production of ^{75}Br and ^{77}Br via the $^{75}\text{As}(^3\text{He},3n)^{75}\text{Br}$ and $^{75}\text{As}(\alpha,2n)^{77}\text{Br}$ reactions using Cu_3As -alloy as a high-current target material. *Int J Appl Radiat Isot* 33: 333–339
- Comar D, Mazière M, Godot JM, Berger G, Soussaline F, Menini C, Arfel G, Naquet R (1979) Visualization of ^{11}C -flunitrazepam displacement in the brain of the live baboon. *Nature* 280: 329–331
- Kloster G, Müller-Platz C, Laufer P (1981) 3- ^{11}C -Methyl-D-glucose, a potential agent for regional cerebral glucose utilization studies: synthesis, chromatography and tissue distribution in mice. *J Labelled Compd Radiopharm* 18: 855–863
- Kloster G, Laufer P, Wutz W, Stöcklin G (1982a) ^{75}Br - and ^{123}I -analogues of D-glucose as potential radiopharmaceuticals. *J Labelled Compd Radiopharm* 19: 1626–1628
- Kloster G, Stöcklin G, Vyska K, Freundlieb C, Höck A, Feinendegen LE, Traupe H, Heiss W-D (1982b) 3- ^{11}C -Methyl-D-glucose, an agent for the assessment of regional glucose transport across the blood-brain barrier. In: Cox PH (ed) *Progress in radiopharmacology*, vol 3. Nijhoff, Den Haag pp 199–211
- Möhler H, Okada T (1977) Benzodiazepine receptor: demonstration in the central nervous system. *Science* 198: 849–851
- Scholl H, Laufer P, Kloster G, Stöcklin G (1982) A potential benzodiazepine receptor-binding radiopharmaceutical for positron emission tomography: 7- ^{75}Br -5-(2'-Fluorophenyl)-1-methyl-1,3-dihydro-2H-1,4-benzodiazepin-2-one. *J Labelled Compd Radiopharm* 19: 1294–1295
- Weinreich R, Alfassi ZB, Blessing G, Stöcklin G (1980) Short-lived neutron-deficient bromine isotopes for applications in nuclear medicine. *Nucl Med [Suppl 17]*: 202–205

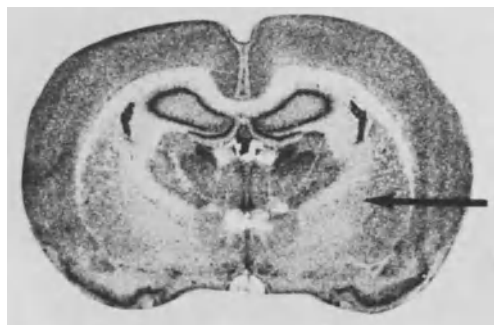
Biochemical Prerequisites for Measurement of Protein Synthesis

By W. Bodsch

One of the basic prerequisites for quantitative determination of cerebral metabolic rates of protein synthesis employing tracer quantities of radioactive-labeled amino acids is the measurement of the time-integrated specific activity of aminoacyl transfer ribonucleic acids (aa-tRNAs), i.e., the actual precursors for protein biosynthesis, and the stability of labeled amino acids trapped in the cerebral proteins. Besides autoradiographic visualization of *in vivo* labeled proteins from cryostat sections of the brain, biochemical analysis can be performed on tissue microsomes taken regionally between sectional planes.

The precursor-product relationship in rat brain protein synthesis was evaluated by a double-isotope incorporation procedure (Bodsch and

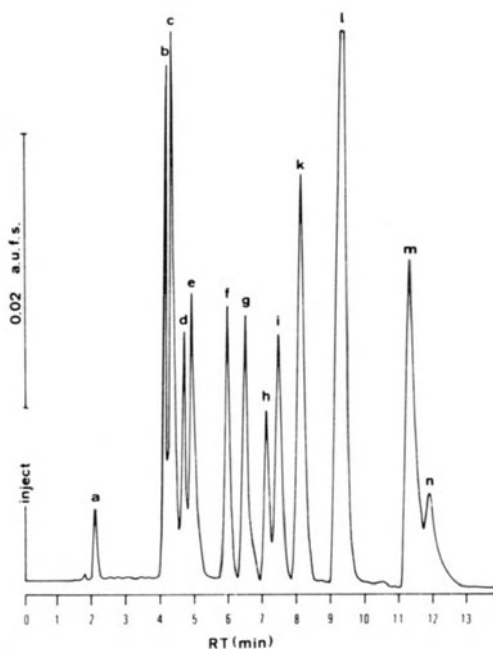
Hossmann 1981) where the labeling experiments were started by intravenous bolus administration of ^{14}C -carboxyl-labeled amino acids (phenylalanine, tyrosine, isoleucine, leucine, and methionine; 20 μCi at 50 mCi/mmol each, total 100 μCi), continued by a pulse of the same mixture of amino acids in the ^3H -labeled form (150 μCi at 90 Ci/mmol each, total 0.75 mCi) at 35 min, and completed at 45 min. Hence the local tissue concentration of cerebral proteins allowed for the identification of two time points on a synthesis curve in the same proteins in the same animal. Furthermore, the direct precursor pool, formed by the aa-tRNAs, is labeled at two different phases: ^{14}C marks the pool conditions at the end of the experiment, while ^3H reflects the aa-tRNA labeling at



a

Fig. 1. a ^{14}C -Autoradiogram of protein synthesis from a coronal section of the rat brain labelled *in vivo* with ^{14}C -carboxyl- and ^3H -labeled amino acids. Autoradiography on Kodak NMB film records exclusively the ^{14}C label. Microsamples of brain tissue (ca. 100 μg) were taken behind the cryostat section from the CA region of the hippocampus. **b** The microsamples were analyzed by HPLC for low-molecular-weight proteins. Quantitative detection of ^3H and ^{14}C radioactivity was performed according to Laskey and Mills (1975) after collection of various peak fractions. The ^3H to ^{14}C ratio of the peptide peaks was: *b*, 0.95; *c*, 0.86; *d*, 0.88; *e*, 0.90; *k*, 1.10; *l*, 1.02; *m*, 1.21. RT, retention times of HPLC peaks

PROTEIN SYNTHESIS



b

10 min after onset of the first administration of labeled amino acids to the animal, where labeled aa-tRNAs already attained maximal levels of specific activities (data not shown). Autoradiography of protein synthesis was monitored on Kodak NMB X-ray films, which are low sensitive to ^3H , and therefore showed ^{14}C -protein-incorporated amino acid activity almost exclusively.

A protein synthesis autoradiogram of the rat brain is illustrated in Fig. 1. Intense autoradiographic density is seen in the CA regions of pyramidal cells in the hippocampus. When tissue microsamples of these cellular structures indi-

cating high protein synthesis were subjected to peptide and protein analysis by high-performance liquid chromatography (HPLC), soluble cellular proteins turned out to belong mainly to the low-molecular-weight proteins, i.e., proteins with a chain length of about 30–100 amino acid residues (as demonstrated in the HPLC profile). The ^3H and ^{14}C radioactivity cochromatographing with the protein peaks was counted, and the ^3H to ^{14}C ratios gave measurable values of the turnover of the desired protein. Proteins with a ^3H to ^{14}C ratio greater than 1.0 are degraded at a higher rate than proteins exhibiting a ^3H to ^{14}C ratio lower than 1.0.

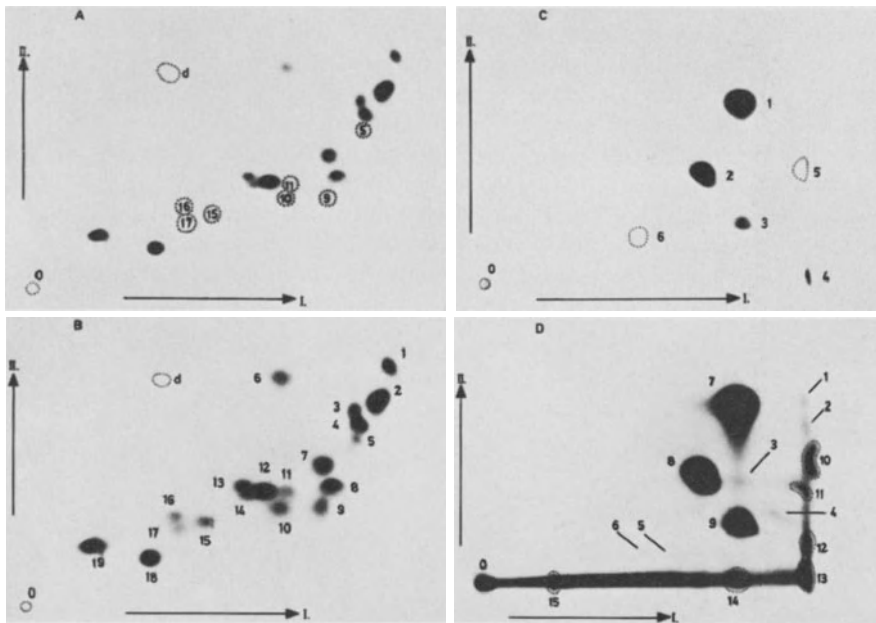


Fig. 2 A–D. Microanalysis of amino acids attached to tRNA molecules from a 100 μg tissue sample of the cerebral cortex of the rat brain. Amino acids released from tRNAs, aa-tRNAs, and free tRNAs were extracted and separated by HPLC as described (Bodsch and Hossmann 1983).

A Autoradiogram of a two-dimensional thin layer separation of a synthetic mixture of ^{14}C -amino acids that do not contain the amino acids used for in vivo labels in brain protein synthesis. **B** A separation of amino acids released from in vivo charged tRNAs by alkaline treatment. Weak spots in the autoradiogram represent the ^3H -labeled forms of tyrosine (9), phenylalanine (10), isoleucine (11), leucine (16), and methionine (15). All the other spots (marked 1–19) were uninvolved as labels in vivo and are therefore contained in the separations as markers. **C** Autoradiography of ^3H aa-tRNAs separated electrophoretically

in the first dimension (I) and chromatographed on PET thin layer plates in the second dimension (II). (^3H -methionyl-tRNA was determined separately by reaction specific for the sulphur group of methionine). Spots on LKB ultrofilm were identified as ^3H -aa-tRNAs of tyrosine (1), phenylalanine (2), leucine (3), and isoleucine (4). Circles 5 and 6 mark the positions of marker dyes. **D** Free tRNAs from rat brain samples were assayed after reaction of the free 3' end of the tRNA molecules with ^{32}P -ATP according to Winter and Brownlee (1978) and separated two-dimensionally as described in C. Autoradiography on Kodak no-screen X-ray film revealed the free tRNA species to tyrosine (7), phenylalanine (8), leucine (9), and isoleucine (13), besides several other spots that represent various fractionated and unfractionated tRNA species. 0, origin of separations

If the ratio is close to 1.0, protein synthesis and degradation are in a steady state. This procedure thus allows evaluation of steady-state relationships of proteins, i.e., verification of the stability of amino acid label in proteins during the labeling experiment.

Microanalysis of the actual precursor pool for brain protein synthesis, i.e., aa-tRNAs, demonstrated in Figure 2, summarized the procedures used to evaluate quantitative precursor measurements. Since charging of tRNA molecules with amino acids occurs at the 3'-end of the tRNA, ^3H -aa-tRNAs were visualized for ^3H radioactivity cochromatographing with the tRNAs (Fig. 2C). Uncharged free tRNAs, which do not contain amino acids at the 3'-end of the molecule, were reacted with a group-specific enzyme and ^{32}P -ATP so that free tRNAs became labeled exclusively (Fig. 2D). This allowed a quantitative determination of the specific activities of amino acids in the aa-tRNA pool used for brain protein synthesis.

Because several tRNA molecules exist for each amino acid in rat and human neuronal cells (Rogg et al. 1977) and are charged with amino acids randomly only a minor fraction of charged aa-tRNAs are involved in brain pro-

tein synthesis. The quantitative assessment of specific activities in this precursor pool seems to be necessary for determination of protein synthesis rates. Regarding positron emission tomography of protein synthesis in man, these relationships should be determined separately in human cell cultures and with purified tRNAs from human sources in vitro.

References

1. Bodsch W, Hossmann K-A (1981) Regional protein and RNA synthesis in rat brain. Autoradiography and quantitative HPLC. *Neurosci Lett [Suppl]* 7: 431
2. Bodsch W, Hossmann K-A (1983) A quantitative regional analysis of amino acids involved in rat brain protein synthesis by high-performance liquid chromatography. *J Neurochem* 40:
3. Laskey RA, Mills AD (1975) Quantitative film detection of ^3H and ^{14}C in poly-acrylamide gels by fluorography. *Eur J Biochem* 56: 335-341
4. Rogg J, Müller P, Keith G, Staehelin M (1977) Chemical basis for brain-specific serine transfer RNA. *Proc Natl Acad Sci USA* 74: 4243-4247
5. Winter G, Brownlee GG (1978) 3'-end labelling of RNA with ^{32}P suitable for rapid gel sequencing. *Nucleic Acids Res* 5: 3129-3139

Discrepancy Between Glucose Availability and Calculated Glucose Consumption During Acute Brain Ischemia

By G. Mies, W. Paschen, and K.-A. Hossmann

Vascular occlusion of the common carotid artery in the gerbil (*Meriones unguiculatus*) is a well-established animal brain infarction model. We have used a modification of this method (Bosma et al. 1981) in order to study the hemodynamic and metabolic changes during the acute stage of graded brain ischemia. Different regional methods were applied in the same animal: an autoradiographic double-tracer technique (Mies et al. 1981) for the simultaneous determination of cerebral blood flow with ^{131}I -iodoantipyrine (Sakurada et al. 1978) and glucose utilization with ^{14}C -deoxyglucose (Sokoloff et al. 1977), and a bioluminescent approach for the assessment of regional content of glucose (Paschen et al. 1981) and ATP (Kogure and Alonso 1978) from intact brain sections.

In control animals, cortical blood flow and glucose consumption were 1.1 ml/g per minute and 0.8 $\mu\text{mol/g}$ per minute respectively. In animals which exhibited no or mild symptoms, an initial depletion of tissue glucose content was detected in circumscribed areas where blood flow decreased to about 0.2 ml/g per minute. At a perfusion rate of 0.12–0.18 ml/g per minute regional ATP concentration diminished and glucose utilization began to increase. However, when blood flow fell below 0.2 ml/g per minute a discrepancy between the local glucose availability and the computed metabolic rates of glucose for identical brain areas became ap-

parent. As demonstrated in Fig. 1, the ^{14}C -deoxyglucose method used with normal constants calculates a value of 2.2 $\mu\text{mol/g}$ per minute. Assuming a 100% extraction for glucose at a plasma glucose concentration of 120 mg%, the maximal glucose consumption at a blood flow of 0.19 ml/g per minute is 1.26 $\mu\text{mol/g}$ per minute glucose. This indicates that the ^{14}C -deoxyglucose method overestimates the local glucose consumption by at least more than 75%.

Several factors may explain this discrepancy. There is a time lag of 30 min between the determination of glucose consumption with ^{14}C -deoxyglucose and blood flow with ^{131}I -iodoantipyrine, and regional perfusion might have changed during this period. However, in studies on the time course of blood flow changes after the onset of carotid artery occlusion in the gerbil, perfusion in the infarcted territories did not change by more than 15% (G. Mies, in preparation). Calculation of the tissue deoxyglucose precursor according to the original equation derived by Sokoloff and co-workers (1977) gives a value of about 25% of free deoxyglucose, which agrees very well with the measured amount determined in intact brain tissue. The possibility could be excluded that in low-perfused brain areas the accumulation of free ^{14}C -deoxyglucose caused the intensive blackening in the ^{14}C -autoradiograms. In tissue samples derived from infarcted brain regions,



Fig. 1. Cerebral blood flow, glucose metabolism, and distribution of glucose and ATP during acute brain ischemia. Quantitative measurement was performed in brain area as shown by the arrows. Blood flow was 0.19 ml/g per minute and calculated glucose con-

sumption was 2.20 $\mu\text{mol/g}$ per minute. Glucose availability, however, was only 1.26 $\mu\text{mol/g}$ per minute indicating an overestimation of local glucose consumption as determined with the ^{14}C -deoxyglucose method

analysis of ^{14}C -deoxyglucose revealed that 90%–95% of deoxyglucose was phosphorylated over a period of 30 min. On the other hand, equilibrium of ^{14}C -deoxyglucose between plasma and brain tissue must occur more rapidly to explain the drastic increase of total ^{14}C tissue concentration in these brain areas, but variation of $k_2^* + k_3^*$ from 0.2 to 0.5 only results in a change of 2% of total plasma specific activity. Since the total ^{14}C -deoxyglucose-6-phosphate concentration is known, it can be inserted in the numerator of the operational equation. Setting $k_2^* + k_3^* = 0.3$, the recalculation of regional glucose utilization using the normal lumped constant with 0.483 still overestimates the metabolic rate of glucose by more than 80%. Recently, it has been reported that the lumped constant increases by more than 100% when tissue glucose content decreases below $1\text{ }\mu\text{mol/g}$ brain tissue (Crane et al. 1981). Since regional glucose concentration is almost zero in areas of overestimated metabolic rates (see Fig. 1), it is concluded that the lumped constant in these particular brain regions may increase as well. The calculation of glucose utilization with a lumped constant of 0.84 gives a metabolic rate of glucose of $1.26\text{ }\mu\text{mol/g}$ per minute which is in agreement with the actual glucose availability.

In summary, at flow rates below 0.2 ml/g per minute in regions of acute brain infarction a discrepancy between glucose availability and glucose utilization as calculated with the ^{14}C -deoxyglucose method has been observed. The most plausible explanation for this finding is a change of the lumped constant.

References

1. Bosma HJ, Paschen W, Hossmann K-A (1981) Cerebral ischemia in gerbils using a modified vascular occlusion model. In: Meyer JS, Lechner H, Reivich M, Ott EO, Aranibar A (eds) *Cerebral Vascular Disease 3*, Excerpta Medica, Amsterdam, pp280–285
2. Crane PD, Pardridge WM, Braun LD, Nyerges AM, Oldendorf WH (1981) The interaction of transport and metabolism on brain glucose utilization: A reevaluation of the lumped constant. *J Neurochem* 36: 1601–1604
3. Kogure K, Alonso OF (1978) A pictorial presentation of endogenous brain ATP by a bioluminescence method. *Brain Res* 154: 273–284
4. Mies G, Niebuhr I, Hossmann K-A (1981) Simultaneous measurement of blood flow and glucose metabolism by autoradiographic techniques. *Stroke* 12: 581–588
5. Paschen W, Niebuhr I, Hossmann K-A (1981) A bioluminescence method for the demonstration of regional glucose distribution in brain slices. *J Neurochem* 36: 513–517
6. Sakurada O, Kennedy C, Jehle J, Brown JD, Garbin GL, Sokoloff L (1978) Measurement of local cerebral blood flow with iodo(^{14}C)antipyrine. *Am J Physiol* 234: H59–H66
7. Sokoloff L, Reivich M, Kennedy C, Des Rosiers MH, Patlak CS, Pettigrew KD, Sakurada O, Shinohara M (1977) The ($2\text{-}^{14}\text{C}$) deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–917

The Relationship of Regional Cerebral Blood Flow, Oxygen Metabolism and Glucose Metabolism Following Acute Stroke

By J. M. Gibbs, C. G. Rhodes, R. J. S. Wise, R. S. J. Frackowiak, and T. Jones

Previous studies using positron emission tomography (PET) have established that the normal coupling of cerebral blood flow (CBF) and oxygen metabolism (CMRO_2) breaks down in acute stroke (Ackerman et al. 1981; Baron et al. 1981; Lenzi et al. 1981). Initially, there are areas of ischaemic but still surviving brain in which CMRO_2 is preserved in relation to the marked fall of blood flow; the oxygen extraction ratio (OER) is high. Prolonged severe ischaemia leads to a subsequent fall of CMRO_2 (and hence also of OER) even if there is no further reduction of blood flow. This sequence of events is considered to represent the transition from potentially reversible ischaemia to established and irreversible cerebral infarction. In order to examine the relationship of regional cerebral glucose metabolism (rCMRGlu) to these pathophysiological changes, we have carried out combined, consecutive measurements of CBF, CMRO_2 and CMRGlu in patients with acute stroke.

Patients and Methods

This preliminary report includes the data from five patients studied between 3 and 21 days after the onset of acute hemispheric cerebral infarction. CBF, OER and hence CMRO_2 were measured by PET during the steady-state inhalation of C^{15}O_2 and $^{15}\text{O}_2$ as described elsewhere in this volume (Jones et al.). CMRGlu was then determined by scanning 50–60 min after intravenous injection of ^{18}F -fluoro-2-deoxyglucose, quantitative values being calculated using a simplified Sokoloff equation (Sokoloff et al. 1977) and a value for the lumped constant quoted for normal brain (Huang et al. 1980). Correction for the effects of blood volume was carried out by means of a final scan after bolus inhalation of ^{11}C -carbon monoxide (Phelps et al. 1979; Lammertsma et al. 1981).

Table 1. Mean values for all patients

	Within infarct	Contralateral cortex
	(mean $\pm$ SD)	
CBF (ml/100 ml/min)	13.5 $\pm$ 5.3	34.3 $\pm$ 5.8
OER	0.16 $\pm$ 0.11	0.42 $\pm$ 0.07
GER	0.17 $\pm$ 0.04	0.12 $\pm$ 0.01
CMRO_2		
(ml O_2 /100 ml/min)	0.32 $\pm$ 0.17	2.77 $\pm$ 0.24
CMRGlu		
(mg/100 ml/min)	2.35 $\pm$ 0.55	4.43 $\pm$ 0.99
MR (ml O_2 /mg glucose)	0.15 $\pm$ 0.08	0.66 $\pm$ 0.20
($\mu\text{mol O}_2/\mu\text{mol}$ glucose)	1.20 $\pm$ 0.64	5.29 $\pm$ 1.60

GER, glucose extraction ratio

Results

A similar pattern of disordered cerebral metabolism was observed in all five patients. Blood flow in the infarcted regions was reduced to a variable extent, but in all cases was sufficient to meet residual oxygen demands, since CMRO_2 was always profoundly depressed and OER low. However, glucose metabolism was only moderately reduced in the regions of infarction as compared to contralateral cerebral cortex, and CMRGlu was invariably preserved in relation to CMRO_2 within the infarct. This relationship can be expressed as a metabolic ratio (MR) $\text{CMRO}_2/\text{CMRGlu}$, the value of which was consistently lower within the infarcted area than in the relatively normal contralateral cortex. Quantitation of these findings is summarised in Table 1.

Discussion

The finding of a low MR – reflecting partial preservation of glucose consumption in relation to the marked fall of CMRO_2 – suggests

that in all five patients there was persistent anaerobic glycolysis in the region of cerebral infarction. This might be predicted in the acute phase of ischaemia, since it is known from animal stroke models that there may be continued and prolonged elevation of lactate levels after restoration of blood flow if this takes place beyond the critical period during which complete recovery of function can occur (Kleihues et al. 1975). This persistent lactic acidosis has been attributed to survival of glycolytic pathways in the cytoplasm at a time when the more vulnerable mitochondrial systems have been irreversibly damaged (Siesjö 1980). However, such a mechanism cannot account for sustained anaerobic glycolysis several days or even weeks after cerebral infarction has occurred. Histopathological studies reveal neutrophil infiltration within 24–48 h of acute infarction, and after 4 or 5 days the accumulation of increasing numbers of macrophages, which may remain the predominant cell population of an infarct for many weeks. It is therefore possible that the observed anaerobic glucose metabolism in lesions more than a few days old may be that of macrophages rather than residual cerebral tissue. There also remains the possibility that without appropriate correction for alteration of the rate constants in ischaemic brain (Hawkins et al. 1981), application of the deoxyglucose model may lead to overestimation of glucose metabolism within the region of a cerebral infarct. However such an error would have to be substantial to account for the marked regional reduction of MR observed in our patients with stroke.

References

1. Ackerman RH, Correia JA, Alpert NM, Baron J-C, Gouliamos A, Grotta JC, Brownell GL, Taveras JM (1981) Positron imaging in ischemic stroke disease using compounds labeled with oxygen 15. *Arch Neurol* 38: 537–543
2. Baron JC, Boussier MG, Comar D, Soussaline F, Castaigne P (1981) Noninvasive tomographic study of cerebral blood flow and oxygen metabolism in vivo. Potentials, limitations, and clinical applications in cerebral ischemic disorders. *Eur Neurol* 20: 273–284
3. Hawkins RA, Phelps ME, Huang S-C, Kuhl DE (1981) Effect of ischemia on quantification of local cerebral glucose metabolic rate in man. *J Cereb Blood Flow Metabol* 1: 37–51
4. Huang S-C, Phelps ME, Hoffman EJ, Sideris K, Selin CJ, Kuhl DE (1980) Noninvasive determination of local cerebral metabolic rate of glucose in man. *Am J Physiol* 238: E 69–E 82
5. Kleihues P, Hossmann K-A, Pegg AE, Kobayashi K, Zimmermann V (1975) Resuscitation of the monkey brain after one hour complete ischemia. III. Indications of metabolic recovery. *Brain Res* 95: 61–73
6. Lammertsma AA, Jones T, Frackowiak RSJ, Lenzi G-L (1981) A theoretical study of the steady-state model for measuring regional cerebral blood flow and oxygen utilisation using oxygen-15. *J Comput Assist Tomogr* 5: 544–550
7. Lenzi G-L, Frackowiak RSJ, Jones T (1981) Regional cerebral blood flow (CBF) oxygen utilization (CMRO₂) and oxygen extraction ratio (OER) in acute hemispheric stroke. *J Cereb Blood Flow Metabol* 1 (Suppl. 1): S504–S505
8. Phelps ME, Huang S-C, Hoffman EJ, Kuhl DE (1979) Validation of tomographic measurement of cerebral blood volume with C-11 labeled carboxyhemoglobin. *J Nucl Med* 20: 328–334
9. Siesjö B (1980) Brain energy metabolism. Wiley, New York, p 512
10. Sokoloff L, Reivich M, Kennedy C, Des Rosiers MH, Patlak CS, Pettigrew KD, Sakurada O, Shinohara M (1977) The [¹⁴C] deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure, and normal values in the conscious and anesthetized albino rat. *J Neurochem* 28: 897–916

Subject Index

- accelerator 53
 - , linac 53
 - , Van de Graaff 53
- accuracy 48
- activating drugs 162, 167
- acute hyperemia 117
- adenylyl cyclase 72
- α -adrenergic receptors 73
- aerobic metabolism 205
- air curve 91
- alanine 66
- alkyl monoamines, ^{11}C 60
- Alzheimer's dementia 208, 210
 - disease 128
- amino acid 59, 66
 - –, ^{11}C 65, 66
 - –, enzymatic 66
 - –, ^3H 19, 229
 - –, ^{13}N 65
 - – pool 14
 - –, synthesis 66
- aminoacyl tRNA 68
- ammonia, ^{13}N 66
- anaerobic glycolysis 111, 130, 234
 - –, glioma 111
 - –, stroke 111
 - metabolism 205
- antagonist ligands, 72
- anticonvulsants 212
- anxiety 212
- aphasia 131
- (–)-apomorphine 71
- arterial blood samples 109
 - samples, ^{18}F -FDG 114
 - sampling 108
- aspartic acid, ^{11}C 59, 66
- astrocytoma 181
 - , grade I 182
 - , grade II 182
 - , grade III 182
 - , grade IV 182, 185
- asymmetry 155, 203
- ATP 20
 - content, regional demonstration 22
- auditory stimulation 140
 - –, LCMRG1 145, 154
- autoradiography 3
 - , brain tumors 20
 - , cerebral blood flow 19
 - , double-tracer technique 232
 - , glucose metabolism 19
 - , image-processing 13
 - , protein synthesis 15, 19
 - , resolution 13
 - , stroke-prone spontaneously hypertensive rats 20
 - , triple-labeled, advantages 27
 - –, quantitative 19
 - –, restrictions 27
- benzodiazepine 227
 - , receptor 218, 227
- BGO detectors 33, 40
 - –, physical parameters 33
- biochemical behavior of ^{14}C -deoxyglucose and glucose 8
 - parameters 65
- bismuth germanate (BGO) 33, 40
- blood brain barrier 93
 - flow 104
 - –, stroke 113
 - samples, arterial 109
 - sampling 109
- bolus injection, ^{15}O -oxygen 104
 - –, ^{15}O -water 104
- brain activity, ^{11}C -DPH 214
 - blood partition coefficient 79
 - tumors 25
- breathing system 108
- bromine-75 226, 227
 - , production methods 226
 - , radiopharmaceuticals 226
- (+)-butaclamol 70, 71
- butyrophenones 71
- carbon-11 54
 - , monoxide 234
 - , yields 55
- carboxyl amino acids, ^{14}C 229
- carotid artery occlusion 116
- ^{11}C 52, 53, 57, 58, 65
- ^{11}C -alkyl monoamines 60
- ^{11}C -amino acid 65, 66
- ^{11}C -aspartic acid 59, 66
- CBF 78, 114, 120, 234
 - /CMRO₂ relationship 120, 123
 - measurements, fluoromethane 90
 - threshold for ischemia 124
- ^{11}C -carbon monoxide 234
- ^{14}C -carboxyl amino acids 229
- ^{11}C -compounds 56

- ¹⁴C-concentrations 8
- ¹¹C-deoxyglucose (¹¹C-DG) 52, 65, 153, 155
- ¹⁴C-deoxyglucose 3, 19, 65, 232
 - , autoradiogram 156
 - , half-life 10
 - , method 9
 - –, error 9
 - –, limitations 9
 - –, operational equation 4
 - , plasma concentration curves 10
 - , transport 10
- ¹¹C-deoxy sugar 58
- ¹¹C-DG, repeat measurement of LCMRGI 155
- ¹¹C-diphenylhydantoin 212
- ¹¹C-DPH 212
 - , brain images 216
 - –, uptake 217
 - , concentration 215
 - , gray matter 215
 - , kinetics 217
 - , white matter 215
- cerebellar diaschisis 118
 - hypometabolism 158
- cerebral activation 200
 - –, procedure 196
 - blood flow 77, 78, 104
 - –, autoradiography 19
 - –, quantitation 108
 - –, quantitative evaluation 21
 - –, stroke 113
 - –, volume 210
 - –, dementia 210
 - glucose metabolic rate, effect of medications 189
 - metabolism, auditory and visual sensory deprivation 139
 - –, lesions of the visual pathway 139
 - –, nonverbal auditory stimuli 139
 - –, verbal auditory stimuli 139
 - –, visual stimulation 139
 - metabolic activity, normal populations 204
 - –, psychiatric populations 204
 - oxygen consumption 104
 - utilization 104
- cerebrovascular disease, LCBF 129
 - –, LCMRGI 129
- ¹¹C-flunitrazepam 59, 227
- ¹¹C-galactose 57
- ¹¹C-glucose 45, 52, 57, 65, 192
- ¹⁴C-glucose 11
- CH₃F 90
- chorea Huntington, asymptomatic carriers 133
 - –, CMRGI 132
- chronic ischaemia, rCMRO₂ 110
 - –, rOER 110
- ¹¹C-iodoantipyrine 88
- ¹⁴C-iodoantipyrine 11
- ¹¹C-leucine 59, 66, 68
- ¹⁴C-leucine 59
- clinical availability, PET 136
 - –, SPET 136
- ¹¹C-L-methionine 208
- ¹¹C-methyl-glucose 169
- CMG 169
 - distribution, infarction 171
 - –, normal brain 171
 - kinetics 173
- CMRGI 114, 234
 - , controls 203
 - , depression 203
 - , epilepsy 133
 - , epileptogenic focus 133
 - , Huntington's chorea 132
 - , mania 203
 - , nonfocal seizures 136
 - , partial epilepsy 133
 - , petit mal seizures 136
 - , schizophrenia 203
 - , sensory deprivation 141
- CMRO₂ 114, 120, 234
 - , threshold for infarction 123
- ¹¹C-norleucine 68
- ¹¹CO 56
 - labeled red cells 108
- CO₂ 113
 - , inhalation 107
- ¹¹CO₂ 56
- C¹⁵O₂ 80, 82, 95, 234
 - , radiation dose 80
- cold injury 23
- collimator 49
- combined instrument schedule 201
- compartmental model 91
- compounds, ¹¹C 56
- computer 42
- concentrations, ¹⁴C 8
- continuous inhalation, carbon dioxide 104
 - –, C¹⁵O₂ 107
 - –, ¹⁵O₂ 107
 - –, ¹⁵O-CO₂ 114
 - –, oxygen 104
- convolution integral equation 79
- cortical dementia 132
- coupling of CBF and CMRO₂ 234
 - of flow, glucose metabolism and protein synthesis 22
- ¹¹C-palmitic acid 57
- ¹¹C-pimozide 59, 212, 217
 - , binding to striatal DA receptors 222
 - , brain images 218
 - concentration, cerebellum 221
 - –, blood 220
 - –, caudate 221
 - , kinetics 221
- ¹¹C, positron emitters 52, 54
- ¹¹C-quinuclidinyl benzilate methiodide 59
- cross-contamination 26
 - slices 43
- crossed cerebellar atrophy 167
- Crow's classification 194
- CsF 31
 - , attenuation coefficient 31

- detectors, physical parameters 33
- , refractive index 31
- , resolution limits 32
- ^{11}C -spiroperidol 58
- cyclotron 53, 54
- corporation 54

- DA receptors** 217
- dead time correction 45
 - – losses 43
- degenerative dementia 109
 - disorders 212
- delayed hyperemia 117
- dementia, methionine metabolism 210
 - , protein metabolism 208
 - , – synthesis 211
 - , rCBF 109
 - , rCMRO₂ 109
- 2-deoxy-2-bromo-D-glucose 226
- deoxyglucose 57, 65
 - , ^{11}C 52, 65, 153, 155
 - , ^{14}C 3, 4, 9, 10, 19, 65, 156, 232
 - , ^{18}F 52, 56, 65, 113, 201
 - method 3
 - –, principle 7
 - , technique 2, 3
- 2-deoxyglucose-6-phosphate 3
- depression 202, 212
 - of glucose metabolism 165
 - of metabolism 155
- design concepts, performances 46
 - parameters 33
- detector motion 46
 - –, rotation 46
 - –, wobble 46
 - ring 46
- determination, quantitative 72
- diaschisis 155, 167
- 3,4-dihydroxyphenylalanine 58
- diphenylhydantoin, ^{11}C 212, 215, 216, 217
- displacement studies 222
- distribution of ^{11}C -DPH 213
 - volumes 101
- dopamine 58, 69, 71
 - agonists 72
 - receptor 70, 72
 - –, topography 71
- dopaminergic antagonist 217
 - ligands 222
 - receptors 217, 221
- double integral method 79
 - isotope incorporation procedure 229
- DPH concentration within the epileptogenic focus 217
- drugs, activating 162
- dynamic FDG studies 43
 - imaging 78
 - plus equilibrium imaging 81
 - positron emission tomography (dPET) 169
 - scanning 164, 166
 - study 46

- ECAT II 91, 95, 107, 108, 128, 139, 169, 182, 197, 208, 213
- EDTA, ^{68}Ga 79
- efficiency, coincidence 34
 - , geometric 34
 - , intrinsic 34
- energy metabolism, schizophrenia 192
- epilepsy 133
 - , focal 212
 - , partial 212
 - , rCBF 109
 - , rCMRO₂ 110
- equilibration of ^{11}C -DPH 215
- equilibrium concentration of H_2 ^{15}O 107
 - , imaging 80
 - –, limitations 81
- experimental procedure 8
- expired air curve 91
- extra-intracranial bypass operation 178
 - – – surgery 179

- false coincidences 48
- ^{18}F 53, 57, 58
 - , compounds 56
- ^{18}F -deoxyglucose (^{18}F FDG) 52, 56, 65, 113, 201
 - , synthesis 58
 - uptake, abnormal brain 111
- ^{18}F -FDG 57, 65, 111, 114
 - , arterial samples 114
 - , technique 153
- ^{18}F -fluorodeoxyglucose (^{18}F -FDG) 57, 58, 65, 111, 114, 139, 153, 162, 196, 234
 - , aging 128
 - , cerebral dysfunction 128
 - , cerebrovascular disorder 128
 - , degenerative disorder 128
 - , epileptic disorders 128
 - , method 153
 - , technique 13, 153
- ^{18}F -3-fluoro-3-deoxy-D-glucose 58
- ^{18}F -fluoromethane 80
- ^{18}F -5-fluorouridine 60
- ^{18}F -haloperidol 58, 72
- Fick principle 85, 105
- flunitrazepam, ^{11}C 59, 227
- fluorine-18 54
 - , yields 55
- 3-fluoro-3-deoxyglucose 90
- fluoromethane 90
 - , ^{18}F 80
 - , synthesis 91
- fluorophenylalanine 68
- fluorouridine, ^{18}F -5- 60
- focal epilepsy 212
- four-ring positron camera 40
 - – emission tomography 193

- ¹⁸F-p-fluorophenylalanine 68
- ¹⁸F, positron emitters 52, 54
- , production 55
- frontal hypermetabolism 142, 149
- /posterior correlations 197
- ¹⁸F-spiroperidol 58, 73
- functional asymmetries 149
- images 87
- inactivation 167
- FWHM 30, 43, 47, 48, 49, 50, 139, 162, 182
- ⁶⁸GaCl₃-labeled transferrin 208
- ⁶⁸Ga-EDTA 79
- galactose, ¹¹C 57
- gallium-67 60
- general principles 2
- gerbils 232
- glioblastoma multiforme 181
- gliomas, glucose utilization 111
- , – rate 185
- , glycolytic rate 181
- , high-grade (astrocytomas III) 183, 184
- , histologic grade 181
- , low-grade (astrocytomas I and II) 183, 184
- , oxygen utilization 111
- , rCBF 110
- , rCMRO₂ 110
- global metabolic rate, depressed 203
- – –, manic 203
- – –, normal 203
- – –, schizophrenic 203
- glucose, availability 232, 233
- , bioluminescent evaluation 22
- , ¹¹C 45, 52, 57, 65, 192
- , ¹⁴C 11
- carrier 175
- – system 176
- consumption 232
- efflux 175
- influx 175
- – rate 173
- – –, normal 176
- metabolic rate 183
- metabolism 127 ff, 162
- –, acute stroke 234
- –, autoradiography 19
- –, cognitive impairment 202
- –, normal 139, 201
- –, primary affective disorders 201
- –, remote effects 167
- –, schizophrenia 201
- –, senile dementia 201
- –, stroke 113
- , phosphorylation 7
- , plasma concentration curves 30
- rates, normal tissue 183
- uptake 169
- utilization and histologic grade 190
- –, mild pain stimulation 196
- –, normal 197
- –, normal subjects 196
- –, quantitative evaluation 21
- – rates, correlation between brain regions, in normals and in schizophrenics 199
- – –, gliomas 185
- –, schizophrenia 196, 197
- glucose-6-phosphatase 12
- , effects in human studies 13
- glucose-6-phosphate 3
- glycolysis, gliomas 181
- glycolytic pathways 234
- rate 190
- half-field stimulations 149
- haloperidol 218
- ³H-amino acids 19, 229
- head positioning device 193
- restraint system 109
- HEADTOME 46
- healthy volunteers 169
- hemianopsia 149
- hemorrhage 117
- hemorrhagic stroke 113
- hexokinase-catalized phosphorylation 3
- HgI₂ 36
- HgI₂-BGO hybrid detector 37
- HgI₂ detector 36
- high-grade gliomas (astrocytomas III) 184
- H₂¹⁸O 56, 80, 95, 105
- , local cerebral blood flow 100
- , radiowater volume 100
- homonomous hemianopsia 116
- ³H-pimozide 73, 217
- Huntington's chorea 70
- hyperemia 117, 158
- hyperfusion 118
- hyperglycemia, rate constants 11
- hypermetabolism, propagated neuronal activity 134
- , seizure origin 134
- hypofrontality 194, 204
- hypometabolism, cerebellar hemisphere 158
- , contralateral cerebellar hemisphere 187
- hypovascularity, in tumors 189
- implementation of SAT 35
- inactivated brain structures, LCMRGI 164
- inert gas exchange 104
- infarct, LCMRGI 130
- infarcted cortex, LCMRGI 164
- infarction, OEF 117
- inhalation, CO₂ 107
- , ¹⁵O-oxygen 105
- ¹²³I, N-isopropyl-p-¹²³I-iodoamphetamine (IMP) 136
- in-plane slices 43
- instrumentation 30
- integrated specific activity of precursor pool 5

- intravenous bolus injection 87
 - injection, ^{15}O -water 105
- inverse metabolic relation 199
- in vivo autoradiography 85
- ^{131}I -iodoantipyrine 19, 232
- iodine-123 60
- iodoantipyrine, ^{11}C 88
 - , ^{14}C 11
- irreversible cerebral infarction 234
- ischemia 88, 110, 232
 - , rate constants 11
- ischemic infarct, LCMRGi 164
 - –, local cerebral blood flow 120
 - –, oxygen consumption 120
 - stroke 113, 164, 169
- (+)-isobutacramol 71
- isotope separation 22

- Japan Steel Works 54

- k^*_1 , k^*_2 , k^*_3 (rate constants) 5
- kinetic constants 3
- “kinking” of the internal carotid artery 179
- ^{77}Kr 79, 82
 - bolus inhalation method 79
 - , radiation dose 80
 - , technique 78

- labeled compounds 52, 56
- lactic acidosis 234
- LCBF 120
 - , cerebrovascular disease 129
 - , early infarcts 121
 - , infarction 121
 - , late infarcts 121
 - , stroke 120, 129
- LCMRGi 153, 162
 - , associative visual cortex 145
 - , auditory stimulation 145, 154
 - , calculation 162
 - , caudate 132
 - , cerebrovascular disease 129
 - , hemianopic subjects 142
 - , inactivated brain structures 164
 - , infarct 130
 - , infarcted cortex 164
 - , ischemic infarcts 164
 - , lesions of the visual pathway 142
 - , normal values 128, 154, 163
 - , piracetam 167
 - , primary visual cortex 145
 - , putamen 132
 - , stroke 129
 - , visual stimulation 154
 - , LCMRO₂ 120
 - , infarction 121
 - , stroke 120
- lesions of visual pathway 140
- leucine, ^{11}C 59, 66, 68
 - , ^{14}C 14, 59
 - , ^{13}N 68
- ligand-receptor binding 70
- line spread function 43
- local cerebral blood flow 120, 121, 129
 - – –, ischemic infarction 120
 - – – volume 105
 - – glucose consumption, stroke 155
 - – – metabolism, stroke 153
 - – – utilization (LCMRGi) 128
 - – – –, cerebrovascular disorders 128
 - – – –, degenerative disorder 128
 - – – –, epileptic disorder 128
 - – – –, monkey 12
 - – – –, normal aging 128
 - – – –, rat 12
 - fractional extraction of oxygen (LOEF) 105
 - perfusion rate (LPR) 169, 175, 176
 - unidirectional glucose transport rate (LUGTR) 169, 175, 176
- LOEF, early infarcts 121
 - , infarction 121
 - , late infarcts 121
- low-grade gliomas (astrocytomas I and II) 184
- LPR 175
 - , infarction 176
 - , normal persons 176
- LUGTR 169, 175
 - , infarction 176
 - , normal persons 176
- lumped constant 5, 11, 233
 - –, cat 12
 - –, dog 12
 - –, normal tissue 11
 - –, rat 11, 12
 - –, rhesus monkey 11, 12
 - –, for tumors 188
- luxury perfusion 27, 123
 - –, syndrome 117, 120, 158
- manic-depressive psychosis 202
- “matched” CBF – CMRO₂ 120
- mathematical integration 96
- mercuric iodide 36
- messenger RNA 68

- metabolic activity, normal 203
 - –, schizophrenic 203
 - asymmetries 147, 149
 - heterogeneity 203
 - hyperfrontality 128
 - pattern, schizophrenia 192
 - rate, calculation 182
 - –, monozygotic twins 204
 - ratio 234
 - –, frontal to nonfrontal 203
 - symmetry 128
- methionine 66

- methionine, ^{11}C 208
 - metabolism, dementia 210
 - –, three-compartment model 208
 - , partition coefficient 209
- method, ^{18}F -FDG 153
- methodology of PET 29ff
- methyl- ^{11}C -methionine 59
- methyl-fluoride 90
- methyl-glucose, ^{11}C 169
- 1-methyl-3,4,6-triacetyl-2-deoxy-2-bromo-D-glucoside 225
- MGH positron camera 114
- middle cerebral artery occlusion, CBF 115
 - – – –, CMRO₂ 115
 - – – –, OEF 115
 - – – –, rCMRO₂ 110
 - – – –, rOER 110
- misery perfusion 123
 - –, syndrome 120
- moya-moya disease 177, 179
- multiparametric methods 27, 28
- multiple infarct dementia 109, 128, 131
- multiring hybrid emission tomograph, design concept 46
 - – – –, performances 46
 - positron camera 40
 - –, electronic system 40
- multislice PET imaging system 88
- muscarinic receptors 218

- NADH 20
 - , fluorescence 22
- NaI detectors 46
- ^{13}N 52, 53, 56, 65
- ^{13}N -amino acids 65
- ^{13}N ammonia 66
- necrosis 120
- neuroECAT 33
- neuroleptics 212
 - , receptors 217
 - , treatment 194
- neurotransmitter receptors 58
- $^{13}\text{NH}_3$ 56, 129
- nitrogen-13 54
- ^{13}N -leucine 68
- $^{13}\text{N}_2\text{O}$ 80
- N_2^{15}O 82
 - , radiation dose 80
- nonequilibrium methods, blood flow 105
 - –, local cerebral oxygen utilization 105
- nonfocal seizures, CMRGl 136
- nonnutritional flow 118
- nonverbal stimulation 141, 146
- no-reflow phenomenon 124
- norleucine, ^{11}C 68
- normal protein synthesis 211
 - subjects, LCMRGl 163
- ^{13}N , positron emitters 52
- nuclear reaction 54

- nucleosides 60
- nucleotides 60
- numerical integration 97

- ^{15}O 52, 53, 56, 65, 114
- $^{15}\text{O}_2$ 113, 234
- ^{15}O -carbon monoxide 105
- ^{15}O -CO₂ 56, 114
- ^{15}O continuous inhalation technique 120
- ocular dominance columns 15
- OEF 114
 - , in infarction 117
- OER 234
- ^{15}O -H₂O 114
- O_2 , inhalation 107
- ^{15}O -labeled water 95, 104
- oligodendroglioma 181
- O^{15}O 105
- ^{15}O -O₂ 114
- operational equation of ^{14}C -deoxyglucose method 4
- ^{15}O , positron emitters 52, 54
- Ostwald solubility coefficient 90, 92
 - – –, fluoromethane 92
 - – –, Xe 92
- ^{15}O -water 104
- oxygen-15 54, 113
 - consumption 103ff
 - –, ischemic infarction 120
 - extraction fraction (OEF) 114, 120
 - – ratio 108
 - metabolism, stroke 113, 234
 - utilization 104, 107
 - –, quantitation 108

- palmitic acid 57
- Parkinson's disease 70
- partial epilepsy 133, 212
 - volume artifact 183
 - – effect 149, 187
- partition coefficient, methionine 209
- PC 384-B (four-ring positron camera) 40
- PC-II 114
 - , FWHM 114
 - , plane thickness 114
- perfusion 169
- petit male seizures, CMRGl 136
- PETT II 155
 - III 202
- phantom 43, 49
- pharmacokinetics 212
 - , labeled drugs 212
- phenethylamines 70
- phosphatase activity, tumors 189
- phosphorylation, hexokinase-catalyzed 3
- photodetector 36
- photoiode 36
- pimozide, ^{11}C 59, 212, 217, 218, 220, 221, 222
- , ^3H 73, 217

- piperazine 72
- piracetam 162, 166
- plasma flow 174
 - ± – rate 174
- point spread function 43
- polyamines 60
- positome III 79
- positron camera PC 384-7 B 162
 - computed tomography 139
 - emission tomography, design concepts 30
 - emitters, carbon-11 52, 54
 - –, fluorine-18, 52, 54
 - –, nitrogen-13 52, 54
 - –, oxygen-15 52, 54
- precursor 2
 - pool, protein synthesis 231
- principles of metabolic determinations 1ff
- production methods, ^{75}Br 226
- proline 66
- protein biosynthesis 67
 - metabolism, dementia 208
 - –, schizophrenia 208
 - synthesis 14, 59, 67, 229
 - –, autoradiography 15, 19, 230
 - –, dementia 211
 - –, dynamic model 208
 - –, fronto-occipital ratio 211
 - –, hypoglossal nucleus 15
 - –, metabolic pathways 69
 - –, models 15
 - –, monocular deprivation 15
 - –, normal 211
 - –, precursor pool 231
 - –, quantitative determination 229
 - –, – evaluation 21
 - –, rates 210
 - – and receptor studies 207ff
 - –, schizophrenia 211
 - –, visual system 15
- PT, functional anatomy 139
- purine 60
- pyrimidines 60

- quantitation, cerebral blood flow 108
 - , – – errors 108
 - , – – method 108
 - , oxygen utilization 108
 - , statistical uncertainty 108
 - , theoretical aspects 108
- quantitative autoradiograms 104
 - determination, protein synthesis 229
- quinuclidinyl benzilate methiodide, ^{11}C 59

- radiation dose, C^{15}O_2 80
 - –, ^{77}Kr 80
 - –, N_2^{15}O 80
- radiochemicals 51ff
- radiolabeled amino acid 67
- radiopharmaceuticals 56, 226
 - , ^{75}Br 226
 - , snythesis 65
- radiowater, input functions 101
 - , volume 95
- ramp infusion technique 87
- rate constants 7, 9, 166, 175, 193
 - –, glucose influx 175
 - –, hyperglycemia 11
 - –, ischemia 11
 - –, monkey 7
 - –, rat 7, 8
 - –, for tumors 188
- rate of glucose utilization, high-grade tumor 183
 - – – –, low-grade tumor 183
 - – – –, per unit mass of tissue 5
 - of local glucose utilization, maps 13
- ratio of CMRGI 193
 - , frontal to nonfrontal 203
- rCBF 85, 86, 95, 99, 108, 111, 115
 - , dementia 109
 - , dynamic imaging 78
 - , – plus equilibrium imaging 78
 - , epilepsy 109
 - , episodic aphasia 93
 - , equilibrium imaging 78
 - , glioma 110
 - , infarction 93
 - measurement 78
 - , schizophrenia 110
 - , stroke 110
 - , tumor 93
 - values, clinical results 80
 - –, normal subjects 80
 - , visual stimulation 93
- rCBV 111
- rCMRGI 111, 234
 - , dementia 204
- rCMRO₂ 108, 111, 115
 - , dementia 109
 - , epilepsy 110
 - , glioma 110
 - , schizophrenia 110
 - , stroke 110
- receptor 212
 - ligands 58
- regional cerebral blood flow 78, 85, 90, 95, 107
 - – – –, acute stroke 234
 - – – volume 108
 - glucose metabolism, normal 196, 201
 - – –, psychiatric 201
 - – –, schizophrenics 196
 - ischaemia 110
- remote functional depression 162
 - metabolic depression 167
- resolution 30
 - , detection system 32
 - , spatial 43, 104
 - , temporal 104
 - , time of flight 31

- reversible ischaemia 234
- RNA 229
- rOER, stroke 110
- samples, arterial 114
 - Scanditronix 54
 - schizophrenia 70, 208, 210, 212, 219
 - , energy metabolism 192
 - , metabolic pattern 192
 - , protein metabolism 208
 - , – synthesis 211
 - , rCBF 110
 - , rCMRO₂ 110
 - senile dementia 132, 202
 - sensitivity 45, 48, 50
 - , BGO detectors 34
 - , CsF detectors 34
 - sensory deprivation 139, 140
 - –, CMRGl 141
 - stimulation 139
 - serotonergic receptors 217
 - serotonine receptors 73
 - signal amplification technique 32
 - to noise ratio 30
 - single photon emission 46
 - single-transit extraction 100
 - slice thickness 43, 48, 50
 - solubility coefficient 92
 - of oxygen 105
 - spatial resolution 43, 48, 50, 104
 - specific binding 217
 - spiperidol 58, 71
 - , ¹¹C 58
 - , ¹⁸F 58, 73
 - , binding 72
 - spiperone 217, 222
 - steady-state, O₂ consumption technique 2
 - , oxygen-15 inhalation method 107
 - stimulation, nonverbal 146
 - , verbal 146
 - stroke 113, 115, 162, 176
 - , blood flow 113
 - , cerebral blood flow 113, 115
 - , CMRO₂ 115
 - , glucose metabolism 113, 234
 - , hemorrhagic 113
 - ischemic 113
 - , LCBF 120, 129
 - , LCMRGl 129
 - , LCMRO₂ 120
 - , local cerebral glucose consumption 155
 - , – – metabolism 153
 - , OEF 115
 - , oxygen metabolism 113, 234
 - prone spontaneously hypertensive rats 25
 - , rCBF 110, 234
 - , rCMRO₂ 110
 - rOER 110
 - syndrome, differential diagnosis 118
 - structured clinical interview 201
 - subcortical dementia 132
 - suppression of glucose utilization 189
 - targetry precursors 55
 - temporal hypermetabolism 150
 - lobectomies 134
 - resolution 104
 - thallium-201 60
 - timbre stimulation, LCMRGl 146
 - test 141
 - time of flight 31
 - – –, resolution 31
 - resolution 31, 47
 - tissue attenuation correction 108
 - /blood partition coefficient 91
 - tomographic mappings of rCBF 83
 - tomography, FWHM resolution 30
 - tonal memory stimulation 145
 - – test 141
 - transcallosal depression 167
 - transfer ribonucleic acids 67, 229
 - transferrin, ⁶⁸GaCl₃ 208
 - transient ischemia, LPR 176
 - –, LUGTR 176
 - transmission scan 48, 108
 - transneural depression 158
 - tryptophan 59
 - tumor grade and glycolytic activity 190
 - uniformity 50
 - verbal stimulation 146
 - –, LCMRGl 145
 - visual cortex, associative 142
 - –, primary 142
 - stimulation 140, 142
 - –, LCMRGl 154
 - water clearance 104
 - volume 95, 99
 - wobbling technique 40
 - yields, carbon-11 55
 - , fluorine-18 55

Experimental Brain Research

Each of these supplements is available to subscribers of the journal "Experimental Brain Research" at 20% off the list price.



Springer-Verlag
Berlin
Heidelberg
New York

Supplement 1: **Afferent and Intrinsic Organization of Laminated Structures in the Brain**

(7th International Neurobiology Meeting)

Editor: O. Creutzfeldt

1976. 127 figures. XXIII, 579 pages

DM 64,-. ISBN 3-540-07923-8

Supplement 2: **Hearing Mechanisms and Speech**

EBBS-Workshop, Göttingen, April 26-28, 1979

Editors: O. Creutzfeldt, H. Scheich, C. Schreiner

1979. 85 figures, 12 tables. XXIII, 413 pages

DM 69,-. ISBN 3-540-09655-8

Supplement 3: **Gonadal Steroids and Brain Function**

IUPS-Satellite-Symposium, Berlin, July 10-11, 1980

Editors: W. Wuttke, R. Horowski

1981. 136 figures, 10 tables. XIII, 373 pages

Cloth DM 92,-. ISBN 3-540-10606-5

Supplement 4: **The Renin Angiotensin System in the Brain**

A Model for the Synthesis of Peptides in the Brain

Editors: D. Ganten, M. Printz, M. I. Phillips, B. A. Schölkens

1982. 108 figures, 46 tables. XVII, 385 pages

Cloth DM 98,-. ISBN 3-540-11344-4

Supplement 5: **The Aging Brain**

Physiological and Pathophysiological Aspects

Editor: S. Hoyer

1982. 52 figures, 66 tables. XIV, 281 pages

Cloth DM 98,-. ISBN 3-540-11394-0

Supplement 6: **The Cerebellum - New Vistas**

Editors: S. L. Palay, V. Chan-Palay

1982. 264 figures, 16 tables. XVII, 637 pages

Cloth DM 198,-. ISBN 3-540-11472-6

Supplement 7: **Neural Coding of Motor Performance**

Editors: J. Massion, J. Paillard, W. Schultz, M. Wiesendanger

1983. 88 figures, 6 tables. Approx. 370 pages

Cloth DM 98,-. ISBN 3-540-12140-4

Current Topics in Neuroendocrinology

Editors: **D. Ganten, D. Pfaff**

The central nervous system and the endocrine system comprise quickly expanding fields at the forefront of medical and biological research. Neuroendocrinology is pursued so intensely that it has become impossible to keep up-to-date on all current research activity. **Current Topics in Neuroendocrinology** helps overcome this problem by treating not only those topics which are exciting because of their topicality, but also those which are reliable, where enough work has been done to treat them from several viewpoints.

Volume 1

Sleep

Clinical and Experimental Aspects

With contributions by numerous experts

1982. 47 figures. VII, 129 pages
ISBN 3-540-11125-5

Volume 1 covers mechanisms of sleep from a neurochemical, endocrine and clinical point of view. In five chapters, in-depth discussions treat neurotransmitters, pharmacologic interference, endocrine and peptidergic functions, a characterization of sleep with regard to hemodynamics, single unit recording and EEG activity, as well as biologic rhythms and sleep characteristics in normal and psychotic patients.



Springer-Verlag
Berlin
Heidelberg
New York

Volume 2

Adrenal Actions on Brain

With contributions by numerous experts

1982. 25 figures. V, 153 pages
ISBN 3-540-11126-3

This volume covers the relations between the adrenal glands and the brain, with emphasis on the regulatory effects of the glucocorticoids. The articles it contains review recent work on hormone receptors and their developmental, endocrinological, electrophysiological and behavioral effects on the central nervous system.
