Distribution Category:
Biology and Medicine
(UC-48)

ANL--83-40

DE83 015756

ARGONNE NATIONAL LABORATORY
9700 South Cass Avenue
Argonne, Illinois 60439

DIVISION OF BIOLOGICAL AND MEDICAL RESEARCH
ANNUAL TECHNICAL REPORT
1982

E. Huberman, Division Director
M. W. Rosenthal, Editor

May 1983

Preceding Report
ANL-82-35, June 1982

COVER

Computer graphic representation of the antigen-binding sites of two Bence-Jones proteins (antibody light chain dimers), Loc (left) and Mcg (right). The spheres represent individual amino acids. Each binding site is composed of two variable domains and each domain consists of framework segments and three hypervariable segments which confer the specificity to the antibody molecule. The three hypervariable segments are distinguished here by different shadings added to the computer printout. The open-appearing antigen binding site of protein Mcg represents the appearance of the binding sites of all antigen-binding fragments studied thus far. The binding site of protein Loc appears to be a new configuration. Prepared by Dr. Chong-Hwan Chang. (See Chapter 11.)

DISCLAIMER

This report was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

ABSTRACT

This report summarizes research during 1982 in the Division of Biological and Medical Research, Argonne National Laboratory. Studies in Carcinogenesis address mechanisms of chemical and radiation carcinogenesis including the processes of tumor initiation and promotion. The studies employ rat liver and mouse skin models as well as human rodent cell culture systems. The use of liposomes for metal mobilization is also explored. Low Level Radiation studies include delineation of the hematopoietic and other responses of dogs to continuous low level gamma irradiation, comparison of lifetime effects in mice of low level neutron and gamma irradiation, and study of the genetic effects of high LET radiations. Molecular Biology research develops two-dimensional electrophoresis systems for diagnosis and detection of cancer and other disease. Fundamental structural and biophysical investigations of immunoglobulins and other key proteins are included, as are studies of cell growth, and of molecular and cellular effects of solar UV light. Research in Toxicology uses cellular, physiological, whole animal, and chronobiological end points and chemical separations to elucidate mechanisms and evaluate hazards of coal conversion by-products, actinides, and toxic metals. The final sections cover support facilities, educational activities, seminars, staff talks, staff, and funding agencies.

TABLE OF CONTENTS

1. Division Director's Introduction
E. Huberman.....1

CARCINOGENESIS

2. Mechanisms of Hepatocarcinogenesis
C. Peraino, W. E. Boernke, J. E. Morris,
B. A. Carnes, J. A. Blomquist, V. A. Ludeman,
A. M. Prapuolenis, and E. F. Staffeldt.....7

3. Mutagenesis and Cell Differentiation in Chemical Carcinogenesis
E. Huberman, C. A. Jones, E. A. Malvoisin,
S.-I. Murao, B. Ryffel, I. Simon,
M. Stodolsky, M. F. Callaham,
C. B. Henning, and B. A. Sedita.....11

4. Radiation-Induced Changes in Mammalian Cells
A. Han, C. K. Hill, R. L. Wells, M. M. Elkind,
M. P. Hagan, D. L. Redman, V. N. Smith,
E. M. Buess, J. L. Dainko, P. J. Dale,
and C.-M. Liu.....17

5. Role of Metals in Cocarcinogenesis and the Use of Liposomes for Metal Mobilization
Y. E. Rahman, P. A. Lagocki, R. A. Schwendener,
E. A. Cerny, and K. M. Rogers.....21

LOW LEVEL RADIATION

6. Radiation Toxicity Studies in Dogs
T. E. Fritz, L. S. Lombard, C. M. Poole,
T. M. Seed, D. V. Tolle, J. M. Angerman,
S. M. Cullen, D. E. Doyle, G. L. Holmblad,
L. V. Kaspar, W. G. Keenan, and A. R. Sallese.....27

7. Hematopoiesis in Chronic Toxicity Studies
T. M. Seed, L. V. Kaspar, and M. Chivers.....33

8. Life Shortening, Tumor Induction, and Tissue Dose for Fission Neutron and Gamma Ray Irradiations
D. Grahn, L. S. Lombard, J. F. Thomson, F. S. Williamson, G. L. Holmblad, J. L. Hulesch, V. A. Ludeman, A. R. Sallese, E. F. Staffeldt, J. E. Trier, and B. J. Wright.....37

9. Mammalian Genetics and Biostatistics
D. Grahn, B. A. Carnes, M. L. Garriott, B. H. Farrington, C.-H. Lee, J. J. Russell, and D. M. Eychner.....41

MOLECULAR BIOLOGY

10. Molecular Anatomy Program: The Human Protein Index
N. G. Anderson, N. L. Anderson, J. J. Edwards, C. S. Giometti, J. Taylor, K. W. Willard-Gallo, M. A. Gemmell, S. L. Nance, and S. L. Tollaksen.....49

11. Structural Studies of Proteins
M. Schiffer, C. S. Borso, C.-H. Chang, M. T. Short, P. Thiyagarajan, C. F. Ainsworth, F. A. Westholm, R. M. Hyslop, R. D. Benioff, and J. A. Guzik.....57

12. Cellular and Molecular Mechanisms of Cell Growth
H. E. Kubitschek, W. W. Baldwin, R. Graetzer, and D. Williams-Hill.....63

13. Molecular and Cellular Effects of Solar Ultraviolet Light
M. J. Peak, J. G. Peak, R. W. Tuveson, M. A. Turner, P. M. Moehring, and L. J. Sammartano.....67

TOXICOLOGY

14. Chemical Toxicology
C. A. Reilly, Jr., F. R. Kirchner, M. J. Peak, K. E. Wilzbach, R. E. Jones, J. M. Leonardo, S. S. Dornfeld, V. A. Pahnke, M. A. Shotola, D. Venters, R. M. Bressette, M. A. Fink, S. M. Kerwin, M. F. Muraski, C. M. Tompkins, and R. J. Wechsler.....75

15. Carcinogen Identification and Metabolism
D. A. Haugen, M. J. Peak, V. C. Stamoudis,
A. S. Boparai, C. A. Reilly, Jr.,
K. E. Wilzbach, K. M. Suhrbier, S. S. Dornfeld,
D. Venters, and F. J. Tremmel.....81

16. Metal Metabolism and Toxicity
M. H. Bhattacharyya, R. P. Larsen,
B. D. Whelton, P. A. Benioff, E. S. Moretti,
D. P. Peterson, R. D. Oldham,
and M. E. Shackelford.....87

17. Neurobehavioral Chronobiology and Toxicology
C. F. Ehret and K. R. Groh.....91

APPENDICES

18. Support Facilities.....97

19. Educational Activities.....101

20. Divisional Seminars, 1982.....107

21. Outside Talks by Divisional Staff during 1982.....113

22. Publications Appearing in Calendar Year 1982.....123

23. Staff and Funding Agencies.....139

AUTHOR INDEX.....147

1. DIVISION DIRECTOR'S INTRODUCTION

The Division of Biological and Medical Research at Argonne National Laboratory conducts multidisciplinary research aimed at defining the biological and medical hazards to man from energy technologies and new energy options. These technically oriented studies have a strong base in fundamental research in a variety of scientific disciplines, including molecular and cellular biology, biophysics, genetics, radiobiology, pharmacology, biochemistry, chemistry, and environmental toxicology.

During 1982, the first full year of my directorship of the Division, the Division was organized into four scientific sections:

Carcinogenesis

Low Level Radiation

Molecular Biology

Toxicology

Each section comprises four groups, as illustrated in the following organizational chart. The purposes of the new sectional organization are to emphasize broad scientific relationships and to encourage interactions among staff. Each section holds biweekly seminars which staff from other sections are encouraged to attend.

The present report is organized into four program areas, reflecting the new divisional structure. The 16 brief scientific chapters summarize the activities of the individual groups during the calendar year 1982 and the early part of 1983, but they also may contain the principal activities over a longer period to provide better continuity and perspective. Because the summaries have been kept short, they do not necessarily describe all the work of a research group.

Each of the scientific chapters are followed by a list of the publications pertinent to the project that appeared in 1981, 1982, and the first quarter of 1983. Also included are publications that were accepted for publication as of May 1, 1983.

The final chapters of this report indicate the range of other activities in the Division. They describe the animal and the radiation facilities, the varied graduate and undergraduate level educational activities, the Divisional seminars presented by visiting and in house speakers, the principal outside talks given by the staff during 1982, and give an inclusive list of Divisional publications - journal papers, reports, and abstracts -

that appeared during the calendar year 1982. The last chapter presents the staff and the Governmental agencies supporting each of the Divisional groups.

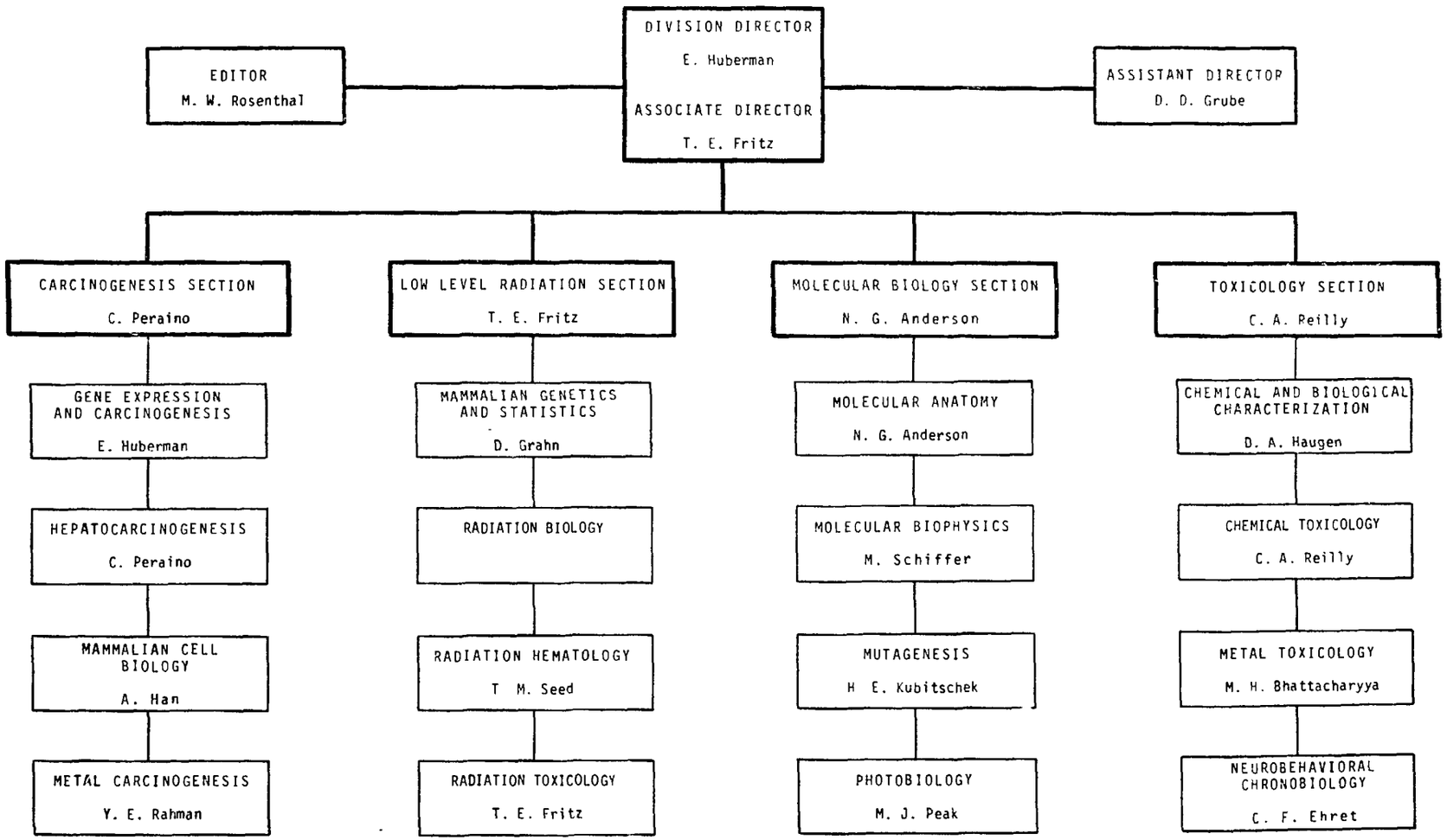
This introduction would not be complete without mention of the special honors received by the staff. The French Minister of National Education conferred on Norman G. Anderson the honorary doctorate degree, "Docteur Honoris Causa," from the University of Nancy I. Dr. Anderson also was presented with the Samuel Natelson Award of the Chicago Section of the American Association for Clinical Chemistry in recognition of his contributions to the advancement of clinical chemistry. Dr. N. Leigh Anderson received the Young Investigator Award of the American Association for Clinical Chemistry, as the young investigator making the most significant contribution to advancing human health through new developments that reach the patient through the clinical laboratory. Dr. Norman G. Anderson and Dr. N. Leigh Anderson received the 1983 Pittsburgh Analytical Chemistry Award, for their pioneering application of two-dimensional gel electrophoresis.

Dr. Charles S. Borso of the Division was honored by an IR 100 award, presented to the Laboratory as one of the 100 most significant technological advances of the year, for his photodiode X-ray detector system. Finally, Dr. Carl Peraino will be awarded a University of Chicago Award for Distinguished Performance at Argonne National Laboratory, for his model of carcinogenesis in the rat liver, a milestone in multistage carcinogenesis research.

E. Huberman

E. Huberman, Director
Division of Biological
and Medical Research

DIVISION OF BIOLOGICAL AND MEDICAL RESEARCH



11/82

C A R C I N O G E N E S I S S E C T I O N

Hepatocarcinogenesis Group, C. Peraino Principal Investigator

Chapter 2. Mechanisms of Hepatocarcinogenesis

Gene Expression and Carcinogenesis Group, E. Huberman Principal Investigator

Chapter 3. Mutagenesis and Cell Differentiation in Chemical Carcinogenesis

Mammalian Cell Biology Group, A. Han Principal Investigator

Chapter 4. Radiation-Induced Changes in Mammalian Cells

Metal Carcinogenesis Group, Y. E. Rahman Principal Investigator

Chapter 5. Role of Metals in Cocarcinogenesis and the Use of Liposomes for Metal Mobilization

2. MECHANISMS OF HEPATOCARCINOGENESIS

C. Peraino, W. E. Boernke,* J. E. Morris,† B. A. Carnes,‡ J. A. Blomquist, V. A. Ludeman, A. M. Prapuolenis, and E. F. Staffeldt

The objectives of this program are to analyze the mechanisms of tumor initiation and promotion, using an experimental rat liver system, and to develop the system as a method for the rapid screening of a broad range of environmental chemicals and other pollutants for tumor initiating or promoting activity. The experimental approach involves the administration (by intraperitoneal injection) of tumor initiators to newborn rats followed by the feeding of promoters beginning at the time of weaning. For mechanistic studies, a standard initiator (e.g., diethylnitrosamine, DEN) and a standard promoter (e.g., phenobarbital, PB) are used; a screening assay involves the substitution of a presumptive initiator or promoter for the appropriate standard agent. Subsequent histochemical analysis of frozen thin liver sections reveals the early formatiom of small foci of liver cells with biochemical changes like those appearing in many of the liver tumors that emerge later. Histochemical characteristics and incidence levels of foci and tumors are then compared.

With this new experimental protocol, altered hepatocyte foci appeared at 4 weeks of age in rats given DEN and PB; tumors appeared within 8 weeks. Substitution of the carcinogen benzo(a)pyrene (BAP) for DEN yielded foci at 11 weeks of age, and tumors appeared within an additional 8 weeks. These results constitute a 5-fold reduction in the time normally required to attain these experimental end points. The substantial activity of BAP in our system suggests that the system may have broad specificity for carcinogens, since BAP normally has minimal hepatic tumor inducing ability.

Histochemical analysis of the foci and tumors in male and female rats treated as described above have yielded the following observations.

> DEN induced more foci and tumors than BAP, and more foci appeared in females than in males.

*Visiting Scientist, Nebraska Wesleyan University.
†Scientist in Residence (Faculty Research Leave at Argonne Appointee), State University College of New York, Brockport.
‡Mammalian Genetics and Statistics Group.

The histochemical phenotypes of the foci were similar in the two carcinogen treatment groups but differed somewhat between males and females.

DEN-induced foci grew more rapidly than those induced by BAP, but focus growth rates were not influenced by sex.

Phenotypic complexity of foci was associated with focus growth in a positive though nonlinear manner; DEN and BAP exerted different effects on this relationship.

A substantial proportion (25%) of the tumors had fewer histochemically detectable phenotypic changes than the most complex and rapidly growing foci, and no one histochemical phenotype invariably occurred in all tumors.

Collectively, the foregoing results suggest that:

Structurally dissimilar carcinogens have common effects on cellular mechanisms concerned with the phenotypic expression of tumor-related markers and the control of cell division.

The mechanisms governing the expression of phenotypes of different foci and growth rates of different foci are dissimilar.

The neoplastic character of tumors is not related to their histochemical phenotypes and cannot be predicted from the histochemical phenotypes of the foci or from focus growth rates.

We conclude that tumorigenesis may not be simply an extension of processes that generate foci with progressively higher growth rates and greater phenotypic complexity. An additional possibility is that the neoplastic transformation may involve the production of a molecular lesion at a specific site that does not necessarily overlap the sites whose alteration is responsible for the derangement of growth control and phenotypic character associated with focus formation.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Effects of Self-Association of Ornithine Aminotransferase on Its Physicochemical Characteristics
W. E. Boernke, F. J. Stevens, and C. Peraino
Biochemistry 20, 115-121 (1981)

Effects of Feeding and Lighting Stimuli on the Synthesis of Ornithine Aminotransferase and Serine Dehydratase in Rat Liver
K. B. Ekelman and C. Peraino
Arch. Biochem. Biophys. 209, 677-681 (1981)

Early Appearance of Histochemically Altered Hepatocyte Foci and Liver Tumors in Female Rats Treated with Carcinogens One Day after Birth
C. Peraino, E. F. Staffeldt, and V. A. Ludeman
Carcinogenesis 2, 463-465 (1981)

1982

Differential Changes in Ornithine Aminotransferase Self-Affinity Produced by Exposure to Basic Amino Acids and Increases in the Intrinsic Electronegativity of the Enzyme Monomer
W. E. Boernke, F. J. Stevens, J. J. Edwards, and C. Peraino
Arch. Biochem. Biophys. 216, 152-157 (1982)

Initiation and Promotion of Liver Tumorigenesis
C. Peraino
Natl. Cancer Inst. Monogr. 58, 55-61 (1981)

Development of a Model to Simulate the Characteristics of Rat Liver Tumor Promotion by Phenobarbital
F. J. Stevens and C. Peraino
Carcinogenesis - A Comprehensive Survey, Vol. 7, Cocarcinogenesis and Biological Effects of Tumor Promoters, E. Hecker, N. E. Fusenig, W. Kunz, F. Marks, and H. W. Thielmann, eds., Raven Press, NY, 1982, pp. 105-109

1983 through April

Liver as a Model System for Analyzing Mechanisms of Tumor Initiation and Promotion
F. J. Stevens and C. Peraino
Organ and Species Specificity in Chemical Carcinogenesis, R. Langenbach, S. Nesnow, and J. M. Rice, eds., Plenum Publishing Corporation, 1983, pp. 231-252

In press as of May 1, 1983

Circadian Manifestations of Barbiturate Habituation, Addiction, and Withdrawal in the Rat
C. F. Ehret, C. Peraino, J. C. Meinert, and K. R. Groh
Intl. J. Chronobiol.

Multistage Hepatocarcinogenesis
C. Peraino, W. L. Richards, and F. J. Stevens
Mechanisms of Tumor Production, Vol. II, CRC Press, Uniscience Series

3. MUTAGENESIS AND CELL DIFFERENTIATION IN CHEMICAL CARCINOGENESIS

E. Huberman, C. A. Jones, E. A. Malvoisin, S.-I. Murao, B. Ryffel,*
I. Simon, M. Stodolsky,† M. F. Callaham, C. B. Henning, and B. A. Sedita

Our project is directed toward studying the mode of action of tumor initiators and promoters in cultured human and rodent cells as a means to obtain basic understanding of the role these chemicals play in the development of human cancers.

The studies involve two principal types of experiments: In the first, potential initiators of tumor development are tested for their ability to induce mutation in different cell/tissue mediated assays. The inherent features of the assay are the detection of mutagenesis in a standard mammalian target cell in the presence of intact cells from a selected organ that activate the chemical carcinogen being tested. Results from such cell mediated mutagenicity tests correlate with carcinogenicity data more closely in regard to potency and cell specificity than similar assays employing subcellular fractions for carcinogen activation.

In the second type of experiment, tumor promoters which are usually devoid of mutagenic activity, are tested for their ability to alter the expression of various markers of differentiation in human cells that are susceptible or resistant to induction of cell differentiation. The markers include acquisition of specific morphological profiles, changes in enzyme activity, alteration of the reactivity of the cells with monoclonal antibodies, and changes in specific functional characteristics, (e.g. phagocytosis, immune suppression, immune cytotoxicity).

In recent mutagenesis studies, we have reexamined this relationship between mutagenicity and carcinogenicity by testing the ability of a series of polycyclic aromatic hydrocarbons (PAH) to induce mutations (6-thioguanine and ouabain resistance) in the cell-mediated assay employing golden hamster embryo fibroblasts for carcinogen/mutagen metabolism and Chinese hamster V79 cells as the target cell. The PAH included benzo(a)pyrene (BP), dibenzo(a,h)pyrene, dibenz(a,i)pyrene [DB(a,i)P], 2,10-difluoro DB(a,i)P, 3,10-difluoro DB(a,i)P, and the ubiquitous PAH cyclopenta(c,d)-pyrene (CPP). The results indicated that the degree of mutagenicity of these PAH in the cell mediated assay was in agreement with the activity of these chemicals as measured by *in vivo* mouse tumorigenesis studies. No such relationship was established using the *Salmonella typhimurium* assay

*Resident Associate (Sandoz, Ltd., Basel, Switzerland).
†Visiting Scientist.

in which activation of chemicals is obtained by subcellular liver fraction (S-9). More specifically, the two difluoro dibenzopyrenes that are not tumorigenic in vivo and not mutagenic in the cell mediated assay were potent mutagens in the Salmonella assay. CPP, which we found to be many times less active than BP in initiating tumors in the mouse skin and in inducing mutations in the cell mediated assay was more active than BP in inducing mutations in the Salmonella system. We plan to extend these studies to investigate organ-specific differences in chemical carcinogenesis using epithelial cells from human cell lines as target cells for the mutagenic insult, and primary hepatocytes and epithelial esophageal and colonic cells from various species for carcinogen activation.

The effect of tumor promoters was analyzed in human T lymphoid (CEM) cells and promyelocytic myeloid (HL-60) leukemia cells. Treatment of CEM leukemia cells with low concentrations of phorbol-12-myristate-13-acetate (PMA) causes the cells to express a phenotype that resembles that of a suppressor T lymphocyte. This phenotype can be characterized in part by the appearance of a specific antigenic pattern. More specifically, PMA causes the cells to exhibit increased reactivity with OKT3 and OKT8 monoclonal antibodies that characterize mature suppressor and cytotoxic T lymphocytes, respectively. PMA also reduces the reactivity of the treated cells with the monoclonal antibodies that detect inducer/helper and immature "common" thymocytes. In addition, treated CEM cells, in common with mature suppressor T lymphocytes, are able to suppress [^{3}H]thymidine incorporation into phytohemaglutinin-activated peripheral blood lymphocytes but, unlike cytotoxic lymphocytes, are unable to induce a cytotoxic response in a number of human and rodent target cells.

The ability of PMA to induce differentiation of CEM cells into suppressor cells raises the possibility that tumor promotion by PMA and related agents in the mouse skin may, among other things, involve a suppression of the immunity against "initiator"-induced papillomas and carcinomas. Such a suppression could result from an increase in the number of mature suppressor T lymphocytes in the promoted animal.

In other studies, we have shown that treatment of HL-60 cells with PMA stimulated the incorporation of label from L-[methyl-^{3}H]methionine into the cellular phospholipids in a dose and time dependent manner. Such a stimulation of phospholipid methylation was not observed in an HL-60 cell variant that is resistant to phorbol ester-induced differentiation. The stimulation in phospholipid methylation by PMA was confirmed by measuring the activity of phosphatidylethanolamine methyltransferase in cellular lysates. Phospholipid methylation was also stimulated after treatment with two other tumor promoters and inducers of cell differentiation in HL-60 cells, phorbol-12,13-dibutyrate and teleocidin. Phorbol-12,13-diacetate and 4-O-methyl PMA, which are inactive as tumor promoters or inducers of cell differentiation, did not stimulate phospholipid methylation. These studies suggest that phospholipid methylation can be involved in the process of cell differentiation in HL-60 cells induced by tumor promoting phorbol esters and teleocidin.

Based on these experiments, we suggest that studies on the control of mutagenesis in the cell mediated assays and studies that analyze the control of cell differentiation of specific cell types, including those that mature into suppressor T lymphocytes, can be useful in explaining basic concepts in carcinogenesis.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Mutagenesis by Chemical Agents in V79 Chinese Hamster Cells: A Review and Analysis of the Literature. A Report of the Gene-Tox Program
M. O. Bradley, B. Bhuyan, M. C. Francis, R. Langenbach, A. Peterson, and E. Huberman
Mutat. Res. 87, 81-142 (1981)

Mutagenesis and Differentiation Induction in Mammalian Cells by Environmental Chemicals (ORNL work)
J. Friedman and E. Huberman
Postepy Mikrobiol. 20/1-2, 93-102 (1981)

The Induction of Mutation and Cell Differentiation by Chemicals which Initiate or Promote Tumor Formation (ORNL work)
E. Huberman
Mutation, Promotion, and Transformation, C. Heidelberger, N. Inui, and T. Kuroki, eds., Gann Monograph on Cancer Research 27, 129-145 (1981)

Induction of Mutation and Cell Differentiation in Cultured Mammalian Cells as Indicators of Potential Carcinogenic Risk (ORNL work)
E. Huberman, B. S. Hass, and C. A. Jones
Health Risk Analysis - Proceedings of the Third Life Sciences Symposium, C. R. Richmond, P. J. Walsh, and E. D. Copenhaver, eds., Franklin Institute Press, Philadelphia, PA, 1981, pp. 287-309

Mutagen-Induced Resistance to Mycophenolic Acid in Hamster Cells Can Be Associated with Increased Inosine 5'-Phosphate Dehydrogenase Activity (ORNL work)
E. Huberman, C. K. McKeown, and J. Friedman
Proc. Natl. Acad. Sci., 78, 3151-3154 (1981)

Alterations in Polyamine Levels Induced by Phorbol Diesters and Other Agents That Promote Differentiation in Human Promyelocytic Leukemia Cells (ORNL work)
E. Huberman, C. Weeks, A. Herrmann, M. Callaham, and T. Slaga
Proc. Natl. Acad. Sci. 78, 1062-1066 (1981)

The Relationship between the Carcinogenicity and Mutagenicity of Nitrosamines in a Hepatocyte-Mediated Mutagenicity Assay (ORNL work)
C. A. Jones, P. J. Marlino, W. Lijinsky, and E. Huberman
Carcinogenesis 2, 1075-1077 (1981)

Down Regulation of Specific Binding of [20-^{3}H]Phorbol-12,13-Dibutyrate and Phorbol Ester-Induced Differentiation of Human Promyelocytic Leukemia Cells (ORNL work)
V. Solanki, T. J. Slaga, M. Callaham, and E. Huberman
Proc. Natl. Acad. Sci. 78, 1722-1725 (1981)

1982

Cell-Mediated Mutagenicity in Chinese Hamster V79 Cells of Dibenzopyrenes and Their Bay-Region Fluorine-substituted Derivatives (ORNL work)
B. S. Hass, C. K. McKeown, D. J. Sardella, E. Boger, P. K. Ghoshal, and E. Huberman
Cancer Res. 42, 1646-1649 (1982)

The Control of Phospholipid Methylation by Phorbol Diesters in Differentiating Human Myeloid HL-60 Leukemia Cells (ORNL work)
D. R. Hoffman and E. Huberman
Carcinogenesis 3, 875-880 (1982)

Induction of Differentiation of Human Promyelocytic Leukemia (HL-60) Cells by Teleocidin and Phorbol-12-Myristate-13-Acetate (ORNL work)
E. Huberman, G. R. Braslawsky, M. Callaham, and H. Fugiki
Carcinogenesis 3, 111-114 (1982)

Cell Differentiation, Alterations in Polyamine Levels, and Specific Binding of Phorbol Diesters in Cultured Human Cells (ORNL work)
E. Huberman, C. Weeks, V. Solanki, M. Callaham, and T. Slaga
Carcinogenesis - A Comprehensive Survey, Vol. 7, Cocarcinogenesis and Biological Effects of Tumor Promoters, E. Hecker, N. E. Fusenig, W. Kunz, F. Marks, and H. W. Thielmann, eds., Raven Press, NY, 1982, pp. 405-416

The Effect of Tumor-Causing Chemicals on the Control of Mutagenesis and Differentiation in Mammalian Cells (ORNL work)
C. A. Jones and E. Huberman
Biological Carcinogenesis, M. A. Rich and P. Furmanski, eds., Marcel Dekker, Inc., New York and Basel, 1982, pp. 107-121

Cell-Mediated Mutagenesis and Tumor-Initiating Activity of the Ubiquitous Polycyclic Hydrocarbon, Cyclopenta[c,d]pyrene (ORNL work)
D. Raveh, T. J. Slaga, and E. Huberman
Carcinogenesis 3, 763-766 (1982)

1983 through April

[Committee on Chemical Environmental Mutagens, National Research Council]
J. F. Crow, S. Abrahamson, C. Denniston, D. G. Hoel, E. Huberman, P. N. Magee, D. W. Nebert, T. Roderick, M. W. Shaw, F. Sherman, V. F. Simmon, H. E. Sutton, and S. Wolff
Identifying and Estimating the Genetic Impact of Chemical Mutagens, N. Grossblatt, ed., National Academy Press, Washington, D. C., 1982

Studies on the Mode of Action of Chemical Carcinogens in Cultured Mammalian Cells (ORNL work)
C. Jones and E. Huberman
Organ and Species Specificity in Chemical Carcinogenesis, R. Langenbach, S. Nesnow, and J. M. Rice, eds., Plenum Publishing Corporation, 1983, pp. 405-427

Differentiation of Human T-Lymphoid Leukemia Cells into Cells That Have a Suppressor Phenotype Is Induced by Phorbol 12-Myristate 13-Acetate
B. Ryffel, C. B. Henning, and E. Huberman
Proc. Natl. Acad. Sci. USA 79, 7336-7340 (1982)

In press as of May 1, 1983

A Microtiter Plate Assay for the Selection of 6-Thioguanine Resistant Mutants in Chinese Hamster V79 Cells in the Presence of Phorbol-12-Myristate-13-Acetate (ORNL work)
D. Raveh and E. Huberman
Mutat. Res.

4. RADIATION-INDUCED CHANGES IN MAMMALIAN CELLS

A. Han, C. K. Hill, R. L. Wells, M. M. Elkind,* M. P. Hagan,†
D. L. Redman,‡ V. N. Smith,** E. M. Buess, J. L. Dainko, P. J. Dale, and
C.-M. Liu

The primary objective of this project is to understand the mechanisms of radiation-induced changes in mammalian cells that lead to cell lethality, mutation, and neoplastic transformation. The influence of repair processes on these end points is of particular interest. Radiations used include different qualities of ionizing radiations and nonionizing radiations of different wavelengths.

The principal techniques that are used include clonal growth of single cells to measure cell lethality or induction of mutants, labeling of macromolecules with radioisotope-labeled precursors, chemical identification of protein and nucleic acid components, and measurement of damage to DNA using sedimentation and elution techniques. In studies of neoplastic transformation *in vitro*, tests for altered properties are made by measuring the ability of cells to grow in semisolid agar, increased production of plasminogen activator, and tumorigenicity of transformed cells after their inoculation into suitable hosts.

Neoplastic transformation of mammalian cells exposed to ^{60}Co gamma rays or fission neutrons. Protracted exposures to ^{60}Co gamma rays or fission-spectrum neutrons (JANUS reactor) were given either at a low dose rate or as multifractionated regimes using a high dose rate. Dose protraction of ^{60}Co gamma ray irradiation by multifractionation at a high dose rate (50 rads/min) or by delivery at a low dose rate (0.1 rad/min) results in significant reduction in cell killing and, independent of the effect on cell survival, dose protraction also significantly reduces the incidence of neoplastic transformation. In contrast to gamma rays, the protracted exposures to fission-spectrum neutrons, given either at a low dose rate (0.086 rad/min) or in multiple fractions at a high dose rate (10 rads/min), result in significant enhancement of the frequency of transformation in

---

*Dual Appointment, Senior Biophysicist, Argonne National Laboratory, and Chairman, Department of Radiology and Radiation Biology, Colorado State University, Fort Collins.
†Scientist, Armed Forces Radiobiology Research Institute, Bethesda, MD.
‡Fall 1982 participant in the Undergraduate Honors Program, Ohio Wesleyan University.
**Spring 1982 participant in the Undergraduate Honors Program, Goucher College.

the low dose region (2.4-80 rads). However, the survival of cells for these different modes of exposure to neutrons is essentially unchanged from the survival after a single dose of neutrons given at an acute dose rate. The initial parts of the neoplastic transformation induction curves for acute and protracted exposures of both ^{60}Co gamma rays and fission-spectrum neutrons fit a straight line quite well. However, the slope of the regression line for protracted exposures with neutrons is about 10 times steeper than the slope for single acute exposures. The estimates of the relative biological effectiveness for transformation by fission neutrons, as measured by the slopes of the initial portions of the induction curves, increases from about 2.5 for acute delivery to about 50 for protracted irradiation.

Repair of potentially lethal damage and mutagenesis. The repair of potentially lethal X-ray damage in actively growing Chinese hamster cells can be inhibited by anisotonic phosphate-buffered saline. Treatment of cells (20 minutes at 37°C) immediately after X-irradiation increases the frequency of mutants that are resistant to 6-thioguanine. Within the dose range examined (100-500 rads) the effect increases with dose. If the treatment with anisotonic saline is delayed and cells are allowed to repair the damage, the mutation frequency is rapidly reduced to the levels of frequencies for irradiated but nontreated cells. Measurements indicate a half time of this repair process to be about 15-20 minutes.

Molecular damage to DNA by far-UV and near-UV light. The efficiency of pyrimidine dimer induction is significantly less following 313-nm irradiation than after 254 nm, and pyrimidine dimers do not seem to be of biological importance following exposure of cells to 365-nm light. Thus, we have measured the induction of DNA-to-protein cross-links in Chinese hamster cells. Measurements of the elution rates of DNA from cells exposed to 365-nm or 405-nm light, by the alkaline elution method, demonstrate a dose dependent increase in the rate of DNA elution. Treatment of DNA with proteinase K, an enzyme that breaks the covalent bonds between DNA and protein residues cross-linked to it, results in an increased rate of DNA elution. The cross-linking of DNA and proteins is readily demonstrable for exposures of 365 nm and 405 nm within the first decade of survival. Induction of pyrimidine dimers, however, as measured by the number of UV-endonuclease (T4) sensitive sites, is insignificant by 365-nm light and no dimers are induced by 405-nm light.

The foregoing studies with mammalian cells exposed to ionizing or nonionizing radiations indicate the important role of repair processes in radiation damage. They suggest that the repair of transformational and/or mutational damage is mediated by a repair process(es) independent of those concerned with the repair of damage leading to cell lethality. The studies are being extended to molecular mechanisms, involving DNA in particular, that might be important in the expression of damage that results in mutation or neoplastic transformation.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Recovery after Exposure to Near-Ultraviolet Light of Cells Containing 5-Bromodeoxyuridine
M. P. Hagan and M. M. Elkind
Biophys. J. 34, 367-374 (1981)

Hydroxyurea Revisited: A Decade of Clinical Effects Studies
W. K. Sinclair
Int. J. Radiat. Oncol. Biol. Phys. 7, 631-637 (1981)

Spectral Dependencies of Killing, Mutation, and Transformation in Mammalian Cells and Their Relevance to Hazards Caused by Solar Ultraviolet Radiation
F. Suzuki, A. Han, G. R. Lankas, H. Utsumi, and M. M. Elkind
Cancer Res. 41, 4916-4924 (1981)

"Single-Hit" Potentially Lethal Damage: Evidence of Its Repair in Mammalian Cells
H. Utsumi, C. K. Hill, E. Ben-Hur, and M. M. Elkind
Radiat. Res. 87, 576-591 (1981)

1982

Keynote Address - Modifiers of Radiation Response in Tumor Therapy: Strategies and Expectations
M. M. Elkind
Int. J. Radiat. Oncol. Biol. Phys. 8, 89-100 (1982)

Neoplastic Transformation and Dose Fractionation: Does Repair of Damage Play a Role? [Summary of paper published in Radiat. Res. 79, 233-240 (1979)]
M. M. Elkind and A. Han
The Year Book of Cancer 1981, R. L. Clark, R. W. Cumley, and R. C. Hickey, eds., Year Book Medical Publishers, Inc., Chicago, 1981, pp. 368-370

Enhanced Transformation of Mouse 10T1/2 Cells by 12-O-Tetradecanoyl-phorbol-13-Acetate following Exposure to X-Rays or to Fission-Spectrum Neutrons
A. Han and M. M. Elkind
Cancer Res. 42, 477-483 (1982)

Fission-Spectrum Neutrons at Reduced Dose Rates Enhance Neoplastic Transformation
C. K. Hill, F. M. Buonaguro, C. P. Myers, A. Han, and M. M. Elkind
Nature 298, 67-69 (1982)

Spectral Dependencies of Killing, Mutation, and Transformation in Mammalian Cells and Their Relevance to Hazards Caused by Solar Ultraviolet Radiation
F. Suzuki, A. Han, G. R. Lankas, H. Utsumi, and M. M. Elkind
Cancer Res. 41, 4916-4924 (1981)

1983 through April

Alkali-Labile Lesion and Uracil-DNA-Glycosylase-Sensitive Site Removal after BrdUrd and UVB Treatment of Chinese Hamster Cells
M. P. Hagan, T. Matsushita, T. Bonura, and A. Shotola
Photochem. Photobiol. 35, 371-377 (1982)

Neoplastic Transformation in Vitro: Dose Rate Dependence of the Relative Effectiveness of Fission-Spectrum Neutrons versus ^{60}Co γ-Rays
C. K. Hill, A. Han, and M. M. Elkind
Radiation Protection: Neutron Carcinogenesis, EUR 8084 EN, J. J. Broerse and G. B. Gerber, eds., European Seminar jointly organized by the Commission of the European Communities, DG XII Biology, Rad. Protection and Medical Research and the Radiobiological Institute TNO, NL - Rijswijk, March 30-April 1, 1982, pp. 397-405

Suppressive Action of Near-Ultraviolet Light on Ouabain Resistance Induced by Far-Ultraviolet Light in Chinese Hamster Cells
F. Suzuki, A. Han, C. K. Hill, and M. M. Elkind
Mutat. Res. 119, 399-406 (1983)

In press as of May 1, 1983

Modifiers of Radiation Response in Tumor Therapy: Strategies and Expectations
M. M. Elkind
Biological Bases & Clinical Implications of Tumor Radioresistance, Chapter 37

Non-Standard Fractionation: Clinical Observations
L. R. Holsti, M. Salmo, and M. M. Elkind
2nd Rome International Symposium, Biological Bases and Clinical Implications of Tumor Resistance, Rome, Italy, September 23-25, 1980

Caffeine-Enhanced Survival of Radiation-Sensitive Repair-Deficient Chinese Hamster Cells
H. Utsumi and M. M. Elkind
Radiat. Res.

5. ROLE OF METALS IN COCARCINOGENESIS AND THE USE OF LIPOSOMES FOR METAL MOBILIZATION

Y. E. Rahman, P. A. Lagocki, R. A. Schwendener, E. A. Cerny, and K. M. Rogers*

The objectives of this program are to determine the role of metals in tumor induction, elucidate the mechanisms of their carcinogenic action, and develop new approaches for reducing the tumorigenicity and toxicity of the relevant metals.

Studies on the role of metals in tumor induction currently focus on arsenic. In a study of carcinogenesis of the skin, mice are given arsenic at different levels in the drinking water, concomitantly first with a tumor initiator (physical or chemical) and then with application of a skin tumor promoter. The mice are then monitored for the development of skin tumors. In other studies focusing on hepatocarcinogenesis, rats are similarly exposed to arsenic in the drinking water, administered a hepatocarcinogen, and then fed phenobarbital as a promoter. The livers are examined histochemically at intervals for the development of preneoplastic foci.

In the development of a new approach to mobilize toxic metals from storage organs of animals, two mouse models for experimental iron overload are used. (1) Iron overload in the reticuloendothelial system is produced by administration of heat damaged ^{59}Fe-labeled red blood cells. (2) Iron overload in the liver parenchymal cells is produced by administration of purified ^{59}Fe-labeled ferritin. Lipid vesicles (liposomes) differing in size and lipid composition are used to encapsulate various metal chelators for delivery to specific metal storage organs. Short-term cultures of liver parenchymal cells and peritoneal macrophages are used in <u>in vitro</u> studies of liposome-cell interactions.

Results from a first experiment testing the cocarcinogenic action of arsenic and ultraviolet (UV) radiation in the induction of skin cancer in SKH:hairless-1 mice (in collaboration with Drs. M. J. Peak and L. S. Lombard) have provided evidence for an enhancing effect of arsenic on skin tumor induction. This study shows that the toxic metal arsenic can play an important role as a cocarcinogen. The results can be summarized as follows: (1) The previous work by others showing that arsenic alone is not a primary carcinogen has been confirmed. (2) Arsenic trioxide at subtoxic levels in the drinking water, given concomitantly first with UVB (290-320 nm) and then with the tumor promoter 12-O-tetradecanoyl

*Resident Student Associate, Summer 1982.

phorbol-13-acetate (TPA) enhances induction of skin tumors (papillomas and squamous cell carcinomas) in female SKH:hairless-1 mice. (3) Arsenic at toxic levels in the drinking water given concomitantly with UV and then TPA almost totally inhibits skin tumor induction in the mice. This result may explain the negative findings obtained by previous workers, who gave arsenic in drinking water at a level of 100 mg/L compared with our highest chronic level of 60 mg/L. (4) Arsenic at subtoxic levels in the drinking water inhibits the spontaneous regression of the skin papillomas induced by UV and TPA in the mice.

Analysis of a study involving the induction of preneoplastic foci and tumors in the liver of rats, in response to oral arsenic with diethylnitrosamine (as tumor initiator) and phenobarbital, has so far shown (1) female rats have a higher incidence of preneoplastic foci than males in all treatment groups, and (2) arsenic appears to have an enhancing effect on the induction of preneoplastic foci in the liver of female rats (results from animals up to 12 weeks after initiator administration).

Results of basic studies on liposome-cell interactions, using both in vivo and in vitro systems, include the following. (1) Liver parenchymal cells have endocytic capabilities; that is, they can take up not only small single membrane liposomes of about 25-nm diameter, but can also effectively take up large single membrane liposomes of up to 160-nm diameter. Endocytic properties have not been previously recognized in these cells. (2) Reticuloendothelial cells (e.g., macrophages) can more effectively take up liposomes with a positive surface charge. (3) Addition of a galactolipid to the membrane bilayers of liposomes increases the liposome uptake by a liver tumor cell line (H-129).

A new iron chelator of low water solubility, N,N'-bis(2-hydroxybenzyl)-ethylenediamine-N,N'-diacetic acid (HBED), has been encapsulated in the lipid bilayers of liposomes. We have found that HBED is effective in removing excess iron specifically loaded in liver parenchymal cells; desferrioxamine, the iron chelator currently in use for treatment of patients with iron overload, is ineffective in this regard.

A study on the interaction of a phospholipid-nucleoside conjugate with serum lipoproteins has shown that high density lipoprotein interacts rapidly with the nucleoside conjugate, and that the extent of interaction is dependent upon the characteristic gel-liquid crystalline phase transition of the conjugate's phospholipid (work in collaboration with Jesse J. Edwards and Malcolm MacCoss).

In conclusion, although epidemiologic studies in man have clearly shown that chronic exposure to arsenic is associated with increased incidence of cancer, especially skin tumors, arsenic has not been found to be tumorigenic in experimental animals. We have now obtained results that show, for the first time in the laboratory, an enhancing effect of arsenic upon the induction of skin tumors by UV irradiation. Confirmation of arsenic-enhanced skin tumorigenesis caused by a physical or chemical carcinogen would provide a model for studies of arsenic carcinogenesis.

The finding of an endocytic property for liver parenchymal cells is unexpected. Further understanding of the cell surface behavior of these cells should be of importance not only in basic cell biology, but also in designing suitable carrier systems for drug targeting.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Liposome-encapsulated Desferrioxamine in Experimental Iron Overload
E. H. Lau, E. A. Cerny, and Y. E. Rahman
Br. J. Haematol. 47, 505-518 (1981)

1982

In Vitro Uptake and Therapeutic Application of Liposome-encapsulated Methotrexate in Mouse Hepatoma 129
K. R. Patel, M. M. Jonah, and Y. E. Rahman
Eur. J. Cancer Clin. Oncol. 18, 833-843 (1982)

Liposomes as Delivery Systems for Iron Chelators
Y. E. Rahman
Proceedings of the Second Symposium on Development of Iron Chelators for Clinical Use, San Francisco, CA, August 23-24, 1980, A. E. Martell, W. F. Anderson, and D. G. Badman, eds., Elsevier North Holland, Inc., 1981, pp. 211-225

Differential Uptake of Liposomes Varying in Size and Lipid Composition by Parenchymal and Kupffer Cells of Mouse Liver
Y. E. Rahman, E. A. Cerny, K. R. Patel, E. H. Lau, and B. J. Wright
Life Sci. 31, 2061-2071 (1982)

1983 through April

In press as of May 1, 1983

Liposomes Targeted to Cellular Receptors
M. M. Jonah and Y. E. Rahman
Proceedings of the 8th International Subcellular Methodology Conference on "Membrane Located Receptors," London, England, July 12-17, 1982

Improvement of Iron Removal from the Reticuloendothelial System by Liposome Encapsulation of N,N'-bis[2-hydroxybenzyl]-ethylenediamine-N,N'-diacetic Acid (HBED). Comparison with Desferrioxamine.
E. H. Lau, E. A. Cerny, B. J. Wright, and Y. E. Rahman
J. Lab. Clin. Med. 101, 806-816 (1983)

Liposomes as Carriers for Chelators
Y. E. Rahman
Liposome Letters, A. D. Bangham, ed., Academic Press, London, Chapter 33

Enhanced Iron Removal from Liver Parenchymal Cells in Experimental Iron Overload. Liposome Encapsulation of HBED and Phenobarbital Administration
Y. E. Rahman, E. A. Cerny, E. H. Lau, and B. A. Carnes
Blood

LOW LEVEL RADIATION SECTION

Radiation Toxicology Group, T. E. Fritz Principal Investigator

Chapter 6. Radiation Toxicity Studies in Dogs

Radiation Hematology Group, T. M. Seed Principal Investigator

Chapter 7. Hematopoiesis in Chronic Toxicity Studies

Radiation Biology Group, D. Grahn Principal Investigator

Chapter 8. Life Shortening, Tumor Induction, and Tissue Dose for Fission Neutron and Gamma Ray Irradiations

Mammalian Genetics and Statistics Group, D. Grahn Principal Investigator

Chapter 9. Mammalian Genetics and Biostatistics

6. RADIATION TOXICITY STUDIES IN DOGS

T. E. Fritz, L. S. Lombard, C. M. Poole, T. M. Seed, D. V. Tolle, J. M. Angerman, S. M. Cullen, D. E. Doyle, G. L. Holmblad, L. V. Kaspar, W. G. Keenan, and A. R. Sallese

The basic objective of this program is the generation of comprehensive data on the late effects of low doses of ionizing radiation in a large, relatively long-lived animal, the dog, that will be useful for extrapolation to man. The specific objectives are: (1) to determine the influence of two major factors of exposure - the daily exposure rate and the accumulated total exposure; (2) to provide data for estimates of radiation-specific excess mortality in the dog to enable interspecies comparisons to existing rodent data and for development of a unifying concept of damage that has predictive value in man; and (3) to provide base-line data and specimens to characterize the pathogenesis and mechanisms of induction of leukemia, aplastic anemia, and other diseases, including late occurring soft tissue tumors.

These experiments utilize protracted whole-body ^{60}Co gamma radiation given to young adult beagles 22 hours/day, 7 days/week. They characterize the responses at various exposure rates given either until death (0.4, 1.0, and 2.5 R/day) or until predetermined total exposures of irradiation are accumulated (600, 1400, 2000, and 4000 R). The latter are delivered at exposure rates of 5, 10, 17, and 35 R/day to compare with dogs previously irradiated until death at these exposure rates. The dogs are maintained for their entire life-span, and are monitored and evaluated at regular intervals by hematologic, clinical, and pathological examinations, and a complete necropsy is performed at death. All data are entered into a computerized data base.

Responses of adult beagles to continuous exposure to ^{60}Co gamma rays. We have continued to monitor the dogs being irradiated continuously at 2.5, 1.0, and 0.4 R/day. As of April, 1983, these dogs had been irradiated for times varying from 1539 to 2609 days after their entry into the experiment at 400 days of age. They are in generally good health and cannot be differentiated clinically from the controls, except for depression of circulating blood cell numbers and testicular atrophy in males.

The deaths since last year of an additional 3 dogs gives a total of 20 decedents (19 experimental dogs and 1 control). There have now been 12 deaths in the 2.5 R/day group, 3 in the 1.0 R/day group, and 4 in the 0.4 R/day group. In the latter two groups, none of the deaths are believed related to the irradiation (one case of encephalitis, two cases of a herniated vertebral disc, and one case of septicemia secondary to mastitis). The dead control dog died with signs of central nervous system

disease related to cerebellar hypoplasia, a developmental anomaly. In the 2.5 R/day group, the deaths of 8 of the dogs were related to the hematopoietic system: 6 were from leukemia, 1 was caused by a mast cell tumor, and 1 was from aplastic anemia. Two of the remaining dogs in this group have died of spondylitis with secondary myelitis (compression and hemorrhage), one with virus enteritis and one with a widely metastasizing hemangiosarcoma of the abdominal cavity originating in the spleen.

As reported previously, there was early depression of thrombocyte and leukocyte values in all three exposure groups, but erythrocyte values remained stable for more than 1000 days, even at the highest exposure rate. There are exposure rate-dependent differences in the peripheral blood values for the three groups. In earlier reports, we pointed out that these differences are not seen when the blood values are plotted against total accumulated exposure rather than against time in the radiation field. However, the additional data obtained over the last two years, with continuing irradiation, tend to show that values for both thrombocytes and leukocytes are stabilizing at differing exposure rate-dependent levels when the data are plotted against the greater accumulated total dose. At 600 R total exposure and greater, the leukocyte values for the 0.4 R/day group remain higher than those for the 1.0 and 2.5 R/day groups, and at 1200 R total exposure and greater, the leukocyte values for the 1.0 R/day group remain higher than those in the 2.5 R/day group.

Influence of dose rate and total dose on late effects of terminated exposures of adult beagles to ^{60}Co gamma rays. The dogs given terminated exposures to predetermined total doses range between 2417 and 5633 days of age as of April 1, 1983. Although only 106 of 257 chronic survivors (41%) and a similar percentage of the controls have died in these experimental groups, several interim observations and tentative conclusions can be made.

Myelogenous leukemia occurs as the earliest occurring malignancy in these dogs as well as in dogs given continuous irradiation. The incidence of the disease is lower than in dogs irradiated continuously. In both groups, irradiation continued beyond a certain time or total dose may not influence the incidence. For example, in the dogs irradiated continuously at 10 R/day, the longest term survivors died of other malignancies, following an early wave of deaths from leukemia.

Our previous conclusions regarding limiting factors for radiation induction of myeloproliferative disease (MPD) with these terminated exposures still stand. These conclusions are (1) the minimum terminated exposure to produce MPD is about 1700 R [2000 R in these studies, but 1700 R (17 R/day for 100 days) in an earlier pilot study]; (2) exposure rates of 5, 10, and 17 R/day but not 35 R/day produce MPD; and (3) the minimum and maximum times to death with MPD are 400 and 1800 days, respectively.

Although the effects of exposure rate and total exposure cannot yet be determined for these terminated exposures, several generalizations on causes of death and tumor incidences can be made. At the present time, the incidence of malignancies in all the terminated exposure groups is not significantly different from that in controls. Whether the total fraction

of each group (all irradiated dogs versus controls) developing a malignancy is compared, or the fraction of the decedents in each group is compared, the incidence is not different. However, the age at the time of death from all causes, and also the age of those dogs dying of a malignancy, is significantly different between the irradiated and control dogs. For example, the irradiated dogs are dying approximately 600 days earlier than controls [2968 ± 95 (S.E.) days vs. 3536 ± (S.E.) days; $P < .001$]. The most common malignancy among both control and irradiated groups is the mammary carcinoma. In the irradiated dogs, sarcomas, particularly nerve sheath and vascular sarcomas of the spleen, have occurred in large numbers, but these tumors are infrequent in the control population. The numbers of lymphoid tumors did not differ significantly between irradiated and control dogs, but these tumors are occurring significantly earlier in irradiated dogs. The early occurrence of myelogenous and lymphoid malignancies as well as sarcomas and mammary carcinomas represent the major factor in the shorter survival time of the irradiated groups.

Another significant observation is a decreased incidence of thyroid tumors among the terminated irradiation groups as compared with controls. This reduced frequency of thyroid malignancies is related to the competing risk of death from other tumors in the irradiated dogs: The mean time to death from thyroid carcinomas in the controls is 4153 ± 223 (S.E.) days whereas the life-span of irradiated dogs is 2968 ± 95 (S.E.) days.

In summary, for the continuous exposures, the data to date support the hypothesis that the biological responses below 5 R/day are related to total dose rather than dose rate, as previously shown for life shortening in mice. For the terminated exposures, the data thus far available suggest that certain specific tumor types are occurring in excess of those observed in the control groups and at earlier times. Observation and monitoring of all dogs will continue, in order to provide data for further comparisons with the shorter lived laboratory species. These studies will help achieve our overall goal of assessing the hazards and understanding the mechanisms of radiation damage to man.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Viral Antibody Studies of Laboratory Dogs with Diarrheal Disease
L. N. Binn, R. H. Marchwicki, E. H. Eckermann, and T. E. Fritz
Am. J. Vet. Res. 42, 1665-1667 (1981)

Autoimmune Diseases of Canines
T. E. Fritz and N. R. Rose
The Canine as a Biomedical Research Model: Immunological, Hematological, and Oncological Aspects, M. Shifrine and F. D. Wilson, eds., 1980, Chap. 11, pp. 231-243

1982

A Direct Data Entry System for Toxicologic Pathology
D. E. Doyle, L. S. Lombard, and T. E. Fritz
Toxicol. Pathol. 9, 22-29 (1981)

Hematologic Responses of Beagles Exposed Continuously to Low Doses of ^{60}Co γ-Radiation
T. E. Fritz, D. V. Tolle, D. E. Doyle, T. M. Seed, and S. M. Cullen
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 28, pp. 229-240

Effects of Chronic Continuous Low Level γ-radiation Exposure (CCLLR) on Hematopoiesis in the Mouse
W. H. Knospe, S. S. Adler, S. Husseini, T. Fritz, and F. D. Wilson
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 28, pp. 251-259

Radiosensitivity and Life Span of Dog Peripheral Blood Lymphocytes
A. Leonard, G. Decat, and T. E. Fritz
Mutat. Res. 92, 257-263 (1982)

Sequential Changes in Bone Marrow Architecture during Continuous Low Dose Gamma Irradiation
T. M. Seed, G. T. Chubb, and D. V. Tolle
Scanning Electron Microscopy/1981/IV, O. Johari, R. M. Albrecht, and T. M. Seed, eds., SEM, Inc., AMF O'Hare (Chicago), IL, 1982, pp. 61-72

Leukemia Induction in Beagles Exposed Continuously to ^{60}Co γ-Irradiation: Hematology
D. V. Tolle, T. E. Fritz, T. M. Seed, S. M. Cullen, L. S. Lombard, and C. M. Poole
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 29, pp. 241-249

1983 through April

Hemopathologic Predisposition and Survival Time under Continuous Gamma Irradiation: Responses Mediated by Altered Radiosensitivity of Hemopoietic Progenitors
T. M. Seed, L. V. Kaspar, D. V. Tolle, and T. E. Fritz
Exp. Hematol. 10, Suppl. No. 12, 232-248 (1982)

Circulating Micromegakaryocytes Preceding Leukemia in Three Dogs Exposed to 2.5 R/day Gamma Radiation
D. V. Tolle, S. M. Cullen, T. M. Seed, and T. E. Fritz
Vet. Pathol. 20, 111-114 (1983)

Aberrant Megakaryocytopoiesis Preceding Radiation-Induced Leukemia in the Dog

D. V. Tolle, T. M. Seed, S. M. Cullen, C. M. Poole, and T. E. Fritz
Scanning Electron Microscopy/1982/I, O. Johari, R. M. Albrecht, and T. M. Seed, eds., SEM Inc., AMF O'Hare (Chicago), Il., 1983, pp. 367-376

In press as of May 1, 1983

Increased Incidence of Malignant Tumors in Dogs after Total Body Irradiation and Marrow Transplantation

H. J. Deeg, R. Prentice, T. E. Fritz, G. E. Sale, L. S. Lombard, E. D. Thomas, and R. Storb
Int. J. Radiat. Oncol. Biol. Phys.

7. HEMATOPOIESIS IN CHRONIC TOXICITY STUDIES

T. M. Seed, L. V. Kaspar, and M. Chivers*

Aplastic anemia and myelogenous leukemia occur in man as "spontaneous" diseases as well as following human exposure to radiation and chemical toxicants (e.g., benzene). This project uses the dog exposed to protracted low level gamma radiation to study the early changes in bone marrow structure and function involved in the prepatent stages of these two hemopathologies. Characterization of the early cellular events involved in the initial stages of aplastic anemia and leukemia is important in order to determine criteria for early diagnosis and prognosis and perhaps to develop preventive measures. The objectives of the project are to (1) define the nature of pathological progression of both aplastic anemia and myeloid leukemia in beagles continuously exposed to low daily doses of gamma radiation and (2) elucidate cellular mechanisms of hematopoietic accommodation under chronic low dose irradiation, and the relationship between the accommodation and the induction and development of the major hemopathological end points.

Both whole animal and cell culture techniques are used to assess the sequential changes within the blood forming system of dogs irradiated at 7.5 rads per 22 hour day. The clinical monitoring of the animals included in this experiment is supported by the staff of the Radiation Toxicology group: D. V. Tolle and S. M. Cullen, hematology; D. E. Doyle, data processing; T. E. Fritz and L. S. Lombard, pathology; and C. M. Poole and W. G. Keenan, veterinary care. G. L. Holmblad, Radiation Biology group, provides radiological assistance. Particular attention is being given to the structural and functional properties of the blood forming stem cell - the suspected cellular target for the effects.

Four preclinical stages of radiation-induced leukemia have been partially characterized. Stage II, "partial recovery," is prerequisite to leukemia development; it appears to be mediated by two processes working in concert, namely (1) acquired shifts in radiosensitivity of vital hematopoietic progenitors and (2) osteogenic-dependent restructuring of a hematopoietically conducive microenvironment. In contrast to these obligate regenerative hematopoietic changes observed in the long surviving leukemia-prone animals, dogs prone to aplastic anemia fail to exhibit such vital microenvironmental and targeted stem cell modifications under chronic irradiation.

---

*Summer 1982 participant in the Undergraduate Honors Program, Boise State College.

In working toward the elucidation of the molecular/cellular basis of the acquired shift in radiation sensitivity of the targeted stem cell, the following observations have been made. First, the response of isolated marrow progenitors from leukemia prone animals to graded doses of gamma irradiation, delivered in vitro, falls into distinct patterns of acquired radioresistance: pattern a, characterized by high dose tolerance; pattern b, characterized by low dose tolerance; and pattern c, tolerance of both high dose and low dose irradiation. Second, clonal acquisition of radioresistance appears not to be mediated by altered cell cycle kinetics, i.e., the selective enrichment of the targeted marrow population with progenitors having prolonged residence times in the more resistant S-phase of the cell-cycle. Third, acquired stem cell resistance appears more likely to involve modified cellular repair capacity as suggested in our split-dose type assays by the time-dependent recovery of clonogenic activity. Fourth, the modified repair capacity concept is supported by the ablative effects of high LET neutron irradiation on the acquired gamma ray resistance of stem cells isolated from leukemia-prone animals. The fifth and final observation involves the detection of repair-dependent clonal radioresistance of progenitors cloned in vitro under the selective pressure of continuous low dose rate (0.02 rad/min) gamma irradiation.

Interim conclusions drawn from these initial observations support the concept that modified repair capacity of marrow progenitors under continuous gamma irradiation provides the cellular basis of the process of acquired radioresistance that we have defined. This radioresistance is a pivotal process in the induction and development of the major hemopathologies seen under chronic ionizing radiation exposure. Accordingly, it is highly important to characterize fully this predisposing factor(s) of acquired radioresistance in order to provide the essential criteria necessary for early diagnosis and prognosis of leukemia and for preventive measures.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Antiviral Properties of Polyinosinic Acids Containing Thio and Methyl Substitutions
E. W. Chan, C. K. Lee, P. J. Dale, K. Nortridge, S. S. Hom, and T. M. Seed
J. Gen. Virol. 52, 291-299 (1981)

Cell Shape in the Alga Pediastrum (Hydrodictyaceae; Chlorophyta)
W. F. Millington, G. T. Chubb, and T. M. Seed
Protoplasma 105, 169-176 (1981)

1982

Hematologic Responses of Beagles Exposed Continuously to Low Doses of ^{60}Co γ-Radiation
T. E. Fritz, D. V. Tolle, D. E. Doyle, T. M. Seed, and S. M. Cullen
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 28, pp. 229-240

Sequential Changes in Bone Marrow Architecture during Continuous Low Dose Gamma Irradiation
T. M. Seed, G. T. Chubb, and D. V. Tolle
Scanning Electron Microscopy/1981/IV, O. Johari, R. M. Albrecht, and T. M. Seed, eds., SEM, Inc., AMF O'Hare (Chicago), IL, 1982, pp. 61-72

Early Effects of Castration and Replacement of Androgen on the Expression of Retrovirus-like Particles in Rat Ventral Prostate Epithelial Cells
T. M. Seed, M. Rubenstein, and K. M. Anderson
Proc. Soc. Exp. Biol. Med. 169, 470-479 (1982)

Leukemia Induction in Beagles Exposed Continuously to ^{60}Co γ-Irradiation: Hematology
D. V. Tolle, T. E. Fritz, T. M. Seed, S. M. Cullen, L. S. Lombard, and C. M. Poole
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 29, pp. 241-249

1983 through April

Phospholipid-Nucleoside Conjugates: The Aggregational Characteristics and Morphological Aspects of Selected 1-β-D-Arabinofuranosylcytosine 5'-Diphosphate-L-1,2-Diacylglycerols
M. MacCoss, J. J. Edwards, T. M. Seed, and S. P. Spragg
Biochim. Biophys. Acta 719, 544-555 (1982)

The Ultrastructure of Radiation-Induced Endosteal Myelofibrosis in the Dog
T. M. Seed, G. T. Chubb, D. V. Tolle, T. E. Fritz, C. M. Poole, D. E. Doyle, L. S. Lombard, and L. V. Kaspar
Scanning Electron Microscopy/1982/I, O. Johari, R. M. Albrecht, and T. M. Seed, eds., SEM Inc., AMF O'Hare (Chicago), IL, 1983, pp. 377-391

Hemopathologic Predisposition and Survival Time under Continuous Gamma Irradiation: Responses Mediated by Altered Radiosensitivity of Hemopoietic Progenitors
T. M. Seed, L. V. Kaspar, D. V. Tolle, and T. E. Fritz
Exp. Hematol. 10, Suppl. No. 12, 232-248 (1982)

Circulating Micromegakaryocytes Preceding Leukemia in Three Dogs Exposed to 2.5 R/day Gamma Radiation
D. V. Tolle, S. M. Cullen, T. M. Seed, and T. E. Fritz
Vet. Pathol. 20, 111-114 (1983)

Aberrant Megakaryocytopoiesis Preceding Radiation-Induced Leukemia in the Dog
D. V. Tolle, T. M. Seed, S. M. Cullen, C. M. Poole, and T. E. Fritz
Scanning Electron Microscopy/1982/I, O. Johari, R. M. Albrecht, and T. M. Seed, eds., SEM Inc., AMF O'Hare (Chicago), IL, 1983, pp. 367-376

8. LIFE SHORTENING, TUMOR INDUCTION, AND TISSUE DOSE FOR FISSION NEUTRON AND GAMMA RAY IRRADIATIONS

D. Grahn, L. S. Lombard, J. F. Thomson, F. S. Williamson, G. L. Holmblad, J. L. Hulesch, V. A. Ludeman, A. R. Sallese, E. F. Staffeldt, J. E. Trier, and B. J. Wright

This program obtains information on the late effects of whole body exposure to low doses of a high-LET and a low-LET ionizing radiation in experimental animals to provide guidance for the prediction of radiation hazards to man. This information takes the form of dose-response curves for life shortening from all causes of death and from numerous specific causes. The program is closely allied with that in Mammalian Genetics and Biostatistics (Chapter 9), in which genetic end points are examined using the same radiation sources, dose rates, total doses, and strain of mouse.

The experiments employ fission neutrons from the JANUS reactor and ^{60}Co gamma rays, delivered as single, weekly, or duration-or-life exposures covering the following range of doses and dose rates:

	Neutrons	Gamma Rays
Single exposures (rads)	1-240	22.5-788
Fractionated, 2-60 fractions (rads, total dose)	2-320	100-3820
Duration of life (rads/weekly fraction)	0.67-2.67	7-32
Continuous (rads/day)	--	1.3-6

In most experiments we have employed the house mouse, *Mus musculus*, specifically the C57BL/6 x BALB/c F_1 hybrid ($B6CF_1$). To provide an interspecies comparison, we have used the wild-type white footed mouse, *Peromyscus leucopus*, a cricetid rodent the same size as *Mus* but with a significantly longer life-span and a different spectrum of tumors.

Animals are observed for their lifetimes; gross and microscopic examinations of the decedents are carried out to establish the probable cause of death. The biological studies are supported and complemented by dosimetric analysis and data processing programs.

The status of current studies as of April 1983 is as follows:

JM-9 extends the neutron dose-response curves to doses below 20 rads and includes single doses of 1, 2.5, 5, 10, 20, and 40 rads of neutron radiation and 22.5, 45, and 90 rads of gamma radiation. This experiment is designed to determine whether the neutron dose-response curve for life

shortening becomes linear at some point below ~ 20 rads, or whether it continues to conform to the power function that fits the data from 20 to 240 rads. Of the 3950 mice committed to the experiment, 85 percent have died.

JM-10 involves the exposure of P. leucopus to a single exposure of 90, 143, 206, and 417 rads of gamma radiation, and 20, 40, 80, and 160 rads of neutron radiation, or fractionated neutron exposures of 40 and 160 rads in 24 once weekly fractions. Out of 2335 animals exposed, about 87 percent have died.

JM-12 is designed to study the effects of neutrons given in 2, 4, or 6 fractions at weekly intervals, compared with the same total dose (240 rads) given as a single exposure. All of the irradiated mice and 80% of the controls are dead. Previous experiments had shown that life shortening after this dose in a single exposure was less than that observed when the dose was divided among 6, 24, or 72 fractions given over a 21 to 24 week period. The phenomenon of augmentation of neutron effect by dose fractionation is now evident even with two fractions, and is most pronounced with four fractions. A study of the causes of death shows that the augmentation may be manifested somewhat differently for each fractionation regime. After two fractions, there is a marked increase in early deaths from thymic lymphomas; after four fractions, both thymic and nonthymic lymphomas appear soon after exposure; and after six fractions, there is an accelerated development of lung tumors.

JM-4L involves gamma radiation only, delivered at instantaneous dose rates of 1.4, 2.8, 6.3, and 12.6 millirads per minute, 22 hours per day, five days per week, for a total of 23 weeks. The total doses accumulated are 206, 417, 959, and 1918 rads, the same as in a previous series in which the mice were irradiated once a week. The purpose is to compare the effects of protracted gamma radiation at low dose rates with fractionated gamma radiation at high dose rates. Of a total of 620 mice, about 50 percent have died.

JM-13, supported by the Nuclear Regulatory Commission, involves weekly exposures to neutrons (0.033, 0.125, 0.225, 0.35, 0.5, and 0.67 rad/weekly fraction) or to gamma rays (1.67, 3.33, 5, 7.5, and 10 rads/weekly fraction) for a total of 60 fractions (half of the life-span of the mouse). This experiment, begun in February 1981, complements and extends the DOE-supported program and will determine the validity of present estimates of dose-response functions for diverse somatic and genetic effects of periodic exposures down to levels being experienced in the nuclear power industry.

We now can describe the dose-response curves for life shortening for most of the exposure schedules and doses given above, except for JM-13. Although some animals are still alive, the following conclusions are not likely to change materially:

1) As is known, fractionation or protraction of gamma irradiation results in longer mean survival times than are observed when the same total doses are given in a single exposure. Fractionation of neutron irradiation, in contrast, decreases the mean survival time relative to that observed after

single doses. This augmentation of injury by protraction of neutron exposure occurs at all total doses of 20 rads or more, and is expressed as an increment of increase in life shortening that is a constant proportion of the value induced by single doses, specifically, the ratio is 1.6 to 1 in $B6CF_1$ mice.

2) The developing data at low single doses of neutrons now indicate there is a final linear term to the dose-response function for life shortening. At neutron doses of 10 rads or less, the coefficient is tentatively estimated to be -5.2 days per rad. Data at doses of 20 rads and above conform to a linear-quadratic equation with a negative D^2 term and a least-squares intercept significantly greater than zero. Preliminary results from the JM-13 experiment would seem to confirm these findings that the response below 10 rads is linear, independent of dose rate, and rapidly rising. The ultimate maximum relative biological effectiveness (RBE) may depend, therefore, on the choice of life shortening coefficient for gamma irradiation. For single doses, the RBE may be about 12. For low intensity exposure over the adult lifetime, wherein the effect of gamma rays is only one-tenth that for single irradiations at high intensity, the RBE could approach 120.

3) The contribution of lymphoreticular tumors (and other tumors of connective tissue origin) to excess mortality is more pronounced with gamma rays than with neutrons; the opposite is true for lung tumors and other tumors of epithelial origin. Consequently, the RBE for specific tumor-related mortality will vary with tissue of origin. It also appears that pulmonary tumors make a significant contribution to the augmented life shortening from fractionated neutron exposures.

4) On the basis of either days of life lost per rad or percent of life-span lost per rad, P. leucopus is more sensitive to neutrons than is M. musculus. However, the excess mortality rates (averaged over the entire life-span) appear to be the same, and the RBE relationships do not appear appreciably different between the two mouse species.

Data from this program, in conjunction with the results from the Mammalian Genetics and Biostatistics program (see Chapter 9), are now being collated and reviewed for the existence of sensible patterns of injury from the two radiation qualities. Establishment of such patterns should provide a more complete understanding of the variation seen in the relative biological effectiveness of the fission neutron. A significant finding this year is the evidence that there is an initial linear relationship between neutron induced injury and dose that covers the range from 0 to about 10 rads. If this finding can be confirmed, then the enigmatic aspects of neutron-induced cell and tissue damage can be studied with greater precision.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Life Shortening in Mice Exposed to Fission Neutrons and Gamma Rays.
I. Single and Short-Term Fractionated Exposures
J. F. Thomson, F. S. Williamson, D. Grahn, and E. J. Ainsworth
Radiat. Res. 86, 559-572 (1981)

Life Shortening in Mice Exposed to Fission Neutrons and Gamma Rays.
II. Duration-of-Life and Long-Term Fractionated Exposures
J. F. Thomson, F. S. Williamson, D. Grahn, and E. J. Ainsworth
Radiat. Res. 86, 573-579 (1981)

1982

Studies on the Multistage Nature of Radiation Carcinogenesis
R. J. M. Fry, R. D. Ley, D. Grube, and E. Staffeldt
Carcinogenesis - A Comprehensive Survey, Vol. 7, Cocarcinogenesis and Biological Effects of Tumor Promoters, E. Hecker, N. E. Fusenig, W. Kunz, F. Marks, and H. W. Thielmann, eds., Raven Press, NY, 1982, pp. 155-165

1983 through April

RBE of Fission Neutrons for Life Shortening and Tumorigenesis
J. F. Thomson, L. S. Lombard, D. Grahn, F. S. Williamson, and T. E. Fritz
Radiation Protection: Neutron Carcinogenesis, EUR 8084 EN, J. J. Broerse and G. B. Gerber, eds., European Seminar jointly organized by the Commission of the European Communities, DG XII Biology, Rad. Protection and Medical Research and the Radiobiological Institute TNO, NL - Rijswijk, March 30-April 1, 1982, pp. 75-93

Life Shortening in Mice Exposed to Fission Neutrons and γ Rays:
III. Neutron Exposures of 5 and 10 Rad
J. F. Thomson, F. S. Williamson, and D. Grahn
Radiat. Res. 93, 205-209 (1983)

In press as of May 1, 1983

Lung Tumors in Mice
R. J. M. Fry and H. P. Witschi
Carcinogenicity, Mutagenicity and Other Short-Term Tests

9. MAMMALIAN GENETICS AND BIOSTATISTICS

D. Grahn, B. A. Carnes, M. L. Garriott, B. H. Farrington, C.-H. Lee,*
J. J. Russell, and D. M. Eychner†

The objectives of this program are threefold: (1) To assess genetic hazards from testicular burdens of ^{239}Pu in comparison with single, weekly, and continuous ^{60}Co gamma irradiation and single and weekly fission neutron irradiation to develop a basis for estimating relative biological effectiveness (RBE) for genetic end points. (2) To develop detailed dose-response data for genetic end points of concern at low doses of neutrons and gamma rays. (3) To develop improved statistical approaches to both the common and the unusual in chemical and radiation toxicology.

The genetic end points are: (1) the dominant lethal mutation rate in both spermatogonial and postspermatogonial cell stages, (2) the frequency of abnormal sperm head morphology measured at various times after irradiation, (3) the frequency of reciprocal chromosome translocations induced in spermatogonia and measured at first meiotic metaphase, and (4) the frequency of chromatin fragments (micronuclei) in the polychromatophilic erythrocyte in the bone marrow. Male hybrid $B6CF_1$ mice, 120 days old, are used for all studies. Measures of the retention, microdistribution, and microdosimetry of plutonium in the testes are obtained by conjoint histological and autoradiographic procedures after intravenous injection of monomeric plutonium citrate. Other males are exposed to external fission neutrons from the JANUS reactor and to ^{60}Co gamma radiation. Breeding tests are performed with the irradiated males at selected periods to obtain data on postmeiotic, meiotic, and premeiotic cell stages in male gametogenesis.

Single neutron doses of 1, 2.5, 5, 10, 20, and 40 rads have been compared with the effects of 22.5, 45, and 145 rads of ^{60}Co gamma rays, using the above genetic end points, and a weekly, long-term exposure series has also been performed for additional neutron and gamma ray comparisons at low weekly increments of 0.67, 1.67, and 2.67 neutron rads vs. 6.95, 17.4, and 32.0 gamma rads. Males were periodically screened for accumulation of dominant lethal mutations and for the frequency of chromosome aberrations. As noted in Chapter 8 of this report, an extensive study is under way for the Nuclear Regulatory Commission on the somatic and genetic effects of 60 once-weekly exposures to low neutron and gamma irradiations.

---

*Special term appointment.
†Summer 1982 participant in the Undergraduate Honors Program, State University of New York at Cortland.

The genetic tests evaluate the effects of 0.125, 0.35, and 0.67 rad/week of neutrons and 5, 7.5, and 10 rads/week of gamma rays. Genetic evaluations have been performed during the 60 weeks of exposure and after the exposure sequence has been completed to determine levels of residual genetic damage.

Genetic effects of gonadal burdens of plutonium. These studies have been completed and a final manuscript is in press. A limited continuing collaboration with the Mammalian Genetics program at Oak Ridge National Laboratory is providing dosimetric data on their mice to support their studies on the rate of induction by ^{239}Pu of specific genetic locus mutations and of dominant mutations causing abnormal skeletal development.

Genetic effects of the external radiations. In the single dose studies, there is a continuous linear response for the induction of dominant lethal mutations in postspermatogonial stages down to 5 rads. Below this level, there is a significant response in excess of linear prediction, which is about a factor of two above expectation. This excess level of response is noted in the rates of both postimplantation and preimplantation losses. Altogether, significant genetic damage is detected for all end points at 2.5 rads, and for all except abnormal sperm at 1 rad. In studies involving once weekly irradiations, males mated regularly during the 6-day radiation-free period show steady levels of dominant lethal mutation frequency that are a direct function of weekly dose rate and show only a small increase with long-term accumulated dose. The neutron irradiated mice show a mutation rate about 50 percent higher than predicted from single dose data, indicating that the frequency of lethal mutations is also augmented by protraction of the neutron exposure as is the case for life shortening (see Chapter 8). Gamma irradiated males show a 25 percent reduction of the dominant lethal mutation rate in the course of weekly exposures compared with single exposures. In the 60-week series for the Nuclear Regulatory Commission, a steady level of weekly dose-rate dependent damage is seen in the frequencies of lethal mutations, micronuclei, and abnormal sperm. This latter response is extremely sensitive to low doses, as a significant effect is seen at 0.125 rad/week of neutrons. The levels of response for all three end points appear to equilibrate after about 10 weeks of exposure and remain nearly constant, relative to the control, throughout the full exposure period of 60 weeks.

Biostatistics. Among a number of programs in this Division and others at Argonne given statistical assistance, several programs have required the development of unusual approaches. These are listed below.

In collaboration with the above genetic studies, important sources of variability were identified and dose-response models were developed for genetic end points at low doses of neutrons and gamma rays in mice. Statistical strategies and collaborative interpretations were provided for C. Peraino in determining whether phenotypic characteristics of rat liver foci could provide predictive information for tumor formation. Statistical analyses were conducted for T. E. Fritz to evaluate the late effects of low doses of ionizing radiation in the dog. A simulation study was generated with M. J. Peak for evaluating mutagenicity decision rules for the effects of nonionizing solar radiation in bacterial genetic systems.

In the area of theoretical applied statistics, Monte Carlo simulations were conducted to compare the behavior of biased regression procedures and determine when a particular biased estimation procedure should be implemented.

In summary, the collation of data in conjunction with the studies on long-term somatic effects are revealing interesting patterns of RBE values in regard to end point and cell stages at risk in the male germ line. In general, the RBE's for genetic effects are less than those for long-term somatic effects, probably because the latter reflect multisystemic pathological interactions that amplify damage. Genetic injury is not subject to as much complication by systemic interactions and can also be categorized by specific cell stage factors. Meiotic and postmeiotic stages, for example, show a narrower spread and lower limit to the greater effectiveness of high LET radiations because of the more restricted opportunities for the repair of genetic damage.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS*

1981

Quantifying General Population Response to Environmental Lead
C. D. Brown
Environmetrics 81, Summaries of Conference Presentations [expanded abstracts], Society for Industrial and Applied Mathematics, 1981, pp. 14-15

Multivariate Methods for Assessing Disease/Environment Association: Conceptual Versus Geographic Regions
B. A. Carnes, M. E. Ginevan, and J. J. Collins
Environmetrics 81, Summaries of Conference Presentations [expanded abstracts], Society for Industrial and Applied Mathematics, 1981, pp. 212-213

The Use of a Demographic Model for Health Risk Assessment
J. J. Collins, M. E. Ginevan, D. Grahn, and R. T. Lundy
Environmetrics 81, Summaries of Conference Presentations [expanded abstracts], Society for Industrial and Applied Mathematics, 1981, pp. 220-221

*Included in this listing are publications arising from the work of the group entitled Human Health Effects and Risk Assessments, J. J. Collins and M. E. Ginevan, Principal Investigators. The group completed its activities September 30, 1982.

Computer Mapping of Birth Weights as an Approach to Environmental Epidemiology
J. R. B. Curtiss
Computer Mapping Applications in Urban, State and Federal Government: Plus Computer Graphics in Education, Vol. 16, 1981, P. A. Moore, ed., Harvard Library of Computer Graphics/1981 Mapping Collection, Laboratory for Computer Graphics and Spatial Analysis, Harvard Graduate School of Design, Cambridge, MA, pp. 77-82

Human Birth Weight Patterns as an Indicator of Populations Subject to Environmental Stress
J. R. B. Curtiss, M. E. Ginevan, and C. D. Brown
Environmetrics 81, Summaries of Conference Presentations [expanded abstracts], Society for Industrial and Applied Mathematics, 1981, pp. 222-223

Mortality in 191 Counties near Energy Facilities: Use of Economic and Social Variables for Standardization
D. K. Dixon-Davis, J. J. Collins, and D. Grahn
Environ. Res. 25, 106-125 (1981)

A Poisson Trials Approach to Interpopulation Comparisons of Cause of Death Data
M. E. Ginevan
Environ. Res. 25, 147-159 (1981)

Reply to R. Bertell (Letter to the Editors)
M. E. Ginevan
Health Phys. 41, 422-424 (1981)

Adult Leukemia Risk and Diagnostic X-Rays: A Reanalysis of the Tri-State Leukemia Survey Data
M. E. Ginevan, J. R. B. Curtiss, and D. Grahn
NUREG Report NUREG/CR-2234; ANL Report, ANL-81-37 (June 1981)

1982

Some Comments on Niche Analysis in Canonical Space
B. A. Carnes, and N. A. Slade
Ecology 63, 888-893 (1982)

The Contribution of Medical Measures to the Decline of Mortality from Respiratory Tuberculosis: An Age-Period-Cohort Model
J. J. Collins
Demography 19, 409-427 (1982)

Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume III, SPAHR Interactive Package Guide
J. J. Collins
NUREG/CR-2364 Volume III, ANL-81-59 Volume III (1982)

Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume II, SPAHR Introductory Guide
J. J. Collins and R. T. Lundy
NUREG/CR-2364 Volume II, ANL-81-59 Volume II (1982)

Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume IV, SPAHR Users Guide
J. J. Collins and R. T. Lundy
NUREG/CR-2364 Volume IV, ANL-81-59 Volume IV (1982)

Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume V, SPAHR Programmer's Guide
J. J. Collins and R. T. Lundy
NUREG/CR-2364 Volume V, ANL-81-59 Volume V (1982)

Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume I, Introduction to the SPAHR Demographic Model for Health Risk
J. J. Collins, R. T. Lundy, D. Grahn, and M. E. Ginevan
NUREG/CR-2364 Volume I, ANL-81-59 Volume I (1982)

Spatio/Temporal Analyses of Human Birth Weight: An Indicator of Subtle Environmental Stress?
J. R. B. Curtiss, M. E. Ginevan, and C. D. Brown
Health Impacts of Different Sources of Energy, IAEA International Symposium on Health Impacts of Different Sources of Energy, Nashville, TN, June 22-26, 1981, International Atomic Energy Agency, Vienna, 1982, pp. 33-50

Neutron and γ-Ray Effects Measured by the Micronucleus Test
M. L. Garriott and D. Grahn
Mutat. Res. Lettr. 105, 157-162 (1982)

1983 through April

In press as of May 1, 1983

A Demographic Model for Performing Site-Specific Health Risk Projections
J. J. Collins, R. T. Lundy, and D. Grahn
Health Phys.

AET Reduces the Frequency of Micronuclei in Bone Marrow Cells of Mice Exposed to γ-Radiation
M. L. Garriott and D. T. Crowe
Radiat. Res.

Genetic Risks Associated with Radiation Exposures during Space Flight
D. Grahn
Adv. Space Research (Life Sciences and Space Research XX)

Morphologic and Phenotypic Analysis of an Outcross Line of Blotchy Mouse
V. Ranga, D. Grahn, and T. M. Journey
Exp. Lung Res.

Interpretation of Cytogenetic Damage Induced in the Germ Line of Male Mice Exposed for Over One Year to ^{239}Pu Alpha Particles, Fission Neutrons, or ^{60}Co γ-Rays
D. Grahn, C. H. Lee, and B. F. Farrington
Radiat. Res.

Enhanced Iron Removal from Liver Parenchymal Cells in Experimental Iron Overload. Liposome Encapsulation of HBED and Phenobarbital Administration
Y. E. Rahman, E. A. Cerny, E. H. Lau, and B. A. Carnes
Blood

MOLECULAR BIOLOGY SECTION

Molecular Anatomy Group, N. G. Anderson Principal Investigator

Chapter 10. Molecular Anatomy Program: The Human Protein Index

Molecular Biophysics Group, M. Schiffer Principal Investigator

Chapter 11. Structural Studies of Proteins

Mutagenesis Group, H. E. Kubitschek Principal Investigator

Chapter 12. Cellular and Molecular Mechanisms of Cell Growth

Photobiology Group, M. J. Peak Principal Investigator

Chapter 13. Molecular and Cellular Effects of Solar Ultraviolet Light

10. MOLECULAR ANATOMY PROGRAM: THE HUMAN PROTEIN INDEX

N. G. Anderson, N. L. Anderson, J. J. Edwards, C. S. Giometti, J. Taylor, K. W. Willard-Gallo, M. A. Gemmell, S. L. Nance, and S. L. Tollaksen

The objectives of this program are to (1) develop methods for the detection of genetic variants in humans which may be due to radiation or to chemical mutagens; (2) characterize, at the protein level, alterations in gene expression due to environmental pollutants, including chemical carcinogens; and (3) characterize changes due to nonmutagenic, noncarcinogenic toxic agents in terms of alterations in gene expression or protein distribution. The effects of all three classes of agents (mutagens, carcinogens, and toxic agents) are now detectable and measurable by quantitative high-resolution two-dimensional electrophoresis. Approximately one-third of mutations involving single amino acid substitutions produce detectable alterations in the charge of a protein gene product; tumor cells exhibit alterations in gene expression that change the protein makeup of cells; and toxic agents may alter gene expression, and/or produce cell leakage (including protein leakage). Hence all three objectives require the development of high-resolution protein analysis systems and methods applicable to relatively large numbers of samples. Also needed are ancillary methods for the identification of as many of the proteins found in the maps as possible.

The long-term goal of the program described above is the development of an index of the proteins seen (The Human Protein Index) and of both the genetic variants and systematic alterations in gene expression found (The Human Protein Data Base).

The objectives can be reached only by using the highest resolution systems for protein separation and analysis currently available; these now center around two-dimensional electrophoresis, with computerized image analysis and data reduction. We summarize below our recent progress.

Human lymphocytes have been separated by cell sorting and other methods, and the major proteins of the subsets obtained have been identified, including the proteins of the cytoskeleton. In other studies of human leukemic cells, specific differences have been noted between the malignant cells and normal cells. In addition it has been shown that the relatively well-differentiated T and B-cell chronic lymphocytic leukemias retain expression patterns characteristic of normal T and B lymphocytes as regards a panel of over 30 marker proteins.

Fibroblasts from 63 individuals (most with known genetic diseases) were examined and 89 major proteins, easily and reproducibly located on the two-dimensional patterns, were intercompared. A total of 11 candidates

for charge change protein variants were discovered. Eight appeared in only one individual or family and were therefore "rare" variants, while one variant was found in three individuals and is considered "common." Thus, of this limited initial panel of 89 proteins, 9% were "rare" variants, and only one "common" polymorphism was found. The average chance that a given protein (from the group of proteins examined) would be detected as charge-heterozygous in a given individual is about 0.2%, indicating very little genetic variation of cellular proteins as compared with the plasma proteins previously studied.

A candidate charge variant of human smooth muscle tropomyosin, found in fibroblasts, was chosen for more extensive analysis and as a model study of how a candidate variant may be demonstrated to be a variant of a spot identified as the "wild-type" protein. Both proteins (Tm:3 and variant Tm:3.1) copurify together during isolation, shift to higher apparent molecular weights on sodium dodecyl sulfate electrophoresis in the presence of 4 M urea, are components of microfilaments, and both lack proline and tryptophan. They vary only in charge. Quantitative computerized analysis showed that the sum of Tm:3 and Tm:3.1 in the heterozygous cells equals the amount of Tm:3 in the homozygous cells.

The TYCHO II image analysis and data reduction system has been used for a systematic study of resolution of two-dimensional protein patterns. Depending on the size of the pattern, the theoretical resolution varied from 15,000 proteins for 7 x 7 inch gels to approximately 50,000 for 12 x 12 inch gels. Positional accuracy on the processed patterns for the majority of proteins was less than a spot width (0.5 mm to 2.0 mm). Computerized image analysis can now be used to select and optimize gel systems and methods systematically, and to select optimally resolved sets of proteins for genetic and other quantitative studies.

Relative to the development of improved laboratory techniques, an improved 8 x 10 inch two-dimensional gel system was designed, and substantial progress toward machine manufacturable gels was made. Such gels are a prerequisite for large scale applications of two-dimensional electrophoresis technology, as well as for ultimate interlaboratory standardization.

It is now technically feasible to search for large numbers of human protein genetic variants, and to systematically validate their relationship to wild-type proteins. This capability opens up the possibility of global analysis of the differential sensitivity of human structural genes to mutation by radiation and chemical mutagens.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Identification of Mitochondrial Proteins and Some of Their Precursors in Two-Dimensional Electrophoretic Maps of Human Cells
L. Anderson
Proc. Natl. Acad. Sci. USA 78, 2407-2411 (1981)

The Human Protein Index: Relationship to Genetic Engineering
N. G. Anderson and L. Anderson
Proceedings of the 1981 Battelle Conference on Genetic Engineering, Reston, VA, April 6-10, 1981, M. Keenberg, ed., Vol. IV, 1981, pp. 163-170

The Human Protein Index (Photo/Essay)
N. G. Anderson and N. L. Anderson
J. Am. Med. Assoc. 246, 2620-2621 (1981)

High Resolution Two-Dimensional Electrophoretic Mapping of Immunoglobulin Light Chains
N. L. Anderson
Immunology Lett. 2, 195-199 (1981)

The Potential of High Resolution Protein Mapping as a Method of Monitoring the Human Immune System
N. L. Anderson and N. G. Anderson
Biological Relevance of Immune Supression as Induced by Genetic, Therapeutic and Environmental Factors, J. H. Dean and M. Padarathsingh, eds., 1981, Van Nostrand Reinhold Co., New York, Chapter 9, pp. 136-147

The TYCHO System for Computer Analysis of Two-Dimensional Gel Electrophoresis Patterns
N. L. Anderson, J. Taylor, A. E. Scandora, B. P. Coulter, and N. G. Anderson
Clin. Chem. 27, 1807-1820 (1981)

The Nature of Observed Schlieren Patterns in Isoelectric Focusing Gels and Their Use for Position Location of Banded Proteins
J. J. Edwards and N. G. Anderson
Electrophoresis 1981 2, 161-168 (1981)

Proteins of Human Semen. I. Two-Dimensional Mapping of Human Seminal Fluid
J. J. Edwards, S. L. Tollaksen, and N. G. Anderson
Clin. Chem. 27, 1335-1340 (1981)

Muscle Protein Analysis. III. Analysis of Solubilized Frozen-Tissue Sections by Two-Dimensional Electrophoresis
C. S. Giometti and N. G. Anderson
Clin. Chem. 27, 1918-1921 (1981)

A Variant of Human Nonmuscle Tropomyosin Found in Fibroblasts by Using Two-Dimensional Electrophoresis
C. S. Giometti and N. L. Anderson
J. Biol. Chem. 256, 11840-11846 (1981)

The Use of Carbamylated Charge Standards for Testing Batches of Ampholytes Used in Two-Dimensional Electrophoresis
S. L. Tollaksen, J. J. Edwards, and N. G. Anderson
Electrophoresis 1981 2, 155-160 (1981)

Two-Dimensional Analysis of Human Lymphocyte Proteins. I. An Assay for Lymphocyte Effectors
K. E. Willard and N. G. Anderson
Clin. Chem. 27, 1327-1334 (1981)

1982

The Human Protein Index
N. G. Anderson and L. Anderson
Clin. Chem. 28, 739-748 (1982)

The Human Protein Index
N. G. Anderson and N. L. Anderson
J. Am. Med Assoc. 246 2620-2621 (1981)

The Human Protein Index: A Prospect for Laboratory Medicine
N. G. Anderson and N. L. Anderson
Lab. Manage. 20, 25-32 (1982)

The Human Protein Index Project and the Molecular Pathology Data Base
N. G. Anderson and N. L. Anderson
Med. Lab. 11, 75-94 (1982)

Proteins of Human Milk. I. Identification of Major Components
N. G. Anderson, M. T. Powers, and S. L. Tollaksen
Clin. Chem. 28, 1045-1055 (1982)

Towards a Complete Catalog of Human Proteins
N. L. Anderson
Trends in Anal. Chem. 1, 131-134 (1982)

A Two-Dimensional Electrophoretic Analysis of the Heat-Shock-Induced Proteins of Human Cells
N. L. Anderson, C. S. Giometti, M. A. Gemmell, S. L. Nance, and N. G. Anderson
Clin. Chem. 28, 1084-1092 (1982)

Specific Antiserum Staining of Two-Dimensional Electrophoretic Patterns of Human Plasma Proteins Immobilized on Nitrocellulose
N. L. Anderson, S. L. Nance, T. W. Pearson, and N. G. Anderson
Electrophoresis 3, 135-142 (1982)

The TYCHO System for Computer Analysis of Two-Dimensional Gel Electrophoresis Patterns
N. L. Anderson, J. Taylor, A. E. Scandora, B. P. Coulter, and N. G. Anderson
Clin. Chem. 27, 1807-1820 (1981)

Proteins of Human Urine. II. Identification by Two-Dimensional Electrophoresis of a New Candidate Marker for Prostatic Cancer
J. J. Edwards, N. G. Anderson, S. L. Tollaksen, A. C. von Eschenbach, and J. Guevara, Jr.
Clin. Chem. 28, 160-163 (1982)

Proteins of Human Urine. III. Identification and Two-Dimensional Electrophoretic Map Positions of Some Major Urinary Proteins
J. J. Edwards, S. L. Tollaksen, and N. G. Anderson
Clin. Chem. 28, 941-948 (1982)

Lymphocyte, Monocyte, and Granulocyte Proteins Compared by Use of Two-Dimensional Electrophoresis
M. A. Gemmell and N. L. Anderson
Clin. Chem. 28, 1062-1066 (1982)

Muscle Protein Analysis by Two-Dimensional Gel Eletrophoresis
C. S. Giometti
Critical Reviews in Clinical Laboratory Sciences, Vol. 18, Issue 1, J. Batsakis and J. Savory, eds., CRC Press, Inc., Boca Raton, FL., 1982, pp. 79-109

Muscle Protein Analysis. III. Analysis of Solubilized Frozen-Tissue Sections by Two-Dimensional Electrophoresis
C. S. Giometti and N. G. Anderson
Clin. Chem. 27, 1918-1921 (1981)

A Variant of Human Nonmuscle Tropomyosin Found in Fibroblasts by Using Two-dimensional Electrophoresis
C. S. Giometti and N. L. Anderson
J. Biol. Chem. 256, 11840-11846 (1981)

Cytoskeletal Proteins from Human Skin Fibroblasts, Peripheral Blood Leukocytes, and a Lymphoblastoid Cell Line Compared by Two-Dimensional Gel Electrophoresis
C. S. Giometti, K. E. Willard, and N. L. Anderson
Clin. Chem. 28, 955-961 (1982)

A Simple Method of Eluting Proteins from Two-Dimensional Gels
J. Guevara, Jr., E. A. Chiocca, F. C. Clayton, A. C. von Eschenbach, and J. J. Edwards
Clin. Chem., 28, 756-758 (1982)

Design and Implementation of a Prototype Human Protein Index
J. Taylor, N. L. Anderson, A. E. Scandora, Jr., K. E. Willard, and N. G. Anderson
Clin. Chem. 28, 861-866 (1982)

Two-Dimensional Analysis of Human Lymphocyte Proteins II. Regulation of Lymphocyte Proteins by a Molecule Present in Normal Human Urine
K. E. Willard
Clin. Chem. 28, 1074-1083 (1982)

Two-Dimensional Analysis of Human Lymphocyte Proteins: III. Preliminary Report on a Marker for the Early Detection and Diagnosis of Infectious Mononucleosis
K. E. Willard
Clin. Chem. 28, 1031-1035 (1982)

Two-Dimensional Electrophoretic Analysis of Human Leukocyte Proteins from Patients with Rheumatoid Arthritis
K. E. Willard, A. K. Thorsrud, E. Munthe, and E. Jellum
Clin. Chem. 28, 1067-1073 (1982)

Introduction [to a special issue of Clinical Chemistry]
D. S. Young and N. G. Anderson
Clin. Chem. 28, 737-738 (1982)

1983 through April

Adult-Onset Nemaline Rods in a Patient Treated for Suspected Dermatomyositis
M. J. Danon, C. S. Giometti, J. R. Manaligod, O. H. Perurena, and J. L. Skosey
Arch. Neurol. 38, 761-766 (1981)

Phospholipid-Nucleoside Conjugates: The Aggregational Characteristics and Morphological Aspects of Selected 1-β-D-Arabinofuranosylcytosine 5'-Diphosphate-L-1,2-Diacylglycerols
M. MacCoss, J. J. Edwards, T. M. Seed, and S. P. Spragg
Biochim. Biophys. Acta 719, 544-555 (1982)

In press as of May 1, 1983

High Resolution Protein Separation and Identification Methods Applicable to Virology
N. G. Anderson
Proceedings of the 7th Munich Symposium on Microbiology, Munich, Germany, June 2-3, 1982

Introduction to High Resolution Two-Dimensional Electrophoresis in Clinical Chemistry
N. G. Anderson
5th Conference on Perspective Biology, Nancy, France, October 3-4, 1982

Use of High-Resolution Two-Dimensional Electrophoresis in Human Health Effects Studies
N. G. Anderson and N. L. Anderson
Symposium on Health Risk Analysis, Gatlinburg, TN, October 27-30, 1980

Studies of Gene-Expression in Human Lymphocytes Using High-Resolution Two-Dimensional Electrophoresis
N. L. Anderson
Electrophoresis '81

Use of Two-Dimensional Electrophoresis to Study the Comparative Molecular Anatomy of Human Cell Types
N. L. Anderson
5th Conference on Perspective Biology, Nancy, France, October 3-4, 1982

Analysis of Human Leukemic Cells by Use of High-Resolution Two-Dimensional Electrophoresis. I: Results of a Pilot Study
N. L. Anderson, J. C. Wiltsie, C. Y. Li, K. W. Willard-Gallo, R. P. Tracy, D. S. Young, M. T. Powers, and N. G. Anderson
Clin. Chem.

Analysis of Proteins from Human Muscle Biopsies by Two-Dimensional Electrophoresis
C. S. Giometti, M. J. Danon, and N. G. Anderson
Neurology

A Computerized System for Matching and Stretching Two-Dimensional Gel Patterns Represented by Parameter Lists
J. Taylor, N. L. Anderson, and N. G. Anderson
Electrophoresis '81

Applications of Two-Dimensional Gel Electrophoresis in the Clinical Laboratory
R. P. Tracy and N. L. Anderson
Clin. Lab.

11. STRUCTURAL STUDIES OF PROTEINS

M. Schiffer, C. S. Borso, C.-H. Chang, M. T. Short, P. Thiyagarajan, C. F. Ainsworth, F. A. Westholm, R. M. Hyslop,* R. D. Benioff,† and J. A. Guzik‡

This program consists of two related projects. The overall goal of the first, structural studies of immunoglobulins (M. Schiffer), is to understand the function and specificity of antibodies at the molecular level. Our initial emphasis has been on determining the molecule structure of dimers formed by immunoglobulin light chains, Bence-Jones proteins, which are excellent models of intact antibody molecules. To study the effect of antigen-antibody interaction on the structure of the antibody binding site, work has been initiated on functional antibodies against protein antigens (rheumatoid factors, which are autoantibodies against IgG proteins, and anti-idiotypic antibodies prepared by hybridoma techniques). The objectives of the second project, neutron and X-ray scattering of biological molecules (C. S. Borso), are to develop instrumentation for neutron and X-ray small angle scattering for studying the structures of biological molecules in solution.

Structural studies of immunoglobulins. X-ray diffraction techniques are used to determine the precise three-dimensional structure of immunoglobulins in the crystalline state. The results are complemented by data obtained from gel filtration and small-angle neutron and X-ray scattering studies of antibody molecules in dilute solution.

In the last year, we adapted high pressure gel permeation chromatography (HPGPC) for the study of protein association. Compared with the conventional gel filtration technique used previously, the new procedure requires less material (5 μg vs. 0.5 to 10 mg) and is much faster (5 hours vs. 24 hours). The elution profiles of self-associating κ_I-Bence-Jones proteins are similar in the two systems. The HPGPC technique was used to demonstrate the binding of the anti-idiotypic antibody F6 to T15 protein.

In our study of Bence-Jones proteins, we have now characterized and purified 50 different proteins, and Dr. A. Solomon (University of Tennessee) has characterized them clinically and immunochemically. We were success-

---

*Faculty Research Participant, Illinois Benedictine College.
†Summer 1982 participant in the Undergraduate Honors Program, University of Illinois at Urbana.
‡Summer 1982 participant in the Undergraduate Honors Program, Cornell College.

ful in growing crystals in forms suitable for X-ray diffraction studies from κ_{II}-type protein Fin, λ_I-type proteins Cle and Loc, and from the variable fragment derived from κ_I-type protein Wat. The crystallographic refinement of the Mcg λ_V-type Bence-Jones dimer has been continued. Variation in the individual temperature factors lowered the R factor to 22.8%. Potential water molecules and residues incorrectly placed or disordered were identified.

The electron density map of λ_I-type Bence-Jones protein Loc was calculated using 3.5 Å data. The protein phases were determined by Iterative Single Isomorphous Replacement, in collaboration with B.-C. Wang (University of Pittsburgh). The electron density map was interpreted by finding the best fit of the individual variable and constant domains of the Mcg protein to the electron density of the Loc protein. Methods used were visual and computer fit of the backbones to the electron density and "rigid body" refinement of the domain positions. The agreement factor R = 34%. The two chains of the Loc light chain dimer form a molecule similar to an Fab fragment, and to the Mcg light chain dimer. The "elbow-bend" for the Loc protein is 97°, the smallest yet observed for light chain dimer or Fab fragment. The relative positions of the two C domains in the molecule and their packing in the crystal are similar to those found in the Mcg protein.

The antigen binding site formed by the two variable domains of the Loc light chain dimer is different from that of any other Fab or light chain studied thus far. This difference in the binding site conformation is caused not only by changes in the complementarity determining regions, which no longer appear to form a cavity, but primarily by differences in the relative positions and interactions of the variable domains.

The structure of protein Loc suggests that binding sites of Fab fragments could be different from those observed until now. Continuing structural studies to determine details of amino acid interactions will give insights into whether the mode of packing of the Loc protein is caused by special interactions of the complementarity determining regions.

X-ray small angle scattering studies. A unique self-scanning photodiode array has been developed as a linear position sensitive detector for studies of biological structures. This detector has several advanced features: 25 μM linear resolution, 100:1 aspect ratio, and count rate capabilities of 10^8 to 10^9 photons per cm^2 per second. The excellent linear resolution and large aspect ratio of the detector, coupled with Kratky collimation, have enabled significant improvements in signal-to-noise to be attained, thereby greatly increasing data throughput. Industrial Research and Development has recognized the significance of this development for a wide range of X-ray scattering and diffraction applications by presenting a 1982 IR-100 award for the design of this instrument.

Improved data acquisition techniques have been developed to enable this detector to be implemented at high flux synchrotron sources to study structural changes in macromolecules on a millisecond time scale in real time. This provides the first direct method for obtaining structural information to study kinetics on this time scale.

A detailed study of the structure of detergent-solubilized rhodopsin has been completed using the photodiode array detector. The results suggest that rhodopsin is somewhat more compact than previously thought, with most of the protein within the lipid bilayer. The maximal dimension of the molecule is 60 Å and the molecule is disposed slightly asymetrically with respect to the rod outer segment membrane. More of the protein extends into the cytoplasmic space than into the intradiscal interior.

Studies have been initiated on yeast inorganic pyrophosphatase to relate enzymatic activity, divalent metal ion binding, and conformational changes.

Small angle scattering has been used in association with high performance liquid chromatography to study the self-association of calmodulin in solution and its relation to conformational changes observed with calcium binding by this important regulatory molecule.

Neutron time-of-flight small angle diffractometer. A time-of-flight small angle neutron instrument has been developed at Argonne's Intense Pulsed Neutron Source, IPNS-I, to provide a unique facility for small angle studies requiring both epithermal and thermal neutrons. This small angle facility is the world's first instrument to utilize neutrons from a pulsed spallation neutron source successfully. User-friendly instrumentation is now available for use by the outside scientific community for biological structural studies.

A new technique employing resonant neutron small angle scattering has been developed to utilize the unique spectrum of neutrons available from this source. Various aminopolycarboxylate bifunctional chelating compounds have been synthesized to label specific amino acids of selected proteins. Resonant isotopes have been attached to calmodulin as a probe for studying its interactions with calcium-mediated enzymes.

In summary, several unique X-ray and neutron small angle scattering instruments are now available for studying the structures of biological molecules in solution. The advanced features of these instruments have already provided a means of obtaining new information on a wide variety of macromolecules. These instruments yield complementary information that cannot be obtained by other techniques. While they have been developed specifically to study many of the molecules involved in cellular regulation, the techniques developed will be suitable for studying a wide variety of biological macromolecules.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Computer Simulation of Protein Self-Association during Small-Zone Gel Filtration
F. J. Stevens and M. Schiffer
Biochem. J. 195, 213-219 (1981)

Characterization and Preliminary Crystallographic Data on the V_L-related Fragment of the Human κI Bence Jones Protein Wat
F. J. Stevens, F. A. Westholm, N. Panagiotopoulos, M. Schiffer, R. A. Popp, and A. Solomon
J. Mol. Biol. 147, 185-193 (1981)

Preliminary Crystallographic Data on the Human κIII Bence Jones Protein Dimer Cle
F. J. Stevens, F. A. Westholm, N. Panagiotopoulos, A. Solomon, and M. Schiffer
J. Mol. Biol. 147, 179-183 (1981)

1982

Optimization of Monolithic Solid State Array Detectors for the Position Encoding of Small Angle X-Ray Scattering from Synchrotron Sources
C. S. Borso
Nucl. Instrum. Methods 201, 65-71 (1982)

Small-Angle Scattering at a Pulsed Neutron Source: Comparison with a Steady-State Reactor
C. S. Borso, J. M. Carpenter, F. S. Williamson, G. L. Holmblad, M. H. Mueller, J. Faber, Jr., J. E. Epperson, and S. S. Danyluk
J. Appl. Cryst. 15, 443-448 (1982)

Small-Angle Neutron Scattering Study of Bence-Jones Protein Mcg: Comparison of Structures in Solution and in Crystal
M. Schiffer, F. J. Stevens, F. A. Westholm, S. S. Kim, and R. D. Carlson
Biochemistry 21, 2874-2878 (1982)

1983 through April

Implementation of Self-Scanning Photodiode Arrays for Scattering Applications Using Conventional and Intense X-Ray Sources
C. S. Borso
Trans. Am. Crystallogr. Assoc. 18, 141-148 (1982)

Structural Studies of λ_I-Bence-Jones Protein Loc
M. Schiffer, C.-H. Chang, M. Short, F. Westholm, and A. Solomon
Fed. Proc. 42, 708 (1983)

High Pressure Gel Permeation Chromatography in the Study of Self-Associating Immunoglobulin κ_I Light Chains and an Idiotype:Anti-Idiotype Complex
M. T. Short, F. J. Stevens, and M. Schiffer
Fed. Proc. 42, 931 (1983)

In press as of May 1, 1983

Automated Fail Safe Control System for Independent Operation of Two Radiation Shutters on a Rotating Anode X-Ray Source
M. T. Short, W. J. Eisler, Jr., and F. J. Stevens
J. Appl. Crystallogr.

12. CELLULAR AND MOLECULAR MECHANISMS OF CELL GROWTH

H. E. Kubitschek, W. W. Baldwin,* R. Graetzer,† and D. Williams-Hill

This program is concerned with the elucidation of cellular and molecular mechanisms of control of cell growth that are common to both prokaryotes and eukaryotes. It has led to the proposal of a new model for cell growth in the bacterium *Escherichia coli*: bilinear growth during the cell cycle with coordinate control of increase in cell mass and surface area. The primary objectives are to test this model for *E. coli*, and to determine relevancy to eukaryotic cell growth.

Conventional methods of cell biology and biochemistry are employed in studies of the growth and division cycle. Cell mass or volume increase and cell division are the fundamental features that define the cell growth cycle and establish cell ages at which other cell properties are measured. Synchronous cultures are produced by the Mitchison-Vincent method of separation of cells by velocity sedimentation in density gradients and selection of the smallest cells for synchronous growth. Cell volume increase is determined with a precision Coulter counter-Davidson analyzer cell sizing system of high resolution or, alternatively, cell size and DNA content are determined by flow cytometry. Values of cell buoyant densities, required to calculate cell mass from volume, are obtained by equilibrium centrifugation in Percoll gradients.

Last year, a breakthrough was made on bacterial cell growth that may lead to a new understanding of cell growth and its controls. Using data from electron micrograph measurements of thousands of growing cells of *E. coli*, measured by Woldringh and his co-workers, we found that both cell volume and surface area increase bilinearly during the cell cycle, i.e., rates of increase are initially constant and then double at the same point in the cycle. These results agree with and extend our earlier observations on linear cell growth in bacteria and eukaryotic cells. The fact that very different kinds of cells have similar patterns suggests that very similar mechanisms exist for controlling cell growth. In addition, our analysis of bacterial growth provides the first evidence for closely linked biochemical pathways controlling increase in cell mass and surface enlargement.

---

*Scientist in Residence (Faculty Research Leave at Argonne Appointee), Indiana University School of Medicine, Northwest Center, Gary.
†Faculty Research Participant, Pennsylvania State University.

More recent experiments have concerned cell buoyant densities, because knowledge of these densities is necessary to convert values of cell volume to the more fundamental measurement of cell mass. We determined the buoyant densities of cells in exponentially growing cultures of E. coli B/r NC32 by equilibrium centrifugation in Percoll gradients, and discovered that the mean cell densities were extremely constant during the cell cycle, and that densities varied little from cell to cell (coefficient of variation, 0.15%). We also found that buoyant densities of cells were unchanged even when cells were grown at widely different growth rates, from 0.5 to 2.5 divisions per hour. This result was surprising because bacterial volumes increase by a factor of 5 over this range, and RNA contents by even larger factors. On the other hand, when growth of these bacterial cultures had passed the exponential phase and entered the stationary phase, cell densities abruptly increased by about 1%. These experimental results provide evidence for the existence of well-controlled buoyant density states in these cells.

Evidence also was obtained that cell density is extremely constant in suspension cultures of each of three different mouse cell lines, myeloma S107 and lymphomas 70 Z/3 and Abe 8. Although the mean buoyant densities of the three cell lines differed significantly, with values of 1.0678, 1.0622, and 1.0540, respectively, these mean densities were independent of the phase of the cell cycle, and the ranges of density values were narrow for each cell line, with a coefficient of variation of about 0.3% (in collaboration with M. R. Loken, University of Chicago).

Our discovery of the extreme and highly regulated constancy of buoyant cell density in these prokaryotic and eukaryotic cells provides evidence for a new and heretofore unexpected control system, that of buoyant density control. In addition, our results lead to the following conclusions.

Cell mass is readily determined from measurements of cell volume throughout the cell cycle in E. coli and the murine cultures.

The constancy of buoyant density during the cycle makes unlikely earlier models of cell division that proposed that density variation may provide a signaling function for cell division.

The constancy of buoyant density provides further evidence for similar mechanisms of control of growth in prokaryotes and eukaryotes and, indeed, suggests that buoyant density constancy has been conserved during evolution.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Bilinear Cell Growth of Escherichia coli
H. E. Kubitschek
J. Bacteriol. 148, 730-733 (1981)

Growth Delay Induced in Escherichia coli by Near-Ultraviolet Radiation: Relationship to Membrane Transport Functions
H. E. Kubitschek and R. J. Doyle
Photochem. Photobiol. [illegible], 595-702 (1981)

1982

RecA-Mediated Asymmetric Repair of Lethal DNA Lesions in E. coli
H. E. Kubitschek and C. N. Newman
Chromosome Damage and Repair, E. Seeberg and K. Kleppe, eds., Plenum Publishing Corporation, NY, 1981, pp. 187-191

Modified Protocol for Ames Mutagenicity Assay
H. E. Kubitschek and M. J. Peak
Environ. Mut. 3, 693-694 (1981)

Azaserine: Survival and Mutation in Escherichia coli
H. E. Kubitschek and R. J. Sepanski
Mutat. Res. 94, 31-38 (1982)

Biological Monitoring of Fluidized Bed Coal Combustion Operations: I. Increased Mutagenicity during Periods of Incomplete Combustion
H. E. Kubitschek, D. M. Williams, and F. R. Kirchner
Short-Term Bioassays in the Analysis of Complex Environmental Mixtures II, M. D. Walters, S. S. Sandhu, J. L. Huisingh, L. Claxton, and S. Nesnow, eds., Plenum Publishing Corporation, 1981, pp. 411-420

Survival of the Brine Shrimp, Artemia salina, after Exposure to 290-nm Ultraviolet Radiation, with and without Maximum Photoreactivation
M. J. Peak and H. E. Kubitschek
The Role of Solar Ultraviolet Radiation in Marine Ecosystems, J. Calkins, ed., Plenum Publishing Corporation, 1982, pp. 357-362

1983 through April

Role of AP Endonuclease in DNA Breakage and Cell Inactivation of Escherichia coli Subjected to Mild Heat (52°C)
D. M. Williams-Hill and N. Grecz
Mutat. Res. 107, 13-21 (1983)

In press as of May 1, 1983

Cell Elongation and Division Probability during the Escherichia coli Growth Cycle
H. E. Kubitschek and C. L. Woldringh
J. Bacteriol.

13. MOLECULAR AND CELLULAR EFFECTS OF SOLAR ULTRAVIOLET LIGHT

M. J. Peak, J. G. Peak,* R. W. Tuveson,† M. A. Turner,‡ P. M. Moehring,** and L. J. Sammartano††

This program addresses the basic nature and mechanisms of lethality, mutagenesis, and transformation caused by the nonionizing radiations of environmental solar ultraviolet (UV) light (> 290 nm). The research is of particular significance because of the possibility of enhanced fluxes of carcinogenic UVB radiation (290-330 nm) that would result from attenuation of stratospheric ozone by man's release of halocarbons into the atmosphere. The program studies DNA damages leading to lethal and mutagenic effects and the roles played by naturally occurring and experimentally added molecular probes in the mechanisms of the damage.

Biological systems used include isolated genetically active transforming DNA, strains of bacteria with genetically modified responses to radiation, mammalian cells in culture, and mouse skin. Physicochemical end points measured include DNA breakage (single strand and double strand scissions, alkali labile sites), DNA-protein cross-linking, and DNA photoproducts such as pyrimidine dimers and adducts of the pyrimidine-pyrimidine type. The following summary of some of our recent results includes a series of significant advances in solar UV photobiology.

Solar UV photosensitizers. Using mutant bacteria lacking the rare RNA nucleic acid base 4-thiouridine (s^4U), we had earlier identified s^4U as a naturally occurring photosensitizer involved in at least 20% of the lethal events caused by solar UV in *Escherichia coli*. Extension of these studies to our purified DNA *in vitro* shows that the addition of 0.1 M s^4U to the culture medium increases 7-fold the DNA breakage caused by 334-nm irradiation. This system is being used as a molecular model to probe the mechanisms of DNA breakage caused by natural photosensitizers and solar UV. Anoxia, diazobicyclo(2.2.2)octane, and histidine almost completely inhibit the s^4U enhancement of DNA breakage. This inhibition indicates a role for excited oxygen species, such as singlet oxygen ($^1O_2$), in the breakage. We have also observed similar inhibition by glycerol and benzoate, both known

---

*Mutagenesis Group.
†Scientist in Residence (Faculty Research Leave at Argonne Appointee), University of Illinois at Urbana.
‡Laboratory Graduate Program Participant, University of Missouri.
**Faculty Research Participant, Mount Mary College, Milwaukee.
††Spring 1982 participant in the Undergraduate Honors Program, St. Mary's College, Notre Dame, Indiana.

not to react with $^1O_2$: evidence for another excited species of oxygen, as yet unidentified. A minor role of superoxide is indicated by our finding that superoxide dismutase quenches a small proportion of the enhancement of breaks caused by s^4U and oxygen.

The role of $^1O_2$ in solar UV induced DNA breaks has been further elucidated by the use of a specific $^1O_2$ generating system - rose bengal and green light. Using this system, we have shown for the first time that $^1O_2$ has sufficient excitation energy to break purified DNA, consistent with the postulated role of this intermediate in solar UV damage.

We have recently discovered that a range of naturally occurring potential photosensitizers also accelerate the induction of breaks in purified DNA by solar UV. For instance, for the following photosensitizers, the dose enhancement factors at 10^{-3} M were: s^4U, 7.6; 5-methylaminomethyl-2-thiouridine, 7.6; 2-thiouracil, 43.4; nicotinamide adenine dinucleotide, reduced form - 6.8, oxidized form - 3.55; and riboflavine, 15.1.

Mutagenic interactions of solar UV wavelengths. The enhancement of 365-nm-induced mutagenesis in E. coli caused by a prior exposure of a nonmutagenic fluence of 405-nm radiation has been further studied. The enhancement is very strain specific; of the nine mutants with normal or defective repair capabilities that were studied, only two (both repair proficient) showed the phenomenon. It has now been demonstrated that this enhancement is not due to induction of the error-prone rec (SOS) system, since an action spectrum for the induction of this system (using both the prophage induction and β-galactoside-rec protein fusion methods) showed that the induction of SOS repair occurs only below 300 nm.

Solar UV sensitive bacteria. Detailed aerobic and anaerobic action spectra for a strain of E. coli specifically sensitive to solar UV radiations (nur, isolated by R. W. Tuveson) and its wild type resistant parent were completed. The mutant showed a large oxygen enhancement ratio compared with its wild type strain in the solar UV region, with a peak near 340 nm, close to its maximum for sensitivity to lethal effects. This is strong evidence that the physiological basis for nur may be a defect that results in intracellular hyperaccumulation of photosensitizing compounds, thus sensitizing the cells to both the lethality and the DNA breakage caused by these radiations.

DNA-protein cross-links. The observation that 365-nm radiation causes DNA-protein cross-links (proteinase K enhancement of alkaline elution from filters) in cultured Chinese hamster cells has now been extended to 405 nm (collaboration with A. Han). This radiation induced fewer cross-links than 365 nm, but significant numbers were observed well within the dose range for lethality. No pyrimidine dimers were observed in the same dose range of 405-nm radiation.

Solar UV mutagenesis in human cells. Analysis of an action spectrum for mutagenesis in cultured human cells (P3 epithelial cells) has continued (collaboration with C. A. Jones and E. Huberman). Mutagenesis by shorter wavelengths appeared to follow the absorption of DNA. The spectrum showed a minor peak at 365 nm, with a minimum at 334 nm.

In conclusion, advances have been made that increase our understanding of fundamental molecular events caused by solar UV, an environmentally important radiation. Especially significant is the demonstration that several non-DNA, naturally occurring chromophores may interact with the radiation and the DNA in photodynamic-type reactions that result in DNA damage. The possible involvement of non-DNA molecules in the actions of these carcinogenic radiations may extend our understanding of the basic mechanisms of carcinogenesis caused by radiation.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Protection by DABCO against Inactivation of Transforming DNA by Near-Ultraviolet Light: Action Spectra and Implications for Involvement of Singlet Oxygen
J. G. Peak, C. S. Foote, and M. J. Peak
Photochem. Photobiol. 34, 45-49 (1981)

The Effect of Stray Light in the 300-320-nm Range and the Role of Cyclobutyl Pyrimidine Dimers in 365-nm Lethality
R. L. Webb and M. J. Peak
Mutat. Res. 91, 177-182 (1981)

1982

Modified Protocol for Ames Mutagenicity Assay
H. E. Kubitschek and M. J. Peak
Environ. Mut. 3, 693-694 (1981)

Lethality in Repair-Proficient *Escherichia coli* after 365 nm Ultraviolet Light Irradiation is Dependent on Fluence Rate
J. G. Peak and M. J. Peak
Photochem. Photobiol. 36, 103-105 (1982)

Effects of Glycerol upon the Biological Actions of Near-Ultraviolet Light: Spectra and Concentration Dependence for Transforming DNA and for *Escherichia coli* B/r
J. G. Peak, M. J. Peak, and C. S. Foote
Photochem. Photobiol. 36, 413-416 (1982)

Liver-Microsome S9 Enzyme Increases Spontaneous Background Mutation Frequency in the Ames Salmonella Test System in the Absence of Any Added Mutagen
M. J. Peak, S. S. Dornfeld, and D. Venters
Mutat. Res. 103, 263-265 (1982)

Survival of the Brine Shrimp, Artemia salina, after Exposure to 290-nm Ultraviolet Radiation, with and without Maximum Photoreactivation
M. J. Peak and H. E. Kubitschek
The Role of Solar Ultraviolet Radiation in Marine Ecosystems, J. Calkins, ed., Plenum Publishing Corporation, 1982, pp. 357-362

Lethal Effects on Biological Systems Caused by Solar Ultraviolet Light: Molecular Considerations
M. J. Peak and J. G. Peak
The Role of Solar Ultraviolet Radiation in Marine Ecosystems, J. Calkins, ed., Plenum Publishing Corporation, 1982, pp. 325-336

Single-Strand Breaks Induced in Bacillus subtilis DNA by Ultraviolet Light: Action Spectrum and Properties
M. J. Peak and J. G. Peak
Photochem. Photobiol. 35, 675-680 (1982)

1983 through April

The Role of 4-Thiouridine in Lethal Effects and in DNA Backbone Breakage Caused by 334 nm Ultraviolet Light in Escherichia coli
M. J. Peak, J. G. Peak, and L. Nerad
Photochem. Photobiol. 37, 169-172 (1983)

Single-Strand DNA Breaks Induced by 365 nm Radiation in Escherichia coli Strains Differing in Sensitivity to Near and Far UV
R. W. Tuveson, J. G. Peak, and M. J. Peak
Photochem. Photobiol. 37, 109-112 (1983)

In press as of May 1, 1983

Induction of Pyrimidine Dimers in Epidermal DNA of Hairless Mice by UVB: An Action Spectrum
R. D. Ley, M. J. Peak, and L. L. Lyon
J. Invest. Dermatol. 80, 188-191 (1983)

Role of DNA Single Strand Breaks in Lethal and Mutagenic Events Caused by UVB Radiation
M. J. Peak
Workshop on Biological Effects of UVB, U. S. Environmental Protection Agency, Munich, Germany, May 22-27, 1982

Use of Action Spectra for Identifying Molecular Targets and Mechanisms of Action of Solar Ultraviolet Light
M. J. Peak and J. G. Peak
Physiol. Plant.

The following publications by Dr. Robert B. Webb resulted from a previous component of this and the preceding program (Chapter 12).

1981

The Effect of Stray Light in the 300-320-nm Range and the Role of Cyclobutyl Pyrimidine Dimers in 365-nm Lethality
R. B. Webb and M. J. Peak
Mutat. Res. 91, 177-182 (1981)

1982

Photodynamic Effects of Dyes on Bacteria: V. Mutagenesis by Acridine Orange and 460-nm or 500-nm Light in Strains of *Escherichia coli* that Differ in Repair Capability
B. S. Hass and R. B. Webb
Mutat. Res. 94, 39-51 (1982)

Genetic Damage in *Escherichia coli* K12 AB2480 by Broad-Spectrum Near-Ultraviolet Radiation
R. B. Webb and M. S. Brown
Science 215, 991-993 (1982)

Nonreciprocal Synergistic Lethal Interaction between 365-nm and 405-nm Radiation in Wild Type and *uvrA* Strains of *Escherichia coli*
R. B. Webb, M. S. Brown, and R. D. Ley
Photochem. Photobiol. 35, 697-703 (1982)

Mutation Induction by Monochromatic 254-nm and 365-nm Radiation in Strains of *Escherichia coli* That Differ in Repair Capability
R. B. Webb and M. A. Turner
Mutat. Res. 84, 227-237 (1981)

Differential Sensitivity to Inactivation of *nur* and nur^+ Strains of *Escherichia coli* at Six Selected Wavelengths in the UVA, UVB and UVC Ranges
R. B. Webb and R. W. Tuveson
Photochem. Photobiol. 36, 525-530 (1982)

TOXICOLOGY SECTION

Chemical Toxicology Group, C. A. Reilly Principal Investigator

Chapter 14. Chemical Toxicology

Chemical and Biological Characterization Group, D. A. Haugen Principal Investigator

Chapter 15. Carcinogen Identification and Metabolism

Metal Toxicology Group, M. H. Bhattacharyya Principal Investigator

Chapter 16. Metal Metabolism and Toxicity

Neurobehavioral Chronobiology Group, C. F. Ehret Principal Investigator

Chapter 17. Neurobehavioral Chronobiology and Toxicology

14. CHEMICAL TOXICOLOGY

C. A. Reilly, Jr., F. R. Kirchner, M. J. Peak, K. E. Wilzbach,*
R. E. Jones, J. M. Leonardo, S. S. Dornfeld, V. A. Pahnke, M. A. Shotola,
D. Venters, R. M. Bressette,† M. A. Fink,‡ S. M. Kerwin,**
M. F. Muraski,†† C. M. Tompkins,‡‡ and R. J. Wechsler***

The variety of toxicants in the by-products of coal conversion processes and their potential interactions make assessment of the impact of these toxicants on human health and the environment a most difficult task. The abundance of fossil fuel resources in this country necessitates that the potential impacts be quantitated prior to large scale utilization so that unwarranted risks can be identified and avoided. The Chemical Toxicology Group forms one part of a multidisciplinary program that addresses the uncertainties of health risks and provides the data that will allow definitive assessments. Currently, we are investigating the toxicology of complex mixtures with particular emphasis on the effects that chemical interactions have on the resultant toxicity. Historically, the project focused on coal gasification and provided toxicologic screening information about a variety of pilot plant effluents with emphasis on the effects of chronic dermal exposure to high molecular weight organic by-products.

Materials have been studied from three coal gasifiers: the Institute for Gas Technology's HYGAS pilot plant gasifier (fluidized bed); the Grand Forks Energy Technology Center's pilot plant gasifier (slagging fixed-bed); and Mellon Institute's process development unit (coal pyrolysis). Samples from a wood gasifier at Battelle Columbus Laboratory have also been studied. We have found that by-products (principally tars) from the different types of gasifiers are quite similar, both toxicologically and chemically. This finding strongly suggests that thermodynamic and kinetic

*Energy and Environmental Systems Division.
†Summer 1982 participant in the Undergraduate Honors Program, College of St. Benedict.
‡Spring and Summer 1982 participant in the Undergraduate Honors Program, St. Xavier College.
**Summer 1982 participant in the Undergraduate Honors Program, University of Notre Dame.
††Spring and Summer 1982 participant in the Undergraduate Honors Program, Purdue University.
‡‡Fall 1982 participant in the Undergraduate Honors Program, Morris Brown College.
***Fall 1982 participant in the Undergraduate Honors Program, Colorado State University.

factors are controlling the composition of the by-product mixtures, and supports a generic approach to current investigations of the toxic hazards of coal gasification. The general data base is now sufficiently large that evaluation of such things as low dose effects (more relevant to human exposures), dose-response relationships, and the metabolism of major toxicants is justified.

Mammalian toxicology studies focus on carcinogenicity. Microbial and mammalian cellular assays are used in support of these studies and have been especially valuable in the chemical characterization of the organic by-products (Chapter 15).

The standard plate incorporation Salmonella mutagenesis assay has been useful, but the assay is difficult to quantitate and has a limited linear range for some materials, e.g., polycyclic aromatic hydrocarbons (PAH). Currently we are validating a procedure we have devised whereby changes in the concentrations of test chemicals and reagents due to diffusion in the culture plates are eliminated as a variable. We are also exploring biochemical approaches to extending the useful range of mutagenic responses to PAH by reducing the effects of some of their metabolites that inhibit metabolic activation.

Studies on the dermal carcinogenicity of the tar from the Grand Forks Energy Technology Center (GFETC) to hairless mice show it to be strongly carcinogenic, inducing tumors in 100% of the animals at a dose of 75 mg/week. A 50% tumor incidence was reched by 17 weeks, in comparison to 18 weeks required to reach 50% tumor incidence after a much smaller dose (0.03 mg/week) of the known dermal carcinogen benzo[a]pyrene. Histopathological evaluation of the skin tumors induced by the GFETC tar showed that 46% (12/26) were squamous cell carcinomas. No evidence for systemic carcinogenicity of the dermally applied tar was observed histologically, but serum and urine analysis gave indications of liver and kidney damage.

Examinations of acid, base, and neutral fractions of the GFETC tar show that carcinogens are present in all fractions, but that the neutral fraction contains the most potent carcinogens. Comparison of the carcinogenicity of the raw tar with that of the neutral fraction (at its concentration in the raw tar) showed the complete tar to be less potent, indicating that interaction of the components in complex mixtures may inhibit some of the carcinogenic activity. Studies investigating these interactions are in progress.

The basic fraction is highly mutagenic, but induces fewer dermal tumors (50% of the animals with tumors at 42 weeks) than the neutral fraction. This lower carcinogenic response suggests that if the mutagenic components contribute significantly to carcinogenicity, they may do so at a site other than skin. The known bladder carcinogenicity of certain basic compounds, such as aromatic amines, and the fact that mice (the test animal used) do not develop bladder cancer from these agents leave this issue to be resolved by further experimentation.

The acidic fraction, although not mutagenic, does in fact induce skin tumors in some animals (28% at 42 weeks). High molecular weight hydroxy-PAH not detectable in the Ames assay are the suspected carcinogens in this

subfraction. Chemical characterization of this fraction indicates that a number of tumor promoting agents (e.g., phenols and catechols) are present, and studies need to be performed that address the contribution of such agents to carcinogenicity.

To determine the ability of a gasification tar to act solely as an initiator of carcinogenesis, a single dermal exposure of either 25 or 50 mg of GFETC tar was followed by twice weekly dermal treatment with a tumor-promoting agent, tetradecanoylphorbol acetate (TPA). After one year of TPA treatment, skin tumor incidence was 72% for the 50 mg group and 68% for the 25 mg group, compared with an incidence of 20% in the mice that received only the single 50 mg tar exposure (no promotion). It is thus clear that the tar can act as a tumor initiator as well as a complete carcinogen.

The program is currently obtaining information on health impacts of chronic low dose exposure to complex organic mixtures. A great deal of information is available for pure compounds but information is quite limited for complex mixtures. A better understanding of interactions of toxic compounds in complex mixtures will allow more accurate assessment of health risks.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Near-UV Induction of Sister Chromatid Exchanges under Aerobic and Anaerobic Conditions
V. M. Griego, G. C. Matsushita, and T. Matsushita
Photochem. Photobiol. 34, 51-55 (1981)

Dependence on Stage of Growth of Mammalian Cell Sensitivity to Near-UV Irradiation
V. M. Griego, R. B. Webb, and T. Matsushita
Photochem. Photobiol. 33, 211-214 (1981)

Synergistic, Additive, and Antagonistic Mutagenic Responses to Binary Mixtures of Benzo(a)pyrene and Benzo(e)pyrene as Detected by Strains TA98 and TA100 in the Salmonella/Microsome Assay
B. S. Hass, E. E. Brooks, K. E. Schumann, and S. S. Dornfeld
Environ. Mut. 3, 159-166 (1981)

Use of Nitrous Acid-Dependent Decrease in Mutagenicity as an Indication of the Presence of Mutagenic Primary Aromatic Amines: Non-Specific Reactions with Phenols and Benzo(a)pyrene
D. A. Haugen, M. J. Peak, and C. A. Reilly, Jr.
Mutat. Res. 82, 59-67 (1981)

Redispersion of Fly Ash from a Liquid Nitrogen Suspension
F. R. Kirchner, J. O. Hutchens, P. C. Brennan, and W. P. Norris
J. Aerosol Sci. 12, 39-47 (1981)

Health Effects of Synfuels Technology - A Review
L. P. Sanathanan, C. A. Reilly, S. A. Marshall, and K. E. Wilzbach
ANL/ES-111 (April 1981)

Chemical and Toxicological Characterization of HYGAS Pilot Plant Process Streams
K. E. Wilzbach and C. A. Reilly, Jr.
Proceedings of the Second International Gas Research Conference, Los Angeles, CA, September, 1981, Government Industries, Inc., Rockville, MD, 1981, 1550-1561

1982

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process): III. Mass and Mutagenicity Balances in Chemical Fractionations
S. Bourne, A. M. Jirka, and M. J. Peak
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 173-180

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). II. Nitrous Acid Treatment for Detection of Mutagenic Primary Aromatic Amines: Nonspecific Reactions
D. A. Haugen, M. J. Peak, and C. A. Reilly, Jr.
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 115-127

Biological Monitoring of Fluidized Bed Coal Combustion Operations: II. Mammalian Responses following Exposure to Gaseous Effluents
F. R. Kirchner, D. M. Buchholz, V. A. Pahnke, and C. A. Reilly, Jr.
Short-Term Bioassays in the Analysis of Complex Environmental Mixtures II, M. D. Walters, S. S. Sandhu, J. L. Huisingh, L. Claxton, and S. Nesnow, eds., Plenum Publishing Corporation, 1981, pp. 421-430

Modified Protocol for Ames Mutagenicity Assay
H. E. Kubitschek and M. J. Peak
Environ. Mut. 3, 693-694 (1981)

Biological Monitoring of Fluidized Bed Coal Combustion Operations: I. Increased Mutagenicity during Periods of Incomplete Combustion
H. E. Kubitschek, D. M. Williams, and F. R. Kirchner
Short-Term Bioassays in the Analysis of Complex Environmental Mixtures II, M. D. Walters, S. S. Sandhu, J. L. Huisingh, L. Claxton, and S. Nesnow, eds., Plenum Publishing Corporation, 1981, pp. 411-420

Electrical Determination of Viability in Saline-Treated Mouse Myeloma Cells
T. Matsushita, A. M. Brendzel, M. A. Shotola, and K. R. Groh
Biophys. J. 39, 41-47 (1982)

Liver-Microsome S9 Enzyme Increases Spontaneous Background Mutation Frequency in the Ames Salmonella Test System in the Absence of Any Added Mutagen
M. J. Peak, S. S. Dornfeld, and D. Venters
Mutat. Res. 103, 263-265 (1982)

Health and Environmental Studies of Coal Gasification Process Streams and Effluents
C. A. Reilly, Jr., A. S. Boparai, S. Bourne, R. D. Flotard, D. A. Haugen, R. E. Jones, F. R. Kirchner, T. Matsushita, M. J. Peak, V. C. Stamoudis, J. R. Stetter, and K. E. Wilzbach
Symposium Proceedings: Environmental Aspects of Fuel Conversion Technology - VI, A Symposium on Coal-Based Synfuels, Denver, CO., October 26-30, 1981, F. A. Ayer and N. S. Jones, compilers, U.S. Environmental Protection Agency, Washington, DC, EPA-600/9-82-017, 1982, pp. 282-292

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). IV. Biological Activity
C. A. Reilly, Jr., M. J. Peak, T. Matsushita, F. R. Kirchner, and D. A. Haugen
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 310-324

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). I. Chemical Characterization of Mutagenic Fractions
V. C. Stamoudis, S. Bourne, D. A. Haugen, M. J. Peak, C. A. Reilly, J. R. Stetter, and K. Wilzbach
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 67-95

Status of Health and Environmental Research Relative to Coal Gasification: 1976 to the Present
K. E. Wilzbach and C. A. Reilly, Jr., compilers and eds.
U.S. Department of Energy, Washington, DOE/ER-0149, 1982

Environmental Research Program for Slagging Fixed-Bed Coal Gasification
K. E. Wilzbach, J. R. Stetter, C. A. Reilly, Jr., and W. G. Willson
ANL/SER-1 (November 1981)

1983 through April

Mixtures of Polycyclic Aromatic Compounds Inhibit Mutagenesis in the Salmonella/Microsome Assay by Inhibition of Metabolic Activation
D. A. Haugen and M. J. Peak
Mutat. Res. 116, 257-269 (1983)

Isolation and Identification of Mutagenic Primary Aromatic Amines from Synthetic Fuel Materials
D. A. Haugen, V. C. Stamoudis, M. J. Peak, and A. S. Boparai
Proceedings of the 6th International Symposium on Polynuclear Aromatic Hydrocarbons, October, 1981, M. Cook, A. J. Dennis, and G. L. Fisher, eds., Battelle Press, Columbus, OH and Springer-Verlag, NY, 1982, pp. 347-356

In press as of May 1, 1983

An Improved Procedure for Extraction of Aromatic Bases from Synfuel Materials
A. S. Boparai, D. A. Haugen, K. M. Suhrbier, and J. F. Schneider
Proceedings of the DOE Workshop on Chemical Characterization of Hazardous Substances in Synfuels, Seattle, WA, November 4-6, 1981

Toxicologic and Physiochemical Characterization of High-Temperature Combustion Emissions
F. R. Kirchner, P. F. Dunn, and C. B. Reed
Aerosol Sci. Technol.

Toxicological Effects on Mice following Inhalation of Fluidized Bed Coal Combustor Fly Ash
F. R. Kirchner, C. A. Reilly, Jr., D. M. Buchholz, and
V. A. Pahnke, Jr.
Environ. Res.

Bio-Directed Chemical Characterization of Condensates from Synfuel Processes
V. C. Stamoudis, D. A. Haugen, M. J. Peak, and K. E. Wilzbach
Proceedings of the DOE Workshop on Chemical Characterization of Hazardous Substances in Synfuels, Seattle, WA, November 4-6, 1981

Status of Coal Gasification Health and Environmental Research
K. E. Wilzbach and C. A. Reilly, Jr.
Proceedings of the Fifth Life Sciences Symposium, Gatlinburg, TN, October 24-27, 1982

15. CARCINOGEN IDENTIFICATION AND METABOLISM

D. A. Haugen, M. J. Peak,* V. C. Stamoudis,† A. S. Boparai,‡
C. A. Reilly, Jr.,* K. E. Wilzbach,† K. M. Suhrbier, S. S. Dornfeld,*
D. Venters,* and F. J. Tremmel**

The immediate objectives of this project are to identify chemicals primarily responsible for the toxicologic activity of coal conversion materials, to investigate their mode of action and biological fate, and to understand better the expression of their activity when they are present in complex mixtures. The long-range objective is to provide information useful for establishing chemical analyses and short-term bioassays predictive of toxicologic effects to assure that large-scale utilization of fossil fuels is achieved in a manner that minimizes risks to human health.

Previously, we examined effluent and process stream samples from the coal gasification pilot plant operated by the Institute of Gas Technology for development of the HYGAS process. We are now examining materials from the slagging fixed-bed gasifier at the Grand Forks Energy Technology Center, and from the experimental coal pyrolysis unit operated at the Mellon Institute. For comparative purposes we also examine reference materials from other coal conversion technologies.

Materials are fractionated and analyzed by a variety of complementary procedures to successively isolate and identify mutagenic and carcinogenic chemicals by class, subclass, and specific compound. The mutagenicity of the fractions is measured using the Ames *Salmonella*/microsome assay with rat liver preparations for metabolic activation.

For the several coal-derived materials we have examined, the acidic components (predominantly phenolic compounds) constituted about 25% of the materials, but had negligible mutagenicity. The basic and neutral components constituted about 5% and 70% of the material, respectively. Despite their difference in relative abundance, the basic and neutral fractions were each responsible for 40 to 60% of the mutagenicity. In the following, we describe first our studies of basic components and then the information we have obtained concerning the neutral components.

*Chemical Toxicology Group.
†Energy and Environmental Systems Division.
‡Analytical Chemistry Laboratory, Chemical Engineering Division.
**Laboratory Graduate Program Participant, University of Iowa.

We have previously developed procedures for efficiently extracting aromatic bases from coal conversion by-products, and for their separation by high performance liquid chromatography. Mutagenesis assays of chromatographic fractions revealed that 3-aminophenanthrene was a predominant mutagen. It is one of several 3-ring aromatic amines known to be potent carcinogens for rodents. Alkylated 3-ring aromatic amines also contributed to the mutagenicity of the coal conversion by-products. Chemical analysis of basic fractions demonstrated the presence of other carcinogenic aromatic amines (e.g., aminonaphthalene) and azaarenes which were not detected as predominant mutagens. Thus toxicologic and chemical characterization provided complementary and supportive evidence for the presence of carcinogenic bases.

To follow up the identification of 3-ring aromatic amines as toxicologically important chemicals, we are examining the mutagenicity, carcinogenicity, and metabolism of 1- and 2-aminoanthracene and their methyl derivatives as model compounds. Little is known about the metabolism of these compounds; although methylation of 2-ring carcinogenic aromatic amines at certain sites increases their potency, comparable information is not available for alkylated aminophenanthrenes or aminoanthracenes.

In our studies of model compounds, we compared the activities of 1- and 2-aminoanthracene for bacterial mutagenesis and for hepatic tumorigenesis as measured in the short-term initiation/promotion system developed by C. Peraino (see Chapter 3 of this report). 2-Aminoanthracene is a potent bacterial mutagen which is 5 to 100 times more mutagenic than 1-aminoanthracene (depending on the pretreatment of the animals from which the liver preparations for metabolic activation were obtained). Treatment of neonatal rats with either 1- or 2-aminoanthracene followed by promotion with dietary phenobarbital revealed that neither compound was significantly tumorigenic. Neonatal liver preparations had relatively little ability to activate 2-aminoanthracene in the mutagenesis assay. This may be responsible for the lack of tumorigenic activity in the neonatal initiation/promotion system.

We have also examined neutral components of a coal gasification tar. Of the neutral components, 4- to 6-ring polycyclic aromatic hydrocarbons (PAH) were responsible for most of the mutagenicity. Analysis was complicated by the presence of multi-alkylated PAH. The predominant mutagens were the unsubstituted PAH cyclopenta[cd]pyrene, benz[a]anthracene, chrysene, benzo[a]pyrene, benzofluoranthenes, anthanthrene, and dibenzopyrenes, as well as methyl and methylene derivatives of the PAH. Although the unsubstituted 4-6 ring PAH and their methyl or methylene derivatives were primarily responsible for the mutagenicity, the presence of other PAH and their alkyl derivatives raises concerns for two reasons. First, some alkyl derivatives of 3-6 ring PAH are more potent carcinogens than the corresponding parent compounds. Second, although most unsubstituted and alkylated 2- and 3-ring PAH are noncarcinogenic, some have been shown to be cocarcinogens. Further work will address the possible role for the relatively abundant noncarcinogens in the expression of the carcinogenicity and mutagenicity of the mixtures.

In addition to measuring the mutagenicity of relatively small chromatographic fractions, we prepared and characterized sufficient quantities of acidic, basic, and neutral fractions of gasification tar for use in dermal carcinogenesis assays (see Chapter 14). Because the neutral fractions were the most abundant and the most carcinogenic, we will prepare subfractions of the neutral components for further carcinogenicity measurements.

In summary, we have isolated and identified the principal basic and neutral mutagens and carcinogens in by-products of coal conversion by the use of complementary chemical and toxicologic approaches. We are examining the mutagenicity, carcinogenicity, and metabolism of aminoanthracenes as model compounds because the 3-ring aromatic amines are major contributors to the mutagenicity of the by-products, because they are potent mutagens and carcinogens, and because their metabolism has received little attention. Analysis of neutral fractions of the gasification tar revealed the presence of carcinogenic and mutagenic PAH as well as other PAH that are suspected of being cocarcinogens. Further work will provide a better understanding of the effects of the toxicologically important chemicals and will lead to establishment of procedures for predicting risk to human health.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Biphenyl Metabolism by Rat Liver Microsomes: Regioselective Effects of Inducers, Inhibitors, and Solvents
D. A. Haugen
Drug Metab. Disp. 9, 212-218 (1981)

Use of Nitrous Acid-Dependent Decrease in Mutagenicity as an Indication of the Presence of Mutagenic Primary Aromatic Amines: Non-Specific Reactions with Phenols and Benzo(a)pyrene
D. A. Haugen, M. J. Peak, C. A. Reilly, Jr.
Mutat. Res. 82, 59-67 (1981)

Chemical and Toxicological Characterization of HYGAS Pilot Plant Process Streams
K. E. Wilzbach and C. A. Reilly, Jr.
Proceedings of the Second International Gas Research Conference, Los Angeles, CA, September, 1981, Government Industries, Inc., Rockville, MD, 1981, 1550-1561

1982

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process): III. Mass and Mutagenicity Balances in Chemical Fractionations
S. Bourne, A. M. Jirka, and M. J. Peak
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 173-180

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). II. Nitrous Acid Treatment for Detection of Mutagenic Primary Aromatic Amines: Nonspecific Reactions
D. A. Haugen, M. J. Peak, and C. A. Reilly, Jr.
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 115-127

Isolation of Mutagenic Aromatic Amines from a Coal Conversion Oil by Cation Exchange Chromatography
D. A. Haugen, M. J. Peak, and K. M. Suhrbier
Anal. Chem. 54, 32-37 (1982)

Health and Environmental Studies of Coal Gasification Process Streams and Effluents
C. A. Reilly, Jr., A. S. Boparai, S. Bourne, R. D. Flotard, D. A. Haugen, R. E. Jones, F. R. Kirchner, T. Matsushita, M. J. Peak, V. C. Stamoudis, J. R. Stetter, and K. E. Wilzbach
Symposium Proceedings: Environmental Aspects of Fuel Conversion Technology - VI, A Symposium on Coal-Based Synfuels, Denver, CO., October 26-30, 1981, F. A. Ayer and N. S. Jones, compilers, U.S. Environmental Protection Agency, Washington, DC, EPA-600/9-82-017, 1982, 282-292

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). IV. Biological Activity
C. A. Reilly, Jr., M. J. Peak, T. Matsushita, F. R. Kirchner, and D. A. Haugen
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 310-324

Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). I. Chemical Characterization of Mutagenic Fractions
V. C. Stamoudis, S. Bourne, D. A. Haugen, M. J. Peak, C. A. Reilly, J. R. Stetter, and K. Wilzbach
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 67-95

Status of Health and Environmental Research Relative to Coal Gasification: 1976 to the Present
K. E. Wilzbach and C. A. Reilly, Jr., compilers and eds.
U.S. Department of Energy, Washington, DOE/ER-0149, 1982

Environmental Research Program for Slagging Fixed-Bed Coal Gasification
K. E. Wilzbach, J. R. Stetter, C. A. Reilly, Jr., and W. G. Willson
ANL/SER-1 (November 1981)

1983 through April

Mixtures of Polycyclic Aromatic Compounds Inhibit Mutagenesis in the Salmonella/Microsome Assay by Inhibition of Metabolic Activation
D. A. Haugen and M. J. Peak
Mutat. Res. 116, 257-269 (1983)

Isolation and Identification of Mutagenic Primary Aromatic Amines from Synthetic Fuel Materials
D. A. Haugen, V. C. Stamoudis, M. J. Peak, and A. S. Boparai
Proceedings of the 6th International Symposium on Polynuclear Aromatic Hydrocarbons, October, 1981, M. Cook, A. J. Dennis, and G. L. Fisher, eds., Battelle Press, Columbus, OH and Springer-Verlag, NY, 1982, pp. 347-356

1983 through April

In press as of May 1, 1983

An Improved Procedure for Extraction of Aromatic Bases from Synfuel Materials
A. S. Boparai, D. A. Haugen, K. M. Suhrbier, and J. F. Schneider
Proceedings of the DOE Workshop on Chemical Characterization of Hazardous Substances in Synfuels, Seattle, WA, November 4-6, 1981

Isolation of Mutagenic Polycyclic Aromatic Hydrocarbons from Tar Produced during Coal Gasification
D. A. Haugen, V. C. Stamoudis, M. J. Peak, and A. S. Boparai
7th International Symposium of Polynuclear Aromatic Hydrocarbons, Columbus, OH, October 26-28, 1982

Bio-Directed Chemical Characterization of Condensates from Synfuel Processes

V. C. Stamoudis, D. A. Haugen, M. J. Peak, and K. E. Wilzbach
Proceedings of the DOE Workshop on Chemical Characterization of Hazardous Substances in Synfuels, Seattle, WA, November 4-6, 1981

16. METAL METABOLISM AND TOXICITY

M. H. Bhattacharyya, R. P. Larsen,* B. D. Whelton,† P. A. Benioff,‡
E. S. Moretti, D. P. Peterson, R. D. Oldham,* and M. E. Shackelford**

This research program is designed to investigate health effects of toxic metals in animals under conditions relevant to potential human exposures. One facet deals with effects of pregnancy and lactation on susceptibility to toxic effects of compounds of cadmium and lead. A second investigates the gastrointestinal (GI) absorption of plutonium and other actinide elements under conditions relevant to setting of drinking water standards for man.

In both facets, compounds are administered orally and GI absorption factors are determined as a first step in evaluating metabolic and/or toxic responses. Pathways for uptake and tissue deposition are studied using radioactive isotopes (measured by gamma spectrophotometry or alpha spectrometric isotope dilution) or using stable isotopes (measured by atomic absorption spectrophotometry). Toxic responses are evaluated by competitive metabolite binding protein assays; analysis of urinary concentrations of cadmium, amino acids, and proteins; analysis of calcium content of bone; and measurements of blood concentrations of protoporphyrin, calcium, lead, and hemoglobin.

Two program areas, not covered in this report, are scheduled for completion. These areas are (1) preparation of Health and Environmental Effects Documents (HEED's) and (2) study of toxic responses to arsine and stibine in mice. In the latter study, a striking difference in time-course of response to arsine and stibine was demonstrated, along with a 2- to 3-fold difference in toxicity.

Metabolism and Toxicity of Compounds of Cadmium and Lead

Influence of lead exposure during pregnancy and lactation on vitamin D metabolite concentrations. We are determining whether lead exposure during pregnancy and lactation in rats reduces concentrations of circulating

*Radiological and Environmental Research Division.
†Scientist in Residence (Faculty Research Leave at Argonne Appointee), Eastern Washington University.
‡Environmental Impact Studies Division.
**Laboratory Graduate Program Participant, University of Illinois at Chicago Circle.

$1,25(OH)_2$ vitamin D. The concentration of this metabolite, which is essential to maintaining calcium homeostasis during periods of calcium stress, normally increases during pregnancy and lactation, and also during rapid growth years in children. Serum concentrations of $1,25(OH)_2$ vitamin D were recently shown to be strikingly reduced in children with elevated levels of lead in blood. Results to date indicate that rats exposed to lead in the diet during gestation and lactation had blood lead concentrations on lactation day 14 that were almost 10 times higher than those of nonpregnant control rats, and that blood lead concentrations were similar in pups and dams. Decreases in hematocrit, free erythrocyte protoporphyrin, and serum calcium were more striking in both the lactating dams and their nursing pups than in nonpregnant animals. Measurements of concentrations of vitamin D metabolites [25(OH) vitamin D, $1,25(OH)_2$ vitamin D, and $24,25(OH)_2$ vitamin D] in rat blood plasma are under way.

Influence of pregnancy and lactation on cadmium toxicity. We previously demonstrated a 2- to 3-fold increase in the GI absorption and retention of cadmium in the maternal mouse during pregnancy and lactation. The question of whether exposure to cadmium during pregnancy and lactation enhances the development of toxic responses is now being addressed. Increased absorption of cadmium during successive pregnancies may have contributed to the increased incidence of Itai-Itai disease among Japanese women environmentally exposed to cadmium in rice. The highest incidence occurred among postmenopausal women with a history of multiple childbirths (average number of children = 6). Groups of adult female mice (~ 350 mice/group) have been placed on diets containing 0.25, 5, or 50 ppm cadmium. Mice in each group are being bred through six successive rounds of pregnancy and lactation, with the mothers from one round being rebred for the next round immediately after weaning. At the end of each round of pregnancy and lactation, tissues and urine samples are being taken for histological and/or biochemical evaluation.

We also recently demonstrated that almost all (> 90%) of the cadmium transferred from dams to pups during lactation comes from the diet consumed by the dam during lactation. Very little transfer to pups takes place from long-term maternal body stores, even with prolonged cadmium exposure prior to mating (140 days exposure to 5 ppm Cd in the diet).

Gastrointestinal Absorption of Plutonium and Other Actinides

Determination of an f_1 factor for plutonium in man. The following three areas were identified as needing clarification in recommending an absorption factor (f_1) for man applicable to environmental exposures to plutonium in drinking water: (1) effect of fasting on the GI absorption of plutonium; (2) effect of animal age on the GI absorption of plutonium; and (3) extrapolation of laboratory animal data to man. GI absorption values in rats range from 3×10^{-3}, in both the fasted adult and fed neonatal rat, down to 1×10^{-4} in the fed adult rat. Taking into account effects of both fasting and animal age, an f_1 factor of 6×10^{-4} appears appropriate, based on rodent data only. An experiment in baboons, jointly supported by the Nuclear Regulatory Commission and the Department of Energy, is under way to aid in extrapolation of this rodent-based factor to man. In this experiment, multiple isotopes of plutonium (^{239}Pu, ^{236}Pu, ^{238}Pu), neptunium (^{237}Np, ^{239}Np), and uranium (^{233}U, ^{236}U) are being administered

to each baboon via either oral administration or intravenous injection. Results will provide GI absorption factors for all three actinides in each baboon, as well as validation (or not) of a method for determining GI absorption values for plutonium and uranium using noninvasive procedures (analysis of blood and/or urine samples only).

In conclusion: (1) Elevated concentrations of lead in blood of lactating animals and their pups when the dams receive dietary lead during gestation and lactation suggest that dams and their offspring both are at increased risk, compared with nonpregnant animals, of developing toxic responses to lead during this critical period. (2) Cadmium exposure during pregnancy and lactation may cause responses at lower concentrations in the dam than in nonpregnant animals due to increased accumulation of cadmium in the dam's tissues. (3) Results on cadmium transfer to pups during lactation, along with corresponding results for lead (Keller and Doherty, 1980), imply that women occupationally exposed to cadmium or lead should not be exposed during lactation, but that a history of previous exposures does not necessarily result in ready transfer of these toxic metals to the young, provided that exposure during lactation itself is avoided. Finally, (4) the value adopted by the International Commission for Radiological Protection for the fraction of soluble plutonium transferred from the GI tract to blood in man, $f_1 = 1 \times 10^{-4}$, appears to be approximately an order of magnitude too low.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1982

Gastrointestinal Absorption of Cadmium in Mice during Gestation and Lactation. I. Short-Term Exposure Studies
M. H. Bhattacharyya, B. D. Whelton, and D. P. Peterson
Toxicol. Appl. Pharmacol. 61, 335-342 (1981)

Plutonium Retention in Mice and Rats after Gastrointestinal Absorption
R. P. Larsen, R. D. Oldham, M. H. Bhattacharyya, E. S. Moretti, and D. J. Austin
Radiat. Res. 87, 37-49 (1981)

On the Gastrointestinal Absorption of Plutonium
R. P. Larsen, R. D. Oldham, M. H. Bhattacharyya, E. S. Moretti, and D. J. Austin
Actinides in Man and Animals, M. E. Wrenn, ed., RD Press, Salt Lake City, UT, 1981, pp. 303-307

1983 through April

Gastrointestinal Absorption of Cadmium in Mice during Gestation and Lactation: II. Continuous Exposure Studies
M. H. Bhattacharyya, B. D. Whelton, and D. P. Peterson
Toxicol. Appl. Pharmacol. 66, 368-375 (1982)

In press as of May 1, 1983

Bioavailability of Orally Administered Cadmium and Lead to the Mother, Fetus, and Neonate during Pregnancy and Lactation: An Overview
M. H. Bhattacharyya
Biological Availability of Trace Metals, R. E. Wilding and E. A. Jenne, eds., Elsevier Press, Amsterdam, 1983, pp. 327-342.

Plutonium - Its Behavior in Natural Water Systems and Assimilation by Man
R. P. Larsen, D. M. Nelson, M. H. Bhattacharyya, and R. D. Oldham
Health Phys.

Retention of Plutonium in the Beagle after Gastrointestinal Absorption
R. E. Toohey, M. H. Bhattacharyya, R. D. Oldham, R. P. Larsen, and E. S. Moretti
Radiat. Res.

17. NEUROBEHAVIORAL CHRONOBIOLOGY AND TOXICOLOGY

C. F. Ehret and K. R. Groh

The objective of this new program is to show that alterations of the characteristic waveforms of biological oscillations of circadian (about a day) and ultradian (less than a day) periodicities can be used diagnostically to judge the capacity of potentially toxic agents and conditions to cause neurological and behavioral disorders. Emphasis is on the actions of agents that have escaped traditional screening techniques, but that nevertheless pose neurobehavioral health hazards.

Rats are singly housed in automated systems in which inputs (food, light, toxicants) are strictly controlled, and from which physiological information is continuously monitored over prolonged periods. This information shows finely detailed waveforms of physiological oscillations of circadian and ultradian periods that constitute "signatures" that are then used to characterize the functional norm. These signatures are derived from several end points, including core body temperature and gross motor activity; in each instance circadian phase (traditionally referred to as a "time-of-day" effect) is a critical factor, and an altered signature (circadian dyschronism) is an indicator of performance abnormality in the presence of a toxic agent.

New system for circadian data acquisition in the rat. The previously developed 36-channel data acquisition system for circadian data has been expanded to include continual measurement of body weight and motor activity, along with the earlier end points of body temperature and food consumption (in collaboration with W. J. Eisler, Jr., D. A. LeBuis, J. A. Blomquist, and C. A. Fox). Animal weight, weight change, and activity are continuously monitored by supporting the animal cage bottom from a precision load beam force transducer. The transducer puts out an electronic signal that is linearly proportional to the forces applied; this signal is amplified and then split into its AC and DC components. The DC portion of the signal is collected with a long time constant as animal weight, and changes over time enable the determination of animal weight gain or loss under different feeding or other entrainment protocols. The AC portion of the signal is amplified, half-wave rectified, and integrated over time to measure the gross motor activity.

The expansion of our capabilities enables the identification of agent-induced "internal desynchronization" of circadian rhythms among any of the four end points whenever this disorder occurs. The two variables of temperature and activity, which are closely correlated in a normal animal,

are particularly useful indicators of desynchronization. The system also permits the retention of large quantities of data collected at high frequencies over long periods.

Circadian dyschronogenic actions of lithium and cadmium in normal and adrenalectomized rats. The expanded data acquisition system has been used in two experimental series that compared the circadian and ultradian waveforms of normal and adrenalectomized rats exposed to low doses of lithium and cadmium (as low as one-tenth, or less, of the conventional LD_{50} values) with the waveforms of unexposed animals. In these two series, the lithium and cadmium were administered by osmotic pumps (Alzet, subcutaneous) or in drinking water. Recordings were made of weight, activity, and body temperature at 2-minute interrogation intervals, and of food consumption at 15-minute intervals, to yield the waveforms for each function. In the groups exposed to cadmium, dramatic distortions of all recorded waveforms occurred, especially in the adrenalectomized animals. In the highest dose groups exposed to lithium slight changes occurred. In a third series, now underway, the metals are being administered in the food at dose levels approaching the LD_{50} values.

In conclusion, we have revised and expanded our circadian data acquisition system to include measures of weight and activity along with those of temperature and food consumption, and to allow the collection and retention of data at high speed and in very large quantities. Using this new system, normal and distorted waveforms of core temperature and motor activity have been recorded for rats with and without adrenal glands, and for those exposed to low doses of lithium or cadmium.

PUBLICATIONS APPEARING DURING THE NOTED PERIODS

1981

Circadian Variations in the Activity of Tyrosine Hydroxylase, Tyrosine Aminotransferase, and Tryptophan Hydroxylase: Relationship to Catecholamine Metabolism
A. L. Cahill and C. F. Ehret
J. Neurochemistry 37, 1109-1115 (1981)

Chronotypic Induction of Tyrosine Aminotransferase by α-Methyl-p-tyrosine
A. L. Cahill, S. M. Ferguson, and C. F. Ehret
Life Sciences 28, 1665-1671 (1981)

New Approaches to Chronohygiene for the Shift Worker in the Nuclear Power Industry
C. F. Ehret
Advances in the Biosciences Vol. 30: Night and Shift Work: Biological and Social Aspects, A. Reinberg, N. Vieux, and P. Andlauer, eds., 1981, pp. 263-270

Effects of Intermittent 60-Hz High Voltage Electric Fields on Metabolism, Activity, and Temperature in Mice
R. S. Rosenberg, P. H. Duffy, and G. A. Sacher
Bioelectromagnetics 2, 291-303 (1981)

1982

α-Methyl-p-tyrosine Shifts Circadian Temperature Rhythms
A. L. Cahill and C. F. Ehret
Am. J. Physiol. 243 (Regulatory Integrative Comp. Physiol. 12), R218-R222 (1982)

The Chronopharmacology of L-DOPA: Implications for Orthochronal Therapy in the Prevention of Circadian Dyschronism
C. F. Ehret, J. C. Meinert, and K. R. Groh
Chronopharmacology and Chronotherapeutics, C. A. Walker, K. F. A. Soliman, and C. M. Winget, eds., Florida A & M University Foundation, Tallahassee, FL, 1981, pp. 47-65

Glucocorticosteroid Injection Is a Circadian Zeitgeber in the Laboratory Rat
N. D. Horseman and C. F. Ehret
Am. J. Physiol. 243 (Regulatory Integrative Comp. Physiol. 12), R373-R378 (1982)

Effects of Intermittent 60-Hz High Voltage Electric Fields on Metabolism, Activity, and Temperature in Mice
R. S. Rosenberg, P. H. Duffy, and G. A. Sacher
Bioelectromagnetics 2, 291-303 (1981)

1983 through April

In press as of May 1, 1983

Chronobiological Consequences of Various Shiftwork Schedules
A. L. Cahill and C. F. Ehret
Proceedings of the XVth International Conference of the International Society for Chronobiology, Minneapolis, MN, September 13-18, 1981, S. Karger, New York

Tissue Distribution of Cadmium and Metallothionein as a Function of Time of Day and Dosage
A. L. Cahill, D. Nyberg, and C. F. Ehret
Environ. Res.

Circadian Manifestations of Barbiturate Habituation, Addiction, and Withdrawal in the Rat
C. F. Ehret, C. Peraino, J. C. Meinert, and K. R. Groh
Intl. J. Chronobiol.

Relationship between Field Strength and Arousal Response in Mice Exposed to 60-Hz Electric Fields

R. S. Rosenburg, P. H. Duffy, G. A. Sacher, and C. F. Ehret

Bioelectromagnetics

A P P E N D I C E S

18. SUPPORT FACILITIES

ANIMAL FACILITIES

T. E. Fritz, J. M. Angerman, W. G. Keenan, L. S. Lombard, C. M. Poole, A. R. Sallese, and D. V. Tolle

The Laboratory Animal Facilities are a major resource of the Division and occupy approximately 34% or 40,000 square feet of the space in the Biology Building. They consist of four major components: (1) dog kennels with a capacity for approximately 700 dogs; (2) two building wings, Wings E and Q, consisting of 47 animal rooms for housing small laboratory animal species; (3) service areas which include equipment for cleaning and sanitizing cages, water bottles, and cage racks, as well as storage space for food and equipment; and (4) technical equipment areas which include examination and procedure rooms, a surgical operating room, necropsy room, diagnostic X-ray rooms, and "hot room" for handling radioactive materials. During the past year an injection room for treating animals with toxic chemicals and carcinogens was designed and equipped as an added resource in the Animal Facilities.

In addition to the resources listed above, three of the four Divisional ^{60}Co gamma ray exposure facilities, dedicated to whole-body irradiation studies in animals, are physically related to the animal facilities.

During 1982, approximately half of the staff members of the Division were involved in various studies which utilized over 18,000 rodents and other laboratory animal species.

The supporting facilities for the husbandry and clinical monitoring of the resident animals include the following (responsible personnel in parentheses):

Automated cage and bottle washing and filling machinery (Animal Care Specialists)

Steam and gas autoclaves for sterilization of equipment, instruments, and media (Animal Care Specialists and scientific staff)

Diagnostic X-ray facilities and darkrooms (C. M. Poole, W. G. Keenan)

Clinical pathology laboratory (D. V. Tolle)

Hematology laboratory (D. V. Tolle)

Necropsy laboratory (T. E. Fritz)

Histopathology laboratory (T. E. Fritz, L. S. Lombard, A. R. Sallese)

Surgical suite with inhalation anesthesiology equipment (C. M. Poole, W. G. Keenan)

An important aspect of the success of any animal research program is the health of its animal population. To assure a supply of high quality animals, the Facility has concentrated on breeding its own healthy, disease-free animals. Most of the rodents, particularly mice, and all dogs are bred in the Facility. The rodent breeding is managed by J. M. Angerman. The beagle breeding is supervised by C. M. Poole and W. G. Keenan, who are also responsible for the clinical and surgical care of the dogs.

The beagle colony, used primarily for low-level radiation studies in dogs (Chapters 6 and 7) has been a closed colony since 1960. Extensive computerized records are maintained under the supervision of D. E. Doyle on all aspects of the colony, including reproduction, genetics, hematology, pathology, and disease incidence.

As important as the physical plant and its support facilities is the staff available to manage, monitor, treat, and evaluate the animals. The personnel provide a complete range of services to users of the experimental animals. Care of the animals is performed by a group of Animal Care Specialists, listed below, under the supervision of E. W. Jackson.

The Animal Care Specialists during 1982 were the following:

Claude C. Colegrove, Group Leader
William G. McDade, Jr., Group Leader
Adrian J. Cordova
Mose Burrell
Charles J. Fowler
MaDonna Gandalovics
Rita Girard
Carrey R. Herringer
Robert A. Herringer
Betty L. McKay
Richard A. Santarelli
Rika Simmons
Leon L. Stewart
Diane M. Thomas
Jose A. Torres
Joseph N. Wilson

RADIATION FACILITIES

G. L. Holmblad, J. L. Hulesch, J. E. Trier, and F. S. Williamson

The Divisional radiation facilities include a number of gamma, neutron, and X-ray radiation sources with accompanying areas for related equipment and for preparing, handling, and servicing animals. These sources are described in detail below. There are five ^{60}Co irradiation facilities which provide a unique and versatile gamma-ray irradiation capability. A research reactor (JANUS) is a source of fission-spectrum neutrons dedicated to biological research. The gamma-ray facilities are suitable for long-term studies in which animals can be maintained in the radiation field for duration of life or for acute brief or fractionated exposures. They provide a choice of exposure rates ranging from 0.004 to 2×10^4 R/minute. The mean energy of the JANUS neutrons is 0.85 MeV, and the available dose rates range between 0.002 to 80 rads/minute. The contamination from gamma radiation is remarkably low, between 3 and 4 percent of the total absorbed dose. X-ray facilities consist of two X-ray machines, one for experimental irradiations and one for clinical diagnostic purposes. Research using the Divisional radiation facilities is described principally in Chapters 6, 7, and 8 of this report.

In addition to the radiation sources maintained in the Division for the irradiation of biological materials, two other neutron sources in the Chicago area are available to the staff for cell biology studies comparing neutron beams with different characteristics: neutrons from the linear accelerator at the Cancer Therapy Facility of the Fermi National Accelerator Laboratory ($p^+ \rightarrow$ Be, mean energy 25 MeV) and cyclotron produced neutrons ($d^+ \rightarrow$ Be, mean energy 3.6 MeV) at the Franklin McLean Research Institute, The University of Chicago.

^{60}Co Gamma Radiation

F-005 Gamma Room Radiation Field Dosimetry

Cavity configuration. Exposure rates from 500 to 16,000 R/minute with ± 2% variation within volumes as large as 25 cubic centimeters.

Panoramic configuration. Exposure rates from 20 mR/minute to 8 R/minute with a worst case deviation of 7% from the mean within an arc large enough to contain 500 small rodents. Exposure rates as high as 30 R/minute can be achieved with up to 80 animals.

F-101 Gamma Room Radiation Field Dosimetry

Panoramic configuration. Exposure rates from 1 mR/minute to 30 mR/minute by varying distance and attenuation. Up to 500 small rodents may be irradiated simultaneously at four different exposure rates with a worst case deviation of 7% from the mean. Animals are maintained in individual plastic cages, with food and water, on a frame with an N x 5 matrix. N varies from 4 to 8 depending on the frame distance from the source.

F-114 Gamma Room (intermediate) Radiation Field Dosimetry

Panoramic configuration. Exposure rates from 5 R/day to 40 R/day by varying distance and attentuation. Up to 52 dogs may be irradiated simultaneously at four different exposure rates.

X and Y Gamma Rooms Radiation Field Dosimetry

Panoramic configuration. In room X, exposure rates of 1.0 R/day and 2.5 R/day on two arcs, each of which can contain up to 50 individually caged dogs. In room Y, exposure rate of 0.4 R/day on one arc which can contain up to 100 individually caged dogs.

Fission-Spectrum Neutron Radiation

JANUS Reactor Radiation Field Dosimetry

Panoramic configuration. Dose rates from 0.002 to 15 rads/minute over a matrix of 400 mice with a worst case deviation of 8% from the mean. The gamma dose contribution is only 3-4% of the total absorbed dose.

Cavity configuration. A restricted room area near the converter plate may be used for irradiating small samples of cells at up to 65 rads/minute with good uniformity.

X-Radiation

General Electric Maxitron 250-kV X-ray machine, a general Divisional facility.

Westinghouse Flurodex "300" 130-kV X-ray unit equipped with a Machlett, Dynamax-40, rotating anode, dual focal spot tube. Used for clinical and diagnostic purposes for small animals and dogs.

19. EDUCATIONAL ACTIVITIES

POSTGRADUATE TRAINING

During 1982, a total of 18 postdoctoral appointees and research associates contributed to the research programs of the Division. Five of these were new appointments in 1982, while seven finished their assignments during the year.

The temporary appointees, their schools, and the staff members with whom they were affiliated were as follows:

Bruce A. Carnes*	University of Kansas	M. E. Ginevan
Chong-Hwan Chang	University of Pittsburgh	M. Schiffer
Michael L. Garriott	Purdue University	D. Grahn
Colin K. Hill†	Brunel University, Middlesex, England	A. Han
Ross E. Jones	Michigan State University	C. A. Reilly, Jr.
Peter A. Lagocki	University of Chicago	Y. E. Rahman
James M. Leonardo	Temple University	M. J. Peak
Etienne A. Malvoisin	University of Louvain, Brussels, Belgium	E. Huberman
Shin-ichi Murao	Kobe University, Kobe, Japan	E. Huberman
Jennifer G. Peak	Rhodes University	H. E. Kubitschek
Paul R. Reynolds	University of Virginia	H. E. Kubitschek
Reto A. Schwendener	Swiss Federal Institute of Technology, Zurich, Switzerland	Y. E. Rahman
Michael T. Short	University of Illinois at the Medical Center	M. Schiffer
Ido Simon	Weizmann Institute of Science, Rehovat, Israel and the University of Iowa	E. Huberman
Fumio Suzuki	Kanazawa University	A. Han
Pappannan Thiyagarajan	University of Illinois at the Medical Center	C. S. Borso
Robert L. Wells	Colorado State University	A. Han
Karen E. Willard	Virginia Polytechnic Institute and State University	N. G. Anderson

*Now Assistant Statistician.
†Research Associate.

FACULTY RESEARCH PARTICIPATIONS

Argonne and the Division have long had a tradition of involving faculty members from colleges and universities, and staff from industrial laboratories, in Argonne activities. During 1982, there were in the Division two Visiting Scientists and five appointments from the Faculty Research Participation Program and the Faculty Research Leave at Argonne Program. The two latter programs are administered through the Argonne Division of Educational Programs. The program for Faculty Research Leave at Argonne was initiated on a pilot scale in 1980; during 1982 four faculty members participated in the program in the Division. These appointments enable faculty members to participate in the research activities of the Laboratory in order to broaden their perspectives, and they allow university and Argonne scientists to develop rapport and explore mutual interests. The names of the Visiting Scientists, Faculty Research Participants, and Faculty Research Leave at Argonne participants during 1982, their schools, and their staff sponsors were as follows:

William W. Baldwin*	Indiana University School of Medicine, Northwest Center, Gary	H. E. Kubitschek
William E. Boernke†	Nebraska Wesleyan University	C. Peraino
Reinhard Graetzer	Pennsylvania State University	H. E. Kubitschek
Randolph Grayson	California Polytechnic State University	E. Huberman
Richard M. Hyslop	Illinois Benedictine College	M. MacCoss/ C. S. Borso
Donald K. Jasper	Illinois Institute of Technology	T. E. Fritz
Pamela M. Moehring	Mount Mary College, Milwaukee	M. J. Peak
J. Emory Morris*	State University College of New York, Brockport	C. Peraino
Marvin Stodolsky†	Bosphorus University, Istanbul, Turkey	E. Huberman
Robert W. Tuveson*	University of Illinois at Urbana	M. J. Peak
Bartlett D. Whelton*	Eastern Washington University	M. H. Bhattacharyya

GRADUATE PROGRAMS

Three graduate students were Laboratory Graduate Participants working in the Division on Research for their Ph.D. degrees in a program administered and supported by the Division of Educational Programs. The Laboratory Graduate Participants, their schools, and their staff sponsors were as follows:

Mary E. Schackelford	University of Illinois at Chicago Circle	M. H. Bhattacharyya

*Scientist in Residence (Faculty Research Leave at Argonne Appointee).
†Visiting Scientist.

Frederick J. Tremmel	University of Iowa	C. A. Reilly, Jr.
Mary A. Turner	University of Missouri	M. J. Peak

A related program, called Thesis Parts, is also supported by the Division of Educational Programs. It enables graduate students to perform pertinent parts of their research at Argonne. In 1982, one student held this appointment in the Division:

James A. Brainer	Illinois Institute of Technology	T. E. Fritz

In addition, two graduate students carried out research in the Division as Resident Student Associates, supported directly by the Division:

Lynda L. Lanning	Auburn University	T. E. Fritz
Karl M. Rogers	University of Pittsburgh	Y. E. Rahman

UNDERGRADUATE TRAINING

During 1982, a total of 22 college undergraduates received training in the Division of Biological and Medical Research through the Spring, Summer, and Fall Undergraduate Research Participation Programs, sponsored by the Division of Educational Programs. The students, their schools, and their staff supervisors are listed below:

Spring Program

MaryAnn Fink	St. Xavier College	F. R. Kirchner
Andrew S. Gulczynski	Northeastern Illinois College	C. F. Ehret
Carol C. Johnson	Stetson University	D. V. Tolle
Steven J. Mick	Illinois State University	H. E. Kubitschek
Mary F. Muraski	Purdue University	R. E. Jones
James T. Olesen	University of Wisconsin, Green Bay, WI	H. E. Kubitschek
Victoria N. Smith	Goucher College	A. Han
Lauri J. Sammartano	St. Mary's College, Notre Dame, IN	M. J. Peak
Shannon S. Samuels	Nebraska Wesleyan University	M. J. Peak

Summer Program

Ron D. Benioff	University of Illinois at Urbana	M. Schiffer
Renee M. Bressette	College of St. Benedict	C. A. Reilly, Jr.
Mary Chivers	Boise State College	T. M. Seed
Donna M. Eychner	State University of New York at Cortland	B. A. Carnes
MaryAnn Fink*	St. Xavier College	F. R. Kirchner

*Also participant in the Spring Undergraduate Research Participation program.

Joyce A. Guzik	Cornell College	C. S. Borso
Sean M. Kerwin	University of Notre Dame	M. J. Peak
Stephen J. Mick*	Illinois State University	H. E. Kubitschek
Mary F. Muraski*	Purdue University	F. R. Kirchner
James T. Olesen*	University of Wisconsin, Green Bay, WI	H. E. Kubitschek

Fall Program

Kelly Greene	Carroll College	T. E. Fritz
Jennifer Hines	Oberlin College	H. E. Kubitschek
Helen M. Kane	Immaculata College	T. M. Seed
David L. Redman	Ohio Wesleyan University	A. Han
Charles M. Tompkins	Morris Brown College	F. R. Kirchner
Rebecca J. Wechsler	Colorado State University	F. R. Kirchner
Kimberly A. Weyand	Heidelberg College	D. V. Tolle

JOINT ARGONNE-UNIVERSITY APPOINTMENTS

During 1982, 14 staff members held a total of 16 faculty appointments at universities in the Chicago area. These appointments usually comprise limited teaching activities at the graduate level, generally of a specialized nature, which involve regular contact with students. They have led to cosponsorship of graduate students and to collaborative research efforts with faculty members, some of which are described in this report.

The affiliations with Chicago area universities were as follows:

University of Chicago

Mortimer M. Elkind
Eliezer Huberman

Loyola University

Maryka H. Bhattacharyya
Thomas E. Fritz

*Also participant in the Spring Undergraduate Research Participation program.

Northern Illinois University

Maryka H. Bhattacharyya
Thomas E. Fritz
Douglas Grahn
Herbert E. Kubitschek
Carl Peraino
Yueh-Erh Rahman
Christopher A. Reilly, Jr.
John F. Thomson
Robert B. Webb

University of Health Sciences/Chicago Medical School

Antun Han

University of Illinois at the Medical Center

Malcolm MacCoss

Northwestern University

Marianne Schiffer

In addition to holding these formal appointments, Divisional staff have active collaborations with faculty in these and other schools. A recent tally of research collaborations and interactions of Divisional staff with university staff in fiscal year 1982 contained a total of 85 listings; almost 50 were in a category of collaborations that have either resulted, or probably will result, in joint scientific publications.

20. DIVISIONAL SEMINARS, 1982

The Division of Biological and Medical Research Seminar Committee for 1982 consisted of A. Han, Chairman, M. MacCoss, M. J. Peak, M. H. Bhattacharyya, T. M. Seed, and D. A. Haugen.

The General Seminar Program for 1982 consisted of a combination of in-house and visiting speakers, selected on the basis of recommendations from the staff. In addition, a number of informal seminars in specialized subjects were held during the year and during the summer period when the general seminars were not scheduled; some of these informal talks are indicated in the following listing.

VISITING SPEAKERS

Dr. Paul B. Fisher, Columbia University, College of Physicians and Surgeons, New York, NY
"Multiple Interactions and Adenovirus Transformation"
January 7, 1982

Dr. Jonathan Greer, Department of Biological Sciences, Sherman Fairchild Center for the Life Sciences, Columbia University, New York, NY
"Model Structures of Blood Proteins, Haptoglobin-Hemoglobin and Blood Clotting Factor Xa-Prothrombin"
January 13, 1982

Dr. Alan Epstein, Department of Medicine, Medical Oncology Section, Northwestern Medical School, Chicago, IL
"Classification Scheme for Nuclear Proteins Based on Their Immunofluorescence Staining Patterns using Monoclonal Antibodies"
January 21, 1982

Professor Donald J. Fluke, Zoology Department, Duke University, Durham, NC
"Mutagenesis and Damage Inducible Functions in *E. coli*"
February 4, 1982

Dr. Niza Frenkel, Department of Biology, Cummings Life Science Center, University of Chicago, Chicago, IL
"The Herpes Simplex Virus Amplicon - A New Eukaryotic Virus Cloning Vector"
February 11, 1982

Dr. Margaret L. Kripke, Frederick Cancer Research Center, Frederick, MD
"Immunological Aspects of Tumor Induction by Ultraviolet Light"
March 11, 1982

Dr. Robert C. Nowinski, Genetics Systems Corporation, Seattle, Wa
"Diagnosis of Herpes Virus and Other Sexually Transmitted Diseases in Humans with Monoclonal Antibodies"
March 17, 1982

Professor M. Roberfroid, Biochemical Toxicology and Carcinogenesis Group, School of Pharmacy, Faculty of Medicine, Catholic University of Louvain, Brussels, Belgium
"Chemical Promotion of Hepatocarcinogenesis; Rapid Production, Isolation, and Biochemical Characterization of Hyperplastic Nodules"
March 23, 1982

Dr. Robert A. Weinberg, Massachusetts Institute of Technology, Scientific Center for Cancer Research, Cambridge, MA
"Oncogenes of Human Tumor Cells"
April 22, 1982

Dr. Daniel W. Nebert, Developmental Pharmacology Branch, NIH, Bethesda, MD
"Cloning the Genes Encoding Enzymes Which Metabolize Drugs and Chemical Carcinogens"
May 5, 1982

Dr. Thomas P. Coohill, Physics Department, Western Kentucky University, Bowling Green, KY
"Ultraviolet Action Spectroscopy in Mammalian Cells and Viruses"
May 13, 1982

Dr. Heinz Kohler, Department of Molecular Immunology, Roswell Park Memorial Institute, Buffalo, NY
"Regulation of Immune Responses Through Idiotypes"
May 20, 1982

Dr. A. Leonard, Center for Nuclear Energy, Brussels, Belgium
"Cytogenetic Effects of Ionizing Radiations in Somatic Cells from Experimental Mammals, and Extrapolation to Man"
May 21, 1982

Dr. Hans-Georg Schweiger, Max-Planck Institute for Cell Research, Heidelberg, West Germany
"Recent Developments in the Molecular Biology of Acetabularia"
June 3, 1982

Dr. Wen K. Yang, Oak Ridge National Laboratory, Biology Division, Oak Ridge, TN
"New Concepts of Retroviral Genes in Relation to Oncogenesis"
July 1, 1982

*Dr. J. Scott Klingensmith, University of Mississippi Medical Center, Jackson, MS
"The Role of Metabolism in the Potentiation of CCl_4 Toxicity"
July 20, 1982

*Dr. Lynn K. Pershing, University of Utah, Salt Lake City, UT
"Induction of Hepatic Cytochrome P-450 by Macrolide Antibiotics"
July 21, 1982

Dr. Frank T. Robb, Microbiology Department, University of Cape Town, Cape Town, South Africa
"Mechanisms and Regulation of Exoprotease Nitrogen Acquisition Systems in Bacteria"
August 12, 1982

Professor Taketoshi Sugiyama, Department of Pathology, Kobe University School of Medicine, Chuo-ku, Kobe, Japan
"Cancer and Chromosome Abnormality"
September 16, 1982

Dr. John Jagger, University of Texas at Dallas, School of General Studies, Richardson, TX
"Responses of Cells to Solar Ultraviolet Radiation"
September 23, 1982

*Dr. Tenlin Tsai, Packard Instrument Company, Chicago, IL
"The Analytical and Biomedical Applications of Bioluminescence"
October 6, 1982

Dr. Conrad Woldringh, Department of Microbiology, University of Texas, Austin, TX
"Cell Growth and Division in _E. coli_"
October 7, 1982

Dr. William H. Knospe, Section of Clinical Hematology, Rush-Presbyterian St. Luke's Medical Center, Chicago, IL
"Immunologically Mediated Aplastic Anemia in Mice"
October 14, 1982

*Dr. Genevieve Krack, Biochemical Toxicology and Carcinogenesis Group, School of Pharmacy, Faculty of Medicine, Catholic University of Louvain, Brussels, Belgium
"Metabolic Competence of Isolated Hepatocytes in Suspension: A New Tool for _in Vitro_ Toxicological Evaluation"
October 22, 1982

---

*Informal seminar.

Dr. Carmia Borek, Radiological Research Laboratory, Columbia University, New York, NY
"The Neoplastic Transformation in Vitro of Rodent and Human Cells and Its Modulation"
October 28, 1982

Dr. Eugene Cronkite, Medical Department, Brookhaven National Laboratory, Upton, NY
"Regulation of Hemopoiesis"
November 4, 1982

*Dr. Robert L. Ullrich, Biology Division, Oak Ridge National Laboratory, Oak Ridge, TN
"Mammary Tumorigenesis in Mice after Exposure to Ionizing Radiation"
November 8, 1982

Dr. William A. Haseltine, Sidney Farber Cancer Institute, Harvard Medical School, Harvard University, Boston, MA
"New Methods for Detection of Damage to Human DNA by Ultraviolet and Ionizing Radiations"
November 11, 1982

*Dr. Ivan Rich, Department of Biochemistry, University of Chicago, Chicago, IL
"Role of Interleukin-3 in Hemopoiesis"
November 15, 1982

Dr. Peter Blumberg, Laboratory of Cellular Carcinogenesis and Tumor Promotion, National Cancer Institute, Bethesda, MD
"Studies on the Receptors for the Phorbol Ester Tumor Promoters"
November 18, 1982

Dr. Thomas J. MacVittie, Chairman, Experimental Hematology Department, Armed Forces Radiobiology Research Institute, Defense Nuclear Agency, Bethesda, MD.
"Mechanisms of Combined Radiation and Microbial Injury"
December 9, 1982

IN HOUSE SPEAKERS

Dr. Meyrick J. Peak, Division of Biological and Medical Research, Argonne National Laboratory, Argonne, IL
"Biological and Molecular Effects of Long Wavelength Ultraviolet Radiations"
January 28, 1982

*Informal seminar.

Dr. Antun Han, Division of Biological and Medical Research, Argonne National Laboratory, Argonne, IL
"Mutation and Neoplastic Transformation of Mammalian Cells by Ultraviolet Light"
February 25, 1982

Dr. Thomas M. Seed, Division of Biological and Medical Research, Argonne National Laboratory, Argonne, IL
"Hematological Responses under Continuous, Low Dose Irradiation: Mechanisms of Accommodation and Possible Relationships to Leukemia Induction"
June 17, 1982

Mr. David V. Tolle, Division of Biological and Medical Research, Argonne National Laboratory, Argonne, IL
"Radiation-Induced Leukemia and Related Hemoproliferative Disorders in the Dog: Hematology and Pathology"
September 30, 1982

Dr. David A. Haugen, Division of Biological and Medical Research, Argonne National Laboratory, Argonne, IL
"Identification of Mutagens in Coal-Derived Synthetic Fuel Materials"
October 21, 1982

Dr. Marvin Stodolsky, Visiting Scientist, Division of Biological and Medical Research, Argonne National Laboratory
"Chromosomal Elements as Mutagens"
December 2, 1982

21. OUTSIDE TALKS BY DIVISIONAL STAFF DURING 1982*

Anderson, N. G. "The Human Protein Index." Upjohn Co., Kalamazoo, MI, February 10, 1982.

Anderson, N. G. "The Techniques of 2-D Electrophoresis." Department of Urology, Northwestern University, Evanston, IL, April 15, 1982.

Anderson, N. G. "High Resolution Protein Separation and Identification Methods Applicable to Virology." New Horizons in Diagnostic Virology, World Health Organization Meeting, Munich, Germany, June 3, 1982.

Anderson, N. G. "Development of Preparative Analogues of the ISO-DALT System." Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 30, 1982.

Anderson, N. G. "The Development of Centrifugal Analyzers." 14th Congress of the Japan Society of Clinical Laboratory Automation, Kobe, Japan, September 10, 1982.

Anderson, N. G. "The Development of Systems for Mutation Rate Measurements and for Clinical Applications." Radiation Effects Research Foundation, Hiroshima, Japan, September 13, 1982.

Anderson, N. G. "Human Milk Proteins." 28th Annual Midwest Milk and Food Protein Seminar, Northbrook, IL, September 24, 1982.

Anderson, N. G. "Introduction to High Resolution Two-Dimensional Electrophoresis in Clinical Chemistry." Colloque International, Biologie Prospective, Nancy, France, October 4, 1982.

Anderson, N. L. "High Resolution Protein Mapping." Upjohn Company, Kalamazoo, MI, February 10, 1982.

Anderson, N. L. "Protein Mapping and Human Genetic Diseases." 1982 International Conference of the Association for Children with Learning Disabilities, Chicago, IL, March 3, 1982.

Anderson, N. L. "The Human Protein Index." Sigma Xi Lecture, Argonne, IL, March 18, 1982.

---

*Not included in this listing of outside talks are lectures given as part of courses in area universities or in the educational programs at Argonne National Laboratory.

Anderson, N. L. "Making a Catalog of Human Proteins." Physics Division Colloquium, Argonne, IL, April 9, 1982.

Anderson, N. L. "Cataloging Human Proteins." Electrophoresis '82, Athens, Greece, April 23, 1982.

Anderson, N. L. "Catalogs of Human Proteins." Pasteur Institute, Paris, France, April 27, 1982.

Anderson, N. L. "Mapping of Human Proteins." Monsanto Company, St. Louis, MO, July 7, 1982.

Anderson, N. L. "Variants of Human Cellular Proteins." (poster presentation) Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 29, 1982.

Anderson, N. L. "New Iso-Dalt 8" x 10" System: Better Resolution and Adaptable to Manufacture." Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 30, 1982.

Anderson, N. L. "Differentiation in the Human Promyelocytic Cell Line HL-60." (poster presentation) Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 30, 1982.

Anderson, N. L. "Evolutionary Comparison of Cellular Proteins by Two-Dimensional Electrophoresis." (poster presentation) Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 30, 1982.

Anderson, N. L. "Analysis of Human Leukemias by 2-D Electrophoresis." Sloan-Kettering Memorial Institute, New York, NY, September 10, 1982.

Anderson, N. L. "Mapping of Human Proteins." Proctor and Gamble Research Laboratories, Cincinnati, OH, September 23, 1982.

Anderson, N. L. "2-D Mapping of Human Proteins." Basel Institute for Immunology, Basel, Switzerland, October 1, 1982.

Anderson, N. L. "2-D Mapping of Human Proteins." Colloquium "Biologie Perspective," Nancy, France, October 4, 1982.

Anderson, N. L. "2-D Mapping of Human Proteins." World Health Organization Symposium on Parasite and Vector Identification, Bromma, Sweden, November 12, 1982.

Anderson, N. L. "Image Analysis and Database Systems for Cataloguing Human Proteins Separated by 2-D Electrophoresis." Computers in Science, Chicago, IL, December 8, 1982.

Bhattacharyya, M. H. "Influence of Pregnancy and Lactation on Gastrointestinal Absorption of Cadmium and Lead." University of Rochester, meeting on Reproductive and Developmental Toxicity of Metals, Rochester, NY, May 25, 1982.

Borso, C. S. "Implementation of Self-Scanning Photodiode Arrays for Scattering Applications Utilizing Conventional and Intense X-Ray Sources." Winter Meeting of the American Crystallographic Association, Gaithersburg, MD, March 29, 1982.

Doyle, D. E. "A Multi-Parameter, On-Line Data-Access System for a Long Term Animal Research Project." 11th Annual Chicago-Southern Wisconsin Interbranch Meeting of the American Association for Laboratory Animal Science, Chicago, IL, September 17, 1982.

Ehret, C. F. "Neurobehavioral Chronobiology and Toxicology." Behavioral Toxicology Round Table, Health Effects Research Division, DOE, Washington, DC, March 9, 1982.

Ehret, C. F. "Chronobiotic Action of Catecholamines and Catecholamine Biosynthesis Inhibitors in the Mammal." Department of Biochemistry and Biophysics, Loyola University School of Medicine, April 26, 1982.

Ehret, C. F. "Relief from Jet Lag through the Orthochronal Application of Chronobiotics and other Zeitgebers." Fall Meeting of the Midwest Association for the Study of Sleep, Chicago Health Science Center, University of Illinois, Chicago, IL, September 4, 1982.

Ehret, C. F. "Food and Drugs as Circadian Zeitgebers." West Suburban Dietetic Association, Naperville, IL, October 5, 1982.

Ehret, C. F. "Chemical Bases and Control of Circadian Biological Rhythms: Applications for Shift Workers and for World Travelers." Sigma Xi Lecture, Nalco Chemical Company, Naperville, IL, October 27, 1982.

Ehret, C. F. "A Chronobiological Approach to Shift Work: Circadian Chronohygiene and Advanced ROTAS for the Law Enforcement and Security Professions." Aylmer Conference on Stress and Law Enforcement, Ontario Police College, Aylmer, Ontario, Canada, November 2, 1982.

Fritz, T. E. "RBE of Fission Neutrons for Life Shortening and Tumorigenesis." European Seminar on Neutron Carcinogenesis, Rijswijk, Netherlands, March 30- April 1, 1982.

Fritz, T. E. "Late Pathologic Effects of Protracted ^{60}Co Irradiation of Beagles." 30th Annual Meeting of the Radiation Research Society, Salt Lake City, UT, April 14-22, 1982.

Giometti, C. S. "Tropomyosin in Muscle and Nonmuscle Cells." (poster presentation) Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 29, 1982.

Giometti, C. S. "Radiolabeling of Tissue Proteins for Two-Dimensional Electrophoresis." Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 31, 1982.

Giometti, C. S. "Application of Two-Dimensional Electrophoresis to Clinical Samples." Seminar in Medical Technology II, Andrews University at Hinsdale Hospital, Hinsdale, IL, October 6, 1982.

Giometti, C. S. "Tropomyosin Heterogeneity in Nonmuscle Cells." 22nd Annual Meeting of the American Society for Cell Biology, Baltimore, MD, December 2, 1982.

Grahn, D. "Radiation Physics and Dosimetry." IAEA course entitled Planning, Preparedness, and Response to Radiological Emergencies, Argonne, IL, February 2, 1982.

Grahn, D. "Radiation Biology." IAEA course entitled Planning, Preparedness, and Response to Radiological Emergencies, Argonne, IL, February 2, 1982.

Grahn, D. "Genetic and Cytogenetic Responses to Long Continued, Periodic External Neutron and Gamma Irradiation." 30th Annual Meeting of the Radiation Research Society, Salt Lake City, UT, April 20, 1982.

Grahn, D. "Life Shortening In Peromyscus leucopus Exposed to Fission Neutrons." 30th Annual Meeting of the Radiation Research Society, Salt Lake City, UT, April 21, 1982.

Grahn, D. "Some Uncertainties and Controversies in Predicting the Effects in Man of Low-Level Irradiation." Department of Civil Engineering, Northwestern University, Evanston, IL, May 12, 1982.

Grahn, D. "Genetic Risks Associated with Radiation Exposures during Space Flight." Workshop on Radiation Safety Standards, Committee on Space Research (COSPAR/ICSU), XXIV Plenary Meeting, Ottawa, Canada, May 27, 1982.

Grahn, D. "Biological Effects of Radiation." Nuclear Energy Institute for Secondary Teachers, Northern Illinois University, DeKalb, IL, July 14, 1982.

Grahn, D. "Biological Effects of Low Dose Exposure to Neutrons or Gamma Rays." Components Technology Division, Argonne National Laboratory, Argonne, IL, December 13, 1982.

Haugen, D. A. "Isolation and Identification of Mutagenic Components of Synfuel Materials." DOE Workshop on 5-Year Research Plan, Warrenton, VA, January 26-28 1982.

Huberman, E. "The Control of Cell Differentiation and Mutagenesis by Chemicals which Initiate or Promote Tumor Formation." Symposium on Genetic Mechanisms of Carcinogenesis, Oak Ridge National Laboratory, Gatlinburg, TN, April 11-15, 1982.

Huberman, E. "The Control of Mutagenesis and Cell Differentiation by Chemicals which Initiate or Promote Tumor Formation." Michigan State University, E. Lansing, IL, May 17, 1982.

Huberman, E. "The Control of Mutagenesis in Cultured Mammalian Cells." NCI Workshop Meeting on Mammalian Mutagenesis by Low Doses of Ionizing Radiation, Argonne, IL, June 28, 1982.

Huberman, E. "The Use of Liver Cell Cultures in Mutagenesis Studies." 4th International Course on Tumor Biology, Bogota, Colombia, October 18-23, 1982.

Huberman, E. "The Control of Cell Differentiation by Tumor Promoting Agents." 4th International Course on Tumor Biology, Bogota, Colombia, South America, October 18-23, 1982.

Huberman, E. "The Control of Cell Differentiation and Mutagenesis by Chemicals which Initiate or Promote Tumor Formation." University of Chicago, Chicago, IL, November 9, 1982.

Huberman, E. "The Control of Differentiation in Myeloid and T-cell Leukemia." University of Chicago, Chicago, IL, December 27, 1982.

Kirchner, F. R. "Characteristics of Emissions Produced by Two High-Temperature Combustion Sources." First Annual Conference of the American Association for Aerosol Research, Santa Monica, CA, February 17-19, 1982.

Kirchner, F. R. "Characterization of Fine Particles from Two High Temperature Combustion Sources." 13th Annual Meeting of the Fine Particle Society, Chicago, IL, April 12-14, 1982.

Kirchner, F. R. "Aerosol and Inhalation Toxicology." Toxicology Seminar Series, Associated College of the Chicago Area, Argonne, IL, October 5, 1982.

Kubitschek, H. E. "Zonal Growth of Escherichia coli." 82nd Annual Meeting of the American Society for Microbiology, Atlanta, GA, March 8, 1982.

Kubitschek, H. E. "Are There Growth Laws for Bacteria?" XIII International Congress of Microbiology, Boston, MA, August 8-13, 1982.

Kubitschek, H. E. "Chemostats and Turbidostats: Operation and Potential Difficulties." Miles Laboratories, Elkhart, IN, November 9, 1982.

Lombard, L. S. "A Computerized Coding System for Histopathologic Data Used at Argonne National Laboratory in Chronic Toxicity Studies on Beagle Dogs." Drug Information Association, Chicago, IL, October 19, 1982.

MacCoss, M. "Synthetic, Structural, and Biological Aspects of Phospholipid-Nucleoside Conjugates - A New Type of Prodrug." The Upjohn Company, Kalamazoo, MI, February 16, 1982.

MacCoss, M. "Synthetic, Structural, and Biological Aspects of Phospholipid-Nucleoside Conjugates - A New Type of Prodrug." E. I. Dupont de Nemours, Experimental Station, Wilmington, DE, February 22, 1982.

MacCoss, M. "Synthetic, Structural, and Biological Aspects of Phospholipid-Nucleoside Conjugates - A New Type of Prodrug." Merck, Sharp and Dohme Research Laboratories, Rahway, NJ, March 2, 1982.

MacCoss, M. "Synthetic, Structural, and Biological Aspects of Phospholipid-Nucleoside Conjugates - A New Type of Prodrug." Biophysics Department Roswell Park Memorial Institute, Buffalo, NY, March 15, 1982.

MacCoss, M. "Synthetic, Structural, and Biological Aspects of Phospholipid-Nucleoside Conjugates - A New Type of Prodrug." Chicago Medical School, Evanston, IL, May 13, 1982.

Peak, M. J. "Effect of pH upon Mutagenicity in Escherichia coli." 13th Annual Meeting of the Environmental Mutagen Society, Boston, MA, March 1, 1982.

Peak, M. J. "Role of DNA Single Strand Breaks in Lethal and Mutagenic Events Caused by uvB Radiation." Ministry of Research and Technology, Federal Republic of Germany, Biological Effects of uv-B Radiation, Munich, Germany, May 25-27, 1982.

Peak, M. J. "Role of DNA Breaks in Near Ultraviolet Lethality." Molecular Biology Research Institute, University of Paris, and the Institut Gustave-Roussy, Paris, France, June 2, 1982.

Peak, M. J. "Correlations Between Lethal Events and Single-Strand Breaks in DNA Caused by Near-Ultraviolet Light in Microorganisms." 10th Annual Meeting of the American Society for Photobiology, Vancouver, BC, Canada, June 27-July 1, 1982.

Peak, M. J. "Use of Action Spectra for Identifying Molecular Targets and Mechanisms of Action of Solar Ultraviolet Light." International Workshop on the Effects of Ultraviolet Radiation on Plants, Delhi, India, November 1-5, 1982.

Peraino, C. "Liver as an Experimental System for Analyzing Mechanisms of Tumor Initiation and Promotion." Department of Pathology, Case Western Reserve University, Cleveland, OH, February 17, 1982.

Rahman, Y. E. "Liposomes Targeted to Cellular Receptors." 8th International Forum on Subcellular Methodology, Membrane-Located Receptors, Guildford, England, July 12-17, 1982.

Rahman, Y. E. "Enhanced Therapeutic Effect of Intraperitoneally Administered Liposome-Encapsulated 1-β-D-arabinofuranosylcytosine (ara-C) Against Mouse Lewis Lung Carcinoma." 13th International Cancer Congress, Seattle, WA, September 8-15, 1982.

Reilly, C. A., Jr. "Toxicological Effects of Coal Gasification By-products." DOE/Airlie House Retreat, Health and Environmental Research Program Related to Coal Conversion Technologies and their Future Directions, Warrenton, VA, January 26, 1982.

Reilly, C. A., Jr. "Coal Toxicology." CSUI-ANL Conference, Argonne, IL, September 24, 1982.

Reilly, C. A., Jr. "Status of Coal Gasification Health and Environmental Research." Oak Ridge National Laboratory Fifth Life Services Symposium, Synthetic Fossil Fuel Technologies: Results of Health and Environmental Studies, Gatlinburg, TN, October 24, 1982.

Reilly, C. A., Jr. "Toxicological Testing Procedures." Toxicology Seminar Series, Associated College of the Chicago Area, Argonne, IL, October 5, 1982.

Russell, J. J. "The Retention and Microdistribution of Plutonium in the Mouse Ovary." Health Physics Society Annual Meeting, Las Vegas, NV, June 27-July 1, 1982.

Russell, J. J. "The Effect of Electric Field Exposure on Rat Circadian and Infradian Rhythms." 4th Annual Meeting of the Bioelectromagnetics Society, Los Angeles, CA, June 28-July 2, 1982.

Russell, J. J. "Effects of Intermittent 60-Hz Electric Field Exposure: Circadian Phase Shifts, Splitting, Torpor, and Aerosol Responses in Mice." (poster presentation) 4th Annual Meeting of the Bioelectromagnetics Society, Los Angeles, CA, June 28-July 2, 1982.

Schiffer, M. "Structure Determination of Immunoglobulins Using X-Ray Diffraction." Abbott Laboratories, North Chicago, IL, April 14, 1982.

Schiffer, M. "Structure of Bence-Jones Dimer Loc at 5 Å Resolution." (poster presentation) Gordon Conference on Diffraction Methods in Molecular Biology, Andover, NH, June 28, 1982.

Seed, T. M. "Ultrastructural Effects of Radiation on Cells and Tissues: An Overview." Scanning Electron Microscopy/1982 Annual Meeting, Anaheim, CA, April 26-30, 1982.

Seed, T. M. "Ultrastructure of Radiation-Induced Endosteal Myelofibrosis in the Dog." Scanning Electron Microscopy/1982 Annual Meeting, Anaheim, CA, April 26-30, 1982.

Seed, T. M. "Hematopoietic Responses under Continuous Low Daily Dose Irradiation." Clinical Hematology, Rush Presbyterian-St. Luke's Medical Center, Chicago, IL, May 27, 1982.

Seed, T. M. "Specimen Preparation for Electron Microscopy: (a) Carbon Replication; (b) Freeze-Etching; (c) Cytochemistry and Autoradiography." Division of Educational Programs Faculty Institute, The Principles and Practice of Electron Microscopy, Atlanta University, Atlanta, GA, June 1-10, 1982.

Seed, T. M. "Radiation Sensitivities of Hemopoietic Progenitors: Hemopathological Predisposition and Survival Times under Continuous Low Daily Dose Gamma Irradiation." 11th Congress of the International Society for Experimental Hematology, Baltimore, MD, August 12-15, 1982.

Seed, T. M. "Indicators for the Development of Leukemia in Chronically Irradiated Dogs." Conference on Low-Level Effects of Radiation-Carcinogenesis, Brookhaven National Laboratory, Upton, NY, August 16-18, 1982.

Taylor, J. "The Use of Array Processors in Analyzing Two-Dimensional Electrophoresis Patterns of Protein Samples." 1982 ARRAY Conference, Boston, MA, March 17, 1982.

Taylor, J. "Numerical Measures of 2-D Gel Resolution and Reproducibility." Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 31, 1982.

Taylor, J. "Image Processing." Minicomputer Workshop, The DEC "11" Family of Computers, Argonne National Laboratory, Argonne, IL, November 18, 1982.

Tolle, D. V. "Aberrant Megakaryocytopoiesis Preceding Radiation-Induced Leukemia in the Dog." Scanning Electron Microscopy/1982 Annual Meeting, Anaheim, CA, April 26-30, 1982.

Tolle, D. V. "Defective Megakaryocyte Differentiation in Canine Preleukemia: Cytomorphology, Ultrastructure, and Pathology." 11th Congress of the International Society for Experimental Hematology, Baltimore, MD, August 12-15, 1982.

Tolle, D. V. "Effect of Chronic Irradiation on Megakaryocytes." Conference on Low-Level Effects of Radiation-Carcinogenesis, Brookhaven National Laboratory, Upton, NY, August 16-18, 1982.

Willard, K. E. "The Human Leukocyte Protein Index." Hematology/Oncology Conference, Chicago, IL, January 11, 1982.

Willard, K. E. "The Human Leukocyte Protein Index." Department of Microbiology and Molecular Genetics, Harvard Medical School, Boston, MA, January 15, 1982.

Willard, K. E. "Potential of Two-Dimensional Electrophoresis of Human Leukocyte Proteins for Clinical Diagnosis in Rheumatoid Arthritis, Infectious Mononucleosis, and Leukemia." Conference of the Combined Clinical Staffs, National Institute of Health, Bethesda, MD, February 18, 1982.

Willard, K. E. "Two-Dimensional Gel Analysis of Purified Leukocyte Subpopulations and Related Clinical Applications." Department of Immunology, Pasteur Institute, Paris, France, March 10, 1982.

Willard, K. E. "Two-Dimensional Gels and the Analysis of Human Proteins." Ludwig Institute, Brussels, Belgium, March 12, 1982.

Willard, K. E. "Two-Dimensional Gel Analysis of Purified Leukocyte Subpopulations and Related Clinical Applications." Research Laboratories, Beckman Instruments, Fullerton, CA, April 7, 1982.

Willard-Gallo, K. E. "Leukocyte Subset-Specific Proteins Determined by Fluorescence-Activated Cell Sorting with Subsequent Analysis on 2D Gels." Argonne and Mayo Clinic Symposium, Technical Advances in Two-Dimensional Electrophoresis and Clinical Applications of the Technique, Argonne, IL, August 30, 1982.

22. PUBLICATIONS APPEARING IN CALENDAR YEAR 1982

JOURNAL ARTICLES

Anderson, N. G., and L. Anderson
The Human Protein Index
Clin. Chem. 28, 739-748 (1982)

Anderson, N. G., and N. L. Anderson
The Human Protein Index
J. Am. Med Assoc. 246, 2620-2621 (1981)

Anderson, N. G., and N. L. Anderson
The Human Protein Index: A Prospect for Laboratory Medicine
Lab. Manage. 20, 25-32 (1982)

Anderson, N. G., and N. L. Anderson
The Human Protein Index Project and the Molecular Pathology Data Base
Med. Lab. 11, 75-94 (1982)

Anderson, N. G., M. T. Powers, and S. L. Tollaksen
Proteins of Human Milk. I. Identification of Major Components
Clin. Chem. 28, 1045-1055 (1982)

Anderson, N. L.
Towards a Complete Catalog of Human Proteins
Trends in Anal. Chem. 1, 131-134 (1982)

Anderson, N. L., C. S. Giometti, M. A. Gemmell, S. L. Nance, and N. G. Anderson
A Two-Dimensional Electrophoretic Analysis of the Heat-Shock-Induced Proteins of Human Cells
Clin. Chem. 28, 1084-1092 (1982)

Anderson, N. L., S. L. Nance, T. W. Pearson, and N. G. Anderson
Specific Antiserum Staining of Two-Dimensional Electrophoretic Patterns of Human Plasma Proteins Immobilized on Nitrocellulose
Electrophoresis 3, 135-142 (1982)

Anderson, N. L., J. Taylor, A. E. Scandora, B. P. Coulter, and N. G. Anderson
The TYCHO System for Computer Analysis of Two-Dimensional Gel Electrophoresis Patterns
Clin. Chem. 27, 1807-1820 (1981)

Bhattacharyya, M. H., B. D. Whelton, and D. P. Peterson
Gastrointestinal Absorption of Cadmium in Mice during Gestation and Lactation. I. Short-Term Exposure Studies
Toxicol. Appl. Pharmacol. 61, 335-342 (1981)

Boernke, W. E., F. J. Stevens, J. J. Edwards, and C. Peraino
Differential Changes in Ornithine Aminotransferase Self-Affinity Produced by Exposure to Basic Amino Acids and Increases in the Intrinsic Electronegativity of the Enzyme Monomer
Arch. Biochem. Biophys. 216, 152-157 (1982)

Borso, C. S.
Optimization of Monolithic Solid State Array Detectors for the Position Encoding of Small Angle X-Ray Scattering from Synchrotron Sources
Nucl. Instrum. Methods 201, 65-71 (1982)

Borso, C. S., J. M. Carpenter, F. S. Williamson, G. L. Holmblad, M. H. Mueller, J. Faber, Jr., J. E. Epperson, and S. S. Danyluk
Small-Angle Scattering at a Pulsed Neutron Source: Comparison with a Steady-State Reactor
J. Appl. Cryst. 15, 443-448 (1982)

Cahill, A. L., and C. F. Ehret
α-Methyl-p-tyrosine Shifts Circadian Temperature Rhythms
Am. J. Physiol. 243 (Regulatory Integrative Comp. Physiol. 12), R218-R222 (1982)

Carnes, B. A., and N. A. Slade
Some Comments on Niche Analysis in Canonical Space
Ecology 63, 888-893 (1982)

Collins, J. J.
The Contribution of Medical Measures to the Decline of Mortality from Respiratory Tuberculosis: An Age-Period-Cohort Model
Demography 19, 409-427 (1982)

DeMark, B. R., G. T. Everson, P. D. Klein, R. B. Showalter, and F. Kern, Jr.
A Method for the Accurate Measurement of Isotope Ratios of Chenodeoxycholic and Cholic Acids in Serum
J. Lipid Res. 23, 204-210 (1982)

Doyle, D. E., L. S. Lombard, and T. E. Fritz
A Direct Data Entry System for Toxicologic Pathology
Toxicol. Pathol. 9, 22-29 (1981)

Edwards, J. J., N. G. Anderson, S. L. Tollaksen, A. C. von Eschenbach, and J. Guevara, Jr.
Proteins of Human Urine. II. Identification by Two-Dimensional Electrophoresis of a New Candidate Marker for Prostatic Cancer
Clin. Chem. 28, 160-163 (1982)

Edwards, J. J., S. L. Tollaksen, and N. G. Anderson
Proteins of Human Urine. III. Identification and Two-Dimensional Electrophoretic Map Positions of Some Major Urinary Proteins
Clin. Chem. 28, 941-948 (1982)

Elkind, M. M.
Keynote Address - Modifiers of Radiation Response in Tumor Therapy: Strategies and Expectations
Int. J. Radiat. Oncol. Biol. Phys. 8, 89-100 (1982)

Finkel, M. P.
BOOK REVIEW: Known Effects of Low-level Radiation Exposure. Proceedings of a conference held in April 1979 by the Mideast Center for Radiological Physics at the Allegheny General Hospital, Pittsburgh, PA, P. K. Shrivastava, ed. (U.S. Department of Health, Education, and Welfare, NIH Publication 80-2087, January 1980), pp. 147
Health Phys. 43, 288 (1982)

Garriott, M. L., and D. Grahn
Neutron and γ-Ray Effects Measured by the Micronucleus Test
Mutat. Res. Lettr. 105, 157-162 (1982)

Gemmell, M. A., and N. L. Anderson
Lymphocyte, Monocyte, and Granulocyte Proteins Compared by Use of Two-Dimensional Electrophoresis
Clin. Chem. 28, 1062-1066 (1982)

Giometti, C. S., and N. G. Anderson
Muscle Protein Analysis. III. Analysis of Solubilized Frozen-Tissue Sections by Two-Dimensional Electrophoresis
Clin. Chem. 27, 1918-1921 (1981)

Giometti, C. S., and N. L. Anderson
A Variant of Human Nonmuscle Tropomyosin Found in Fibroblasts by Using Two-dimensional Electrohoresis
J. Biol. Chem. 256, 11840-11846 (1981)

Giometti, C. S., K. E. Willard, and N. L. Anderson
Cytoskeletal Proteins from Human Skin Fibroblasts, Peripheral Blood Leukocytes, and a Lymphoblastoid Cell Line Compared by Two-Dimensional Gel Electrophoresis
Clin. Chem. 28, 955-961 (1982)

Guevara, J., Jr., E. A. Chiocca, F. C. Clayton, A. C. von Eschenbach, and J. J. Edwards
A Simple Method of Eluting Proteins from Two-Dimensional Gels
Clin. Chem., 28, 756-758 (1982)

Han, A., and M. M. Elkind
Enhanced Transformation of Mouse 10T1/2 Cells by 12-O-Tetradecanoyl-phorbol-13-Acetate following Exposure to X-Rays or to Fission-Spectrum Neutrons
Cancer Res. 42, 477-483 (1982)

Hass, B. S., C. K. McKeown, D. J. Sardella, E. Boger, P. K. Ghoshal, and E. Huberman
Cell-mediated Mutagenicity in Chinese Hamster V79 Cells of Dibenzopyrenes and Their Bay-Region Fluorine-substituted Derivatives (ORNL work)
Cancer Res. 42, 1646-1649 (1982)

Hass, B. S., and R. B. Webb
Photodynamic Effects of Dyes on Bacteria: V. Mutagenesis by Acridine Orange and 460-nm or 500-nm Light in Strains of Escherichia coli that Differ in Repair Capability
Mutat. Res. 94, 39-51 (1982)

Haugen, D. A., M. J. Peak, K. M. Suhrbier and V. C. Stamoudis
Isolation of Mutagenic Aromatic Amines from a Coal Conversion Oil by Cation Exchange Chromatography
Anal. Chem. 54, 32-37 (1982)

Hill, C. K., F. M. Buonaguro, C. P. Myers, A. Han, and M. M. Elkind
Fission-Spectrum Neutrons at Reduced Dose Rates Enhance Neoplastic Transformation
Nature 298, 67-69 (1982)

Hoffman, D. R., and E. Huberman
The Control of Phospholipid Methylation by Phorbol Diesters in Differentiating Human Myeloid HL-60 Leukemia Cells (ORNL work)
Carcinogenesis 3, 875-880 (1982)

Horseman, N. D., and C. F. Ehret
Glucocorticosteroid Injection Is a Circadian Zeitgeber in the Laboratory Rat
Am. J. Physiol. 243 (Regulatory Integrative Comp. Physiol. 12), R373-R378 (1982)

Huberman, E., G. R. Braslawsky, M. Callaham, and H. Fugiki
Induction of Differentiation of Human Promyelocytic Leukemia (HL-60) Cells by Teleocidin and Phorbol-12-Myristate-13-Acetate (ORNL work)
Carcinogenesis 3, 111-114 (1982)

Irving, C. S., D. A. Schoeller, K.-I. Nakamura, A. L. Baker, and P. D. Klein
The Aminopyrine Breath Test as a Measure of Liver Function: A Quantitative Description of Its Metabolic Basis in Normal Subjects
J. Lab. Clin. Med. 100, 356-373 (1982)

Kubitschek, H. E., and M. J. Peak
Modified Protocol for Ames Mutagenicity Assay
Environ. Mut. 3, 693-694 (1981)

Kubitschek, H. E., and R. J. Sepanski
Azaserine: Survival and Mutation in Escherichia coli
Mutat. Res. 94, 31-38 (1982)

Larsen, R. P., R. D. Oldham, M. H. Bhattacharyya, E. S. Moretti, and D. J. Austin
Plutonium Retention in Mice and Rats after Gastrointestinal Absorption
Radiat. Res. 87, 37-49 (1981)

Leonard, A., G. Decat, and T. E. Fritz
Radiosensitivity and Life Span of Dog Peripheral Blood Lymphocytes
Mutat. Res. 92, 257-263 (1982)

Matsushita, T., A. M. Brendzel, M. A. Shotola, and K. R. Groh
Electrical Determination of Viability in Saline-Treated Mouse Myeloma Cells
Biophys. J. 39, 41-47 (1982)

Nakamura, K., D. L. Hachey, M. J. Kreek, C. S. Irving, and P. D. Klein
Quantitation of Methadone Enantiomers in Humans Using Stable Isotope-Labeled [$^2H_3$]-, [$^2H_5$]-, and [$^2H_8$]Methadone
J. Pharm. Sciences 71, 40-43 (1982)

Patel, K. R., M. M. Jonah, and Y. E. Rahman
In Vitro Uptake and Therapeutic Application of Liposome-encapsulated Methotrexate in Mouse Hepatoma 129
Eur. J. Cancer Clin. Oncol. 18, 833-843 (1982)

Peak, J. G., and M. J. Peak
Lethality in Repair-Proficient *Escherichia coli* after 365 nm Ultraviolet Light Irradiation is Dependent on Fluence Rate
Photochem. Photobiol. 36, 103-105 (1982)

Peak, J. G., M. J. Peak, and C. S. Foote
Effects of Glycerol upon the Biological Actions of Near-Ultraviolet Light: Spectra and Concentration Dependence for Transforming DNA and for *Escherichia coli* B/r
Photochem. Photobiol. 36, 413-416 (1982)

Peak, M. J., S. S. Dornfeld, and D. Venters
Liver-Microsome S9 Enzyme Increases Spontaneous Background Mutation Frequency in the Ames Salmonella Test System in the Absence of Any Added Mutagen
Mutat. Res. 103, 263-265 (1982)

Peak, M. J., and J. G. Peak
Single-Strand Breaks Induced in *Bacillus subtilis* DNA by Ultraviolet Light: Action Spectrum and Properties
Photochem. Photobiol. 35, 675-680 (1982)

Peraino, C.
Initiation and Promotion of Liver Tumorigenesis
Natl. Cancer Inst. Monogr. 58, 55-61 (1981)

Rahman, Y. E., E. A. Cerny, K. R. Patel, E. H. Lau, and B. J. Wright
Differential Uptake of Liposomes Varying in Size and Lipid Composition by Parenchymal and Kupffer Cells of Mouse Liver
Life Sci. 31, 2061-2071 (1982)

Raveh, D., T. J. Slaga, and E. Huberman
Cell-Mediated Mutagenesis and Tumor-Initiating Activity of the Ubiquitous Polycyclic Hydrocarbon, Cyclopenta[c,d]pyrene (ORNL work)
Carcinogenesis 3, 763-766 (1982)

Rosenberg, R. S., P. H. Duffy, and G. A. Sacher
Effects of Intermittent 60-Hz High Voltage Electric Fields on Metabolism, Activity, and Temperature in Mice
Bioelectromagnetics 2, 291-303 (1981)

Ryu, E. K., R. J. Ross, T. Matsushita, M. MacCoss, C. I. Hong, and C. R. West
Phospholipid-Nucleoside Conjugates. 3. Syntheses and Preliminary Biological Evaluation of 1-β-D-Arabinofuranosylcytosine 5'-Monophosphate-L-1,2-Dipalmitin and Selected 1-β-D-Arabinofuranosylcytosine 5'-Diphosphate-L-1,2-Diacylglycerols
J. Med. Chem. 25, 1322-1329 (1982)

Sacher, G. A.
Evolutionary Theory in Gerontology
Perspect. Biol. Med. 25, 339-353 (1982)

Schiffer, M., F. J. Stevens, F. A. Westholm, S. S. Kim, and R. D. Carlson
Small-Angle Neutron Scattering Study of Bence-Jones Protein Mcg: Comparison of Structures in Solution and in Crystal
Biochemistry 21, 2874-2878 (1982)

Schoeller, D. A., P. D. Klein, W. C. MacLean, Jr., J. B. Watkins, and E. Van Santen
Fecal ^{13}C Analysis for the Detection and Quantitation of Intestinal Malabsorption
J. Lab. Clin. Med. 97, 439-448 (1981)

Seed, T. M., M. Rubenstein, and K. M. Anderson
Early Effects of Castration and Replacement of Androgen on the Expression of Retrovirus-like Particles in Rat Ventral Prostate Epithelial Cells
Proc. Soc. Exp. Biol. Med. 169, 470-479 (1982)

Suzuki, F., A. Han, G. R. Lankas, H. Utsumi, and M. M. Elkind
Spectral Dependencies of Killing, Mutation, and Transformation in Mammalian Cells and Their Relevance to Hazards Caused by Solar Ultraviolet Radiation
Cancer Res. 41, 4916-4924 (1981)

Taylor, J., N. L. Anderson, A. E. Scandora, Jr., K. E. Willard, and N. G. Anderson
Design and Implementation of a Prototype Human Protein Index
Clin. Chem. 28, 861-866 (1982)

Webb, R. B., and M. S. Brown
Genetic Damage in *Escherichia coli* K12 AB2480 by Broad-Spectrum Near-Ultraviolet Radiation
Science 215, 991-993 (1982)

Webb, R. B., M. S. Brown, and R. D. Ley
Nonreciprocal Synergistic Lethal Interaction between 365-nm and 405-nm Radiation in Wild Type and uvrA Strains of Escherichia coli
Photochem. Photobiol. 35, 697-703 (1982)

Webb, R. B., and M. A. Turner
Mutation Induction by Monochromatic 254-nm and 365-nm Radiation in Strains of Escherichia coli That Differ in Repair Capability
Mutat. Res. 84, 227-237 (1981)

Webb, R. B., and R. W. Tuveson
Differential Sensitivity to Inactivation of nur and nur^+ Strains of Escherichia coli at Six Selected Wavelengths in the UVA, UVB and UVC Ranges
Photochem. Photobiol. 36, 525-530 (1982)

Willard, K. E.
Two-Dimensional Analysis of Human Lymphocyte Proteins II. Regulation of Lymphocyte Proteins by a Molecule Present in Normal Human Urine
Clin. Chem. 28, 1074-1083 (1982)

Willard, K. E.
Two-Dimensional Analysis of Human Lymphocyte Proteins: III. Preliminary Report on a Marker for the Early Detection and Diagnosis of Infectious Mononucleosis
Clin. Chem. 28, 1031-1035 (1982)

Willard, K. E., A. K. Thorsrud, E. Munthe, and E. Jellum
Two-Dimensional Electrophoretic Analysis of Human Leukocyte Proteins from Patients with Rheumatoid Arthritis
Clin. Chem. 28, 1067-1073 (1982)

Young, D. S., and N. G. Anderson
Introduction [to a special issue of Clinical Chemistry]
Clin. Chem. 28, 737-738 (1982)

BOOKS, REPORTS, AND PROCEEDINGS

Bourne, S., A. M. Jirka, and M. J. Peak
Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process): III. Mass and Mutagenicity Balances in Chemical Fractionations
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 173-180

Collins, J. J.
Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume III, SPAHR Interactive Package Guide
NUREG/CR-2364 Volume III, ANL-81-59 Volume III (1982)

Collins, J. J., and R. T. Lundy
Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume II, SPAHR Introductory Guide
NUREG/CR-2364 Volume II, ANL-81-59 Volume II (1982)

Collins, J. J., and R. T. Lundy
Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume IV, SPAHR Users Guide
NUREG/CR-2364 Volume IV, ANL-81-59 Volume IV (1982)

Collins, J. J., and R. T. Lundy
Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume V, SPAHR Programmer's Guide
NUREG/CR-2364 Volume V, ANL-81-59 Volume V (1982)

Collins, J. J., R. T. Lundy, D. Grahn, and M. E. Ginevan
Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants, Volume I, Introduction to the SPAHR Demographic Model for Health Risk
NUREG/CR-2364 Volume I, ANL-81-59 Volume I (1982)

Curtiss, J. R. B.
Computer Mapping of Birth Weights as an Approach to Environmental Epidemiology
Computer Mapping Applications in Urban, State and Federal Government: Plus Computer Graphics in Education, Vol. 16, 1981, P. A. Moore, ed., Harvard Library of Computer Graphics/1981 Mapping Collection, Laboratory for Computer Graphics and Spatial Analysis, Harvard Graduate School of Design, Cambridge, MA, pp. 77-82

Curtiss, J. R. B., M. E. Ginevan, and C. D. Brown
Spatio/Temporal Analyses of Human Birth Weight: An Indicator of Subtle Environmental Stress?
Health Impacts of Different Sources of Energy, IAEA International Symposium on Health Impacts of Different Sources of Energy, Nashville, TN, June 22-26, 1981, International Atomic Energy Agency, Vienna, 1982, pp. 33-50

Ehret, C. F., J. C. Meinert, and K. R. Groh
The Chronopharmacology of L-DOPA: Implications for Orthochronal Therapy in the Prevention of Circadian Dyschronism
Chronopharmacology and Chronotherapeutics, C. A. Walker, K. F. A. Soliman, and C. M. Winget, eds., Florida A & M University Foundation, Tallahassee, FL, 1981, pp. 47-65

Elkind, M. M., and A. Han
Neoplastic Transformation and Dose Fractionation: Does Repair of Damage Play a Role? [Summary of paper published in Radiat. Res. 79, 233-240 (1979)]
The Year Book of Cancer 1981, R. L. Clark, R. W. Cumley, and R. C. Hickey, eds., Year Book Medical Publishers, Inc., Chicago, 1981, pp. 368-370

Fritz, T. E., D. V. Tolle, D. E. Doyle, T. M. Seed, and S. M. Cullen
Hematologic Responses of Beagles Exposed Continuously to Low Doses of ^{60}Co γ-Radiation
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 28, pp. 229-240

Fry, R. J. M., R. D. Ley, D. Grube, and E. Staffeldt
Studies on the Multistage Nature of Radiation Carcinogenesis
Carcinogenesis - A Comprehensive Survey, Vol. 7, Cocarcinogenesis and Biological Effects of Tumor Promoters, E. Hecker, N. E. Fusenig, W. Kunz, F. Marks, and H. W. Thielmann, eds., Raven Press, NY, 1982, pp. 155-165

Giometti, C. S.
Muscle Protein Analysis by Two-Dimensional Gel Eletrophoresis
Critical Reviews in Clinical Laboratory Sciences, Vol. 18, Issue 1, J. Batsakis and J. Savory, eds., CRC Press, Inc., Boca Raton, FL., 1982, pp. 79-109

Haugen, D. A., M. J. Peak, and C. A. Reilly, Jr.
Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). II. Nitrous Acid Treatment for Detection of Mutagenic Primary Aromatic Amines: Nonspecific Reactions
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 115-127

Huberman, E., C. Weeks, V. Solanki, M. Callaham, and T. Slaga
Cell Differentiation, Alterations in Polyamine Levels, and Specific Binding of Phorbol Diesters in Cultured Human Cells (ORNL work)
Carcinogenesis - A Comprehensive Survey, Vol. 7, Cocarcinogenesis and Biological Effects of Tumor Promoters, E. Hecker, N. E. Fusenig, W. Kunz, F. Marks, and H. W. Thielmann, eds., Raven Press, NY, 1982, pp. 405-416

Jones, C. A., and E. Huberman
The Effect of Tumor-Causing Chemicals on the Control of Mutagenesis and Differentiation in Mammalian Cells (ORNL work)
Biological Carcinogenesis, M. A. Rich and P. Furmanski, eds., Marcel Dekker, Inc., New York and Basel, 1982, pp. 107-121

Kirchner, F. R., D. M. Buchholz, V. A. Pahnke, and C. A. Reilly, Jr.
Biological Monitoring of Fluidized Bed Coal Combustion Operations: II. Mammalian Responses following Exposure to Gaseous Effluents
Short-Term Bioassays in the Analysis of Complex Environmental Mixtures II, M. D. Walters, S. S. Sandhu, J. L. Huisingh, L. Claxton, and S. Nesnow, eds., Plenum Publishing Corporation, 1981, pp. 421-430

Kubitschek, H. E., and C. N. Newman
RecA-Mediated Asymmetric Repair of Lethal DNA Lesions in E. coli
Chromosome Damage and Repair, E. Seeberg and K. Kleppe, eds., Plenum Publishing Corporation, NY, 1981, pp. 187-191

Kubitschek, H. E., D. M. Williams, and F. R. Kirchner
Biological Monitoring of Fluidized Bed Coal Combustion Operations: I. Increased Mutagenicity during Periods of Incomplete Combustion
Short-Term Bioassays in the Analysis of Complex Environmental Mixtures II, M. D. Walters, S. S. Sandhu, J. L. Huisingh, L. Claxton, and S. Nesnow, eds., Plenum Publishing Corporation, 1981, pp. 411-420

Larsen, R. P., R. D. Oldham, M. H. Bhattacharyya, E. S. Moretti, and D. J. Austin
On the Gastrointestinal Absorption of Plutonium
Actinides in Man and Animals, M. E. Wrenn, ed., RD Press, Salt Lake City, UT, 1981, pp. 303-307

MacCoss, M., E. K. Ryu, C. I. Hong, and T. Matsushita
Synthesis and Biological Activity of Novel Nucleoside-Phospholipid Prodrugs
Nucleosides, Nucleotides and Their Biological Applications, Proceedings of the 4th International Round Table, Antwerp, February 4-6, 1981, F. C. Alderweireldt and E. L. Esmans, eds., University of Antwerp, Belgium, 1982, pp. 255-263

Oldfield, D. G.
Determining Three-Dimensional Intracellular Structure Data from Scanning Proton Microscopy Luminescence Data
Proceedings of the Fourth International Congress for Stereology, Gaithersburg, MD, September 4-9, 1975, National Bureau of Standards Special Publication 431, 1976, pp. 211-214

Peak, M. J., and H. E. Kubitschek
Survival of the Brine Shrimp, Artemia salina, after Exposure to 290-nm Ultraviolet Radiation, with and without Maximum Photoreactivation
The Role of Solar Ultraviolet Radiation in Marine Ecosystems, J. Calkins, ed., Plenum Publishing Corporation, 1982, pp. 357-362

Peak, M. J., and J. G. Peak
Lethal Effects on Biological Systems Caused by Solar Ultraviolet Light: Molecular Considerations
The Role of Solar Ultraviolet Radiation in Marine Ecosystems, J. Calkins, ed., Plenum Publishing Corporation, 1982, pp. 325-336

Rahman, Y. E.
Liposomes as Delivery Systems for Iron Chelators
Proceedings of the Second Symposium on Development of Iron Chelators for Clinical Use, San Francisco, CA, August 23-24, 1980, A. E. Martell, W. F. Anderson, and D. G. Badman, eds., Elsevier North Holland, Inc., 1981, pp. 211-225

Reilly, C. A., Jr., A. S. Boparai, S. Bourne, R. D. Flotard, D. A. Haugen, R. E. Jones, F. R. Kirchner, T. Matsushita, M. J. Peak, V. C. Stamoudis, J. R. Stetter, and K. E. Wilzbach
Health and Environmental Studies of Coal Gasification Process Streams and Effluents
Symposium Proceedings: Environmental Aspects of Fuel Conversion Technology - VI, A Symposium on Coal-Based Synfuels, Denver, CO., October 26-30, 1981, F. A. Ayer and N. S. Jones, compilers, U.S. Environmental Protection Agency, Washington, DC, EPA-600/9-82-017, 1982, 282-292

Reilly, C. A., Jr., M. J. Peak, T. Matsushita, F. R. Kirchner, and D. A. Haugen
Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). IV. Biological Activity
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 310-324

Seed, T. M., G. T. Chubb, and D. V. Tolle
Sequential Changes in Bone Marrow Architecture during Continuous Low Dose Gamma Irradiation
Scanning Electron Microscopy/1981/IV, O. Johari, R. M. Albrecht, and T. M. Seed, eds., SEM, Inc., AMF O'Hare (Chicago), IL, 1982, pp. 61-72

Stamoudis, V. C., S. Bourne, D. A. Haugen, M. J. Peak, C. A. Reilly, J. R. Stetter, and K. Wilzbach
Chemical and Biological Characterization of High-Btu Coal Gasification (The HYGAS Process). I. Chemical Characterization of Mutagenic Fractions
Coal Conversion and the Environment: Chemical, Biomedical, and Ecological Considerations, Proceedings of the 20th Annual Hanford Life Sciences Symposium, Richland, WA, October 19-23, 1980, D. D. Mahlum, R. H. Gray, and W. D. Felix, eds., Technical Information Center, U. S. DOE #CONF-801039, 1981, pp. 67-95

Stevens, F. J., and C. Peraino
Development of a Model to Simulate the Characteristics of Rat Liver Tumor Promotion by Phenobarbital
Carcinogenesis - A Comprehensive Survey, Vol. 7, Cocarcinogenesis and Biological Effects of Tumor Promoters, E. Hecker, N. E. Fusenig, W. Kunz, F. Marks, and H. W. Thielmann, eds., Raven Press, NY, 1982, pp. 105-109

Tolle, D. V., T. E. Fritz, T. M. Seed, S. M. Cullen, L. S. Lombard, and C. M. Poole
Leukemia Induction in Beagles Exposed Continuously to ^{60}Co γ-Irradiation: Hematology
Experimental Hematology Today, S. J. Baum, G. D. Ledney, and S. Thierfelder, eds., S. Karger, New York, 1982, Part VIII, Chap. 29, pp. 241-249

Wilzbach, K. E., and C. A. Reilly, Jr., compilers and eds.
Status of Health and Environmental Research Relative to Coal Gasification: 1976 to the Present
U.S. Department of Energy, Washington, DOE/ER-0149, 1982

Wilzbach, K. E., J. R. Stetter, C. A. Reilly, Jr., and W. G. Willson
Environmental Research Program for Slagging Fixed-Bed Coal Gasification
ANL/SER-1 (November 1981)

ABSTRACTS

Bhattacharyya, M. H.
Influence of Pregnancy and Lactation on Gastrointestinal Absorption of Cadmium and Lead
Teratology 26, 47A (1982)

Danon, M. J., C. S. Giometti, and N. G. Anderson
Analysis of Proteins from Human Muscle Biopsies by Two-Dimensional Electrophoresis
Neurology 32, A203 (1982)

Deeg, H. J., T. E. Fritz, R. Prentice, G. E. Sale, L. S. Lombard, E. D. Thomas, and R. Storb
Increased Cancer Risk in Canine Radiation Chimeras
Proceedings of the 13th International Cancer Congress, Seattle, WA, September 8-15, 1982, p. 212

Elkind, M. M., F. Suzuki, A. Han, G. R. Lankas, and H. Utsumi
Spectral Dependencies of Killing, Mutation, and Transformation in Mammalian Cells and Their Relevance to Hazards Caused by Solar Ultraviolet Radiation
Ninth Annual Report of Cancer Research at the University of Chicago, 1981-1982, p. 169

Epperson, J. E., and C. S. Borso
Small Angle Scattering Diffractometer Developed for Metallurgical Type Applications at the Argonne Pulsed Neutron Source, IPNS-I
Proceedings of the American Crystallographic Association Summer Meeting, LaJolla, CA, August 15-19, 1982, p. 11.

Garriott, M. L., B. H. Farrington, and D. Grahn
Investigation of Genetic and Cytogenetic Effects of Weekly Exposure to Occupational Levels of Fission Neutrons and ^{60}Co Gamma Rays
Abstracts of the Thirteenth Annual Meeting of the Environmental Mutagen Society, Boston, MA, February 26-March 1, 1982, p. 100

Giometti, C. S., and N. L. Anderson
Tropomyosin Heterogeneity in Nonmuscle Cells
J. Cell Biol. 95, 282 (1982)

Han, A., and M. M. Elkind
Enhanced Transformation of Mouse 10T1/2 Cells by 12-O-Tetradecanoyl-phorbol-13-Acetate following Exposure to X-Rays or to Fission-Spectrum Neutrons
Ninth Annual Report of Cancer Research at the University of Chicago, 1981-1982, P. 168

Hong, C. I., M. MacCoss, E. K. Ryu, and C. R. West
1-β-D-Arabinofuranosylcytosine Conjugate of Phospholipid as Potential New Antitumor Prodrug
Proceedings of the 73rd Annual Meeting of the American Association for Cancer Research, St. Louis, MO, April 28-May 1, 1982, Proceedings of the American Association for Cancer Research 23, 200 (1982)

Hong, C. I., A. Nechaev, A. J. Kirisits, M. MacCoss, E. K. Ryu, and C. R. West
Nucleoside Conjugates as Potential Antitumor Agents
5th International Round Table, Nucleosides, Nucleotides and Their Biological Applications, Research Triangle Park, NC, October 20-22, 1982, p. 46

Jasper, D. K., and T. E. Fritz
Changes in Amino Acid Uptake by the Jejunum of the $B6CF_1$ Mouse after Irradiation
J. Cell Biol. 95, 384 (1982)

Jones, R. E., F. R. Kirchner, F. J Tremmel, and C. A. Reilly, Jr.
Initial Results of the Dermal Effects of Oil from Coal Gasification
The Toxicologist 2, 138-139 (1982)

Kirchner, F. R., P. F. Dunn, V. A. Pahnke, and R. E. Jones
Characteristics of Emissions Produced by Two High-Temperature Combustion Sources
Extended Abstracts of the First Annual Conference of the American Association for Aerosol Research, Santa Monica, CA, February 17-19, 1982, pp. 26-1 to 26-2

Lagocki, P. A., R. A. Schwendener and Y. E. Rahman
Effects of Charge and Size on the Uptake of Unilamellar Liposomes by Macrophages
J. Cell Biol. 95, 414 (1982)

Rahman, Y. E., K. R. Patel, and E. A. Cerny
Enhanced Therapeutic Effect of Intraperitoneally Administered, Liposome-Encapsulated 1-β-D-Arabinofuranosylcytosine (ara-C) Against Mouse Lewis Lung Carcinoma
Proceedings of the 13th International Cancer Congress, Seattle, WA, September 8-15, 1982, p. 597

Schwartz, H. M., M. MacCoss, and S. S. Danyluk
^{17}O Chemical Shifts of Substituted Uridines and Thymidines
The Multinuclear Approach to Nuclear Magnetic Resonance, NATO Advanced Study Institute, Stirling, Scotland, August 23-September 3, 1982, p. 23

Seed, T. M., L. V. Kaspar, D. V. Tolle, and T. E. Fritz
Radiation Sensitivities of Hemopoietic Progenitors: Hemopathological Predisposition, and Survival Times under Continuous Low Daily Dose Gamma Irradiation
Exp. Hematol. 10, 174 (1982)

Short, M., F. J. Stevens, F. Westholm, A. Solomon, and M. Schiffer
Preliminary X-Ray Crystallographic Studies of Bence-Jones Protein Loc
Program Abstracts of Tenth Annual Meeting of the Midwest Autumn Immunology Conference, Minneapolis, MN, October 18-20, 1981

Short, M., F. J. Stevens, F. Westholm, A. Solomon, and M. Schiffer
Crystallographic Investigation of λ-Bence-Jones Protein LOC
Fed. Proc. 41, 287 (1982)

Tolle, D. V., T. M. Seed, T. E. Fritz, S. M. Cullen, C. M. Poole, and L. S. Lombard
Defective Megakaryocyte Differentiation in Canine Preleukemia: Cytomorphology, Ultrastructure, and Pathology
Exp. Hematol. 10, 85 (1982)

23. STAFF AND FUNDING AGENCIES*

CARCINOGENESIS SECTION

Hepatocarcinogenesis

Regular Staff

C. Peraino (Senior Biochemist)
Principal Investigator

J. A. Blomquist (Programmer)
V. A. Ludeman (Scientific Asst.)
A. M. Prapuolenis (Scientific Asst.)
J. J. Russell (Scientific Assoc.)
E. F. Staffeldt (Scientific Assoc.)

Temporary Staff

W. E. Boernke (Visiting Scientist)
J. E. Morris (Scientist in Residence)

DOE: Carcinogenesis (HA-02-02-01) ANL-60400 "Carcinogenesis-Pollutant Interactions"

Carcinogenesis (HA-02-02-01) ANL-60402 "The Detection of Tumor Initiating, Promoting, and Synergistic Activity among Environmental Pollutants"

NIEHS: Interagency Agreement, ANL-8D410 "Refinement of Newborn Rat Model as a Rapid Bioassay for Tumor Initiators and Promoters"

*Staff listings and funding agencies shown cover the period 1982 through April 1983. Participating staff from divisions other the Division of Biological and Medical Research are not included.

The individual groups are arranged in the order in which the program summaries are presented in this report. Group titles in this listing are those in the organizational chart (p. 3).

Abbreviations: DOE, Department of Energy; NCI, National Cancer Institute; NRC, Nuclear Regulatory Commission; NIADDK, National Institute of Arthritis, Diabetes, and Digestive and Kidney Diseases; NIEHS, National Institute of Environmental Health Sciences.

Gene Expression and Carcinogenesis	DOE:	Carcinogenesis (HA-02-02-01) ANL-60417 "Chemical Carcinogenesis Studies in Cultured Human and Rodent Cells"
Regular Staff		
E. Huberman (Division Director) Principal Investigator	NIEHS:	Interagency Agreement, ANL-8D408 "Transformation of Syrian Hamster Embryo Cells by Carcinogenic Chemicals"
M. F. Callaham (Scientific Assoc.) C. B. Henning (Scientific Assoc.) C. A. Jones (Asst. Biochemist) B. A. Sedita (Scientific Asst.)	NCI:	Interagency Agreement, ANL-8D409 "Malignant Cell Transformation and Mutagenesis Induced by Carcinogenic Chemicals"
Temporary Staff		
K. Kiguchi (Postdoc. Appointee) E. A. Malvoisin (Postdoc. Appointee) S.-I. Murao (Postdoc. Appointee) C. T. Oravec (Postdoc. Appointee) I. Simon (Postdoc. Appointee) M. Stodolsky (Visiting Scientist)		
Mammalian Cell Biology	DOE:	Carcinogenesis (HA-02-02-01) ANL-61302 "Mutation and Transformation: New Cell Systems for Studies of Mechanisms"
Regular Staff		
A. Han (Biophysicist) Principal Investigator	DOE:	Mutagenesis (HA-02-02-02) ANL-61300 "Mechanisms of Radiation-Induced Changes in Mammalian Cell Properties"
E. M. Buess (Scientific Asst.) J. L. Dainko (Scientific Asst.) P. J. Dale (Scientific Asst.) M. M. Elkind (Sr. Biophysicist) C.-M. Liu (Scientific Asst.)	NCI:	5 R01 CA26984-04 "UV-X-Ray Interaction: Mutation and Transformation"
Temporary Staff	NCI:	5 R01 CA29940-03 "Damage-Repair Studies Related to Mammography"
C. K. Hill (Research Associate) F. Suzuki (Postdoc. Appointee) R. L. Wells (Postdoc. Appointee)	NCI:	1 R01 CA33974-01 "Mutation-Transformation: Neutron Damage and Repair"

Metal Carcinogenesis

Regular Staff

Y. E. Rahman (Senior Biologist)
Principal Investigator

E. A. Cerny (Scientific Asst.)

Temporary Staff

P. A. Lagocki (Postdoc. Appointee)
R. A. Schwendener (Postdoc. Appointee)

DOE: Carcinogenesis (HA-02-02-01)
ANL-60414 "The Role of Metals as Cocarcinogens in the Induction of Malignant Tumors"

NIADDK: 5 R01 AM21592-06
"New Iron Chelator Delivery System for Cooley's Anemia"

LOW LEVEL RADIATION SECTION

Radiation Toxicology

Regular Staff

T. E. Fritz (Vet. Pathologist)
Principal Investigator

D. E. Doyle (Scientific Asst.)
G. L. Holmblad (Scientific Assoc.)
L. V. Kaspar (Scientific Assoc.)
W. G. Keenan (Scientific Assoc.)
L. S. Lombard (Vet. Pathologist)
C. M. Poole (Veterinarian)
A. R. Sallese (Scientific Asst.)
T. M. Seed (Biologist)
D. V. Tolle (Asst. Biologist)

DOE: Carcinogenesis (HA-02-02-01)
ANL-63100 "Radiation Toxicity in Dogs"

NCI: Interagency Agreement,
ANL-8D407
"Late Effects of Protracted Irradiation in Dogs"

Radiation Hematology

Regular Staff

T. M. Seed (Biologist)
Prinicpal Investigator

L. V. Kaspar (Scientific Assoc.)

DOE: Carcinogenesis (HA-02-02-01)
ANL-63105 "Sensitive Indicators of Altered Target Cell Function in Chronic Toxicity Studies"

Radiation Biology	DOE:	Carcinogenesis (HA-02-02-01) ANL-60300 "Neutron and Gamma-Ray Toxicity Studies"
Regular Staff		
J. F. Thomson (Senior Biologist) Principal Investigator (to 6/82)	NRC:	Div. of Health, Siting, and Waste Management "Relative Biological Effectiveness of Fission Neutrons and Gamma Rays at Occupational Exposure Levels" ANL-8M456
D. Grahn (Senior Biologist) Principal Investigator (from 6/82)		
G. L. Holmblad (Scientific Assoc.)		
J. L. Hulesch (Scientific Asst.)		
L. S. Lombard (Vet. Pathologist)		
V. A. Ludeman (Scientific Asst.)		
A. R. Sallese (Scientific Asst.)		
E. F. Staffeldt (Scientific Assoc.)		
J. E. Trier (Engineering Asst.)		
F. S. Williamson (Physicist)		
B. J. Wright (Scientific Assoc.)		

Mammalian Genetics and Statistics	DOE:	Mutagenesis (HA-02-02-02) ANL-62100 "Genetic Effects of High LET Radiations"
Regular Staff		
D. Grahn (Senior Biologist) Principal Investigator	NRC:	Div. of Health, Siting, and Waste Management "Relative Biological Effectiveness of Fission Neutrons and Gamma Rays at Occupational Exposure Levels" ANL-8M456
B. A. Carnes (Asst. Biostatistician)		
B. H. Farrington (Scientific Asst.)		
J. J. Russell (Scientific Assoc.)		
Temporary Staff		
M. L. Garriott (Postdoc. Appointee)		

Human Health Effects and Risk Assessments*	DOE:	Human Health Studies (HA-02-01) ANL-68100 "Energy Production and Human Health"
Regular Staff		
J. J. Collins (Asst. Environ. Sci.) Principal Investigator	DOE:	Health and Environmental Risk Analysis (HA-02-06) ANL-68103 "Analysis of Health and Environmental Risks of Coal Gasification"
M. E. Ginevan (Asst. Statistician) Principal Investigator		
C. D. Brown (Scientific Assoc.) B. A. Carnes (Asst. Biostatistician) (Postdoc. Appointee to 4/82) J. R. B. Curtiss (Scientific Asst.) N. Devine (Scientific Assoc.)	NRC:	"Projection Models for Health Effects Assessment in Populations Exposed to Radioactive and Nonradioactive Pollutants" ANL-8M419
	NRC:	"Reanalysis of the Tri-State Leukemia Survey Data with Special Reference to the Leukemogenic Potential of Diagnostic X-Rays" ANL-8M420

MOLECULAR BIOLOGY SECTION

Molecular Anatomy	DOE:	Systems Damage (HA-02-02-03) ANL-61203 "Molecular Perturbations in Man Produced by Energy Related Pollutants"
Regular Staff		
N. G. Anderson (Sr. Physiologist) Principal Investigator	DOE:	Human Health Effects from Energy Generation (HA-02-01-01) ANL-68112 "Development of New Technology for the Use of Isotopic Tracers in the Study of Human Health and Disease"
N. L. Anderson (Biophysicist) J. J. Edwards (Asst. Biologist) M. A. Gemmell (Scientific Asst.) C. S. Giometti (Asst. Biologist) S. L. Nance (Scientific Assoc.) J. Taylor (Computer Scientist) S. L. Tollaksen (Scientific Assoc.)		
Temporary Staff		
K. W. Willard (Postdoc. Appointee)		

*All activities in this program were completed September 30, 1982.

Molecular Biophysics	DOE:	General Life Sciences (HB-01) ANL-61200 "Detection and Characterization of Damage in Molecular, Cellular, and Physiological Systems"

Regular Staff

M. Schiffer (Biophysicist)
Principal Investigator

C. F. Ainsworth (Scientific Asst.)
C. S. Borso (Asst. Biophysicist)
M. MacCoss (Biochemist)
F. A. Westholm (Scientific Asst.)

Temporary Staff

C.-H. Chang (Postdoc. Appointee)
C. W. Hanly (Visiting Scientist)
M. T. Short (Postdoc. Appointee)
P. Thiyagarajan (Postdoc. Appointee)

Mutagenesis	DOE:	General Life Sciences (HB-01) ANL-62201 "Molecular, Genetic, and Cellular Mechanisms of Environmental and Solar-UV Mutagens"

Regular Staff

H. E. Kubitschek (Sr. Biophysicist)
Principal Investigator

D. Williams-Hill (Scientific Asst.)

Temporary Staff

W. W. Baldwin (Scientist in Residence)
J. G. Peak (Postdoc. Appointee)*
P. R. Reynolds (Postdoc. Appointee)

Photobiology	DOE:	General Life Sciences (HB-01) ANL-62201 "Molecular, Genetic, and Cellular Mechanisms of Environmental and Solar-UV Mutagens"

Regular Staff

M. J. Peak (Microbiologist)
Principal Investigator

Temporary Staff

R. W. Tuveson (Scientist in Residence)

*Assistant Biologist, effective February 1, 1983.

TOXICOLOGY SECTION

Chemical Toxicology

Regular Staff

C. A. Reilly, Jr. (Microbiologist)
Principal Investigator

S. S. Dornfeld (Scientific Asst.)
D. A. Haugen (Biochemist)
F. R. Kirchner (Asst. Biologist)
T. Matsushita (Geneticist)
V. A. Pahnke (Scientific Asst.)
M. J. Peak (Microbiologist)
M. A. Shotola (Scientific Asst.)
K. M. Suhrbier (Scientific Asst.)
D. Venters (Scientific Asst.)

Temporary Staff

M. L. Cunningham (Postdoc. Appointee)
R. E. Jones (Postdoc. Appointee)
J. M. Leonardo (Postdoc. Appointee)

DOE: Carcinogenesis (HA-02-02-01) ANL-60404 "Toxicological Evaluation of Coal Gasification Technology"

Carcinogenesis (HA-02-02-01) ANL-62200 "Genetic Toxicology of Coal Gasification"

DOE: Coal: Surface Coal Gasification (AA-85-25-05) ANL-49533 "Process Development Unit Coal Gasification"

Chemical and Biological Characterization

Regular Staff

D. A. Haugen (Biochemist)
Principal Investigator

K. M. Suhrbier (Scientific Asst.)

DOE: Analytical Studies (HA-02-04-01) ANL-60407 "Fractionation/Characterization of Coal Gasification Process Streams for Biological Evaluation"

Metal Toxicology Regular Staff M. H. Bhattacharyya (Biochemist) Principal Investigator E. S. Moretti (Scientific Asst.) D. P. Peterson (Scientific Asst.) Temporary Staff B. D. Whelton (Scientist in Residence)	DOE: DOE: NRC:	Systems Damage (HA-02-02-03) ANL-61209 "Biological Effects of Metals in Developing Energy Technologies" Health and Environmental Risk Analysis (HA-02-06) ANL-61210 "Assessment of the Health and Environmental Effects of Battery Energy Storage Systems" Div. of Health, Siting, and Waste Management "Reanalysis of Gastrointestinal Absorption Factors for Plutonium and Other Actinide Elements" ANL-8M447
Neurobehavioral Chronobiology Regular Staff C. F. Ehret (Sr. Biologist) Principal Investigator P. H. Duffy (Scientific Asst.) K. R. Groh (Scientific Assoc.) J. J. Russell (Scientific Assoc.)	DOE: DOE:	Systems Damage (HA-02-02-03) ANL-61208 "Neurobehavioral Chronobiology and Toxicology" Electric Energy Systems (AK-05-02) ANL-49185 "Biomedical Effects Associated with Energy Transmission Systems"

AUTHOR INDEX

Ainsworth, C. F., 57
Angerman, J. M., 27
Anderson, N. G., 49
Anderson, N. L., 49

Baldwin, W. W., 63
Benioff, P. A., 87
Benioff, R. D., 57
Bhattacharyya, M. H., 87
Blomquist, J. A., 7
Boernke, W. E., 7
Boparai, A. S., 81
Borso, C. S., 57
Bressette, R. M., 75
Buess, E. M., 17

Callaham, M. F., 11
Carnes, B. A., 7, 41
Cerny, E. A., 21
Chang, C.-H., 57
Chivers, M., 33
Cullen, S. M., 27

Dainko, J. L., 17
Dale, P. J., 17
Dornfeld, S. S., 75, 81
Doyle, D. E., 27

Edwards, J. J., 49
Ehret, C. F., 91
Elkind, M. M., 17
Eychner, D. M., 41

Farrington, B. H., 41
Fink, M. A., 75
Fritz, T. E., 27

Garriott, M. L., 41
Gemmell, M. A., 49
Giometti, C. S., 49
Graetzer, R., 63
Grahn, D., 37, 41
Groh, K. R., 91
Guzik, J. A., 57

Hagan, M. P., 17
Han, A., 17
Haugen, D. A., 81
Henning, C. B., 11
Hill, C. K., 17
Holmblad, G. L., 27, 37
Huberman, E., 1, 11
Hulesch, J. L., 37
Hyslop, R. M., 57

Jones, C. A., 11
Jones, R. E., 75

Kaspar, L. V., 27, 33
Keenan, W. G., 27
Kerwin, S. M., 75
Kirchner, F. R., 75
Kubitschek, H. E., 63

Lagocki, P. A., 21
Larsen, R. P., 87
Lee, C.-H., 41
Leonardo, J. M., 75
Liu, C.-M., 17
Lombard, L. S., 27, 37
Ludeman, V. A., 7, 37

Malvoisin, E. A., 11
Moehring, P. M., 67
Moretti, E. S., 87
Morris, J. E., 7
Murao, S.-I., 11
Muraski, M. F., 75

Nance, S. L., 49

Oldham, R. D., 87

Pahnke, V. A., 75
Peak, J. G., 67
Peak, M. J., 67, 75, 81
Peraino, C., 7
Peterson, D. P., 87
Poole, C. M., 27
Prapuolenis, A. M., 7

Rahman, Y. E., 21
Reuman, D. L., 17
Reilly, C. A., Jr., 75, 81
Rogers, K. M., 21
Russell, J. J., 41
Ryffel, B., 11

Sallese, A. R., 27, 37
Sammartano, L. J., 67

Schiffer, M., 57
Schwendener, R. A., 21
Sedita, B. A., 11
Seed, T. M., 27, 33
Shackelford, M. E., 87
Short, M. T., 57
Shotola, M. A., 75
Simon, I., 11
Smith, V. N., 17
Staffeldt, E. F., 7, 37
Stamoudis, V. C., 81
Stodolsky, M., 11
Suhrbier, K. M., 81

Taylor, J., 49
Thiyagarajan, P., 57
Thomson, J. F., 37
Tollaksen, S. L., 49
Tolle, D. V., 27
Tompkins, C. M., 75
Tremmel, F. J. 81
Trier, J. E., 37
Turner, M. A., 67
Tuveson, R. W., 67

Venters, D., 75, 81

Wechsler, R. J., 75
Wells, R. L., 17
Westholm, F. A., 57
Whelton, B. D., 87
Willard-Gallo, K. W., 49
Williams-Hill, D., 63
Williamson, F. S., 37
Wilzbach, K. E., 75, 81
Wright, B. J., 37

Distribution for ANL-83-40

Internal:

W. E. Massey
E. S. Beckjord
J. Rundo (3)
L. Burris
H. W. Conner
P. F. Gustafson
W. J. Hallett
D. P. O'Neil
A. F. Stehney
C. F. Ainsworth
N. G. Anderson
N. L. Anderson
J. M. Angerman
W. Baldwin
M. H. Bhattacharyya
J. A. Blomquist
W. E. Boernke
C. S. Borso
E. M. Buess
M. F. Callaham
B. A. Carnes
E. A. Cerny
C.-H. Chang
S. M. Cullen
M. L. Cunningham
P. J. Dale
J. A. DePadro
T. J. Doody
S. S. Dornfeld
D. E. Doyle
C. F. Ehret
W. J. Eisler
B. F. Farrington
C. A. Fox
T. E. Fritz
M. A. Gemmell
C. S. Giometti
D. Grahn
K. R. Groh
D. D. Grube
A. Han
C. W. Hanly
D. A. Haugen
C. B. Henning
C. K. Hill
G. L. Holmblad
E. Huberman
J. S. Hulesch
E. W. Jackson
C. A. Jones
L. A. Kaspar
W. G. Keenan
K. Kiguchi
F. R. Kirchner
H. E. Kubitschek
P. A. Lagocki
D. A. LeBuis
C.-M. Liu
L. S. Lombard
V. A. Ludeman
E. A. Malvoisin
M. J. McCoy
P. M. Moehring
E. S. Moretti
J. E. Morris
S.-I. Murao
S. L. Nance
C. T. Oravec
V. A. Pahnke
J. G. Peak
M. J. Peak
C. Peraino
D. P. Peterson
C. M. Poole
A. M. Prapuolenis
L. M. Purdy
Y. E. Rahman
C. A. Reilly, Jr.
K. M. Rogers
M. W. Rosenthal
J. J. Russell
A. R. Sallese
M. Schiffer
B. A. Sedita
T. M. Seed
M. E. Shackelford
M. J. Short
E. F. Staffeldt
M. Stodolsky
K. M. Suhrbier
J. Taylor
P. Thiyagarajan
S. L. Tollaksen
D. V. Tolle
F. J. Tremmel
M. A. Turner
T. A. Vanisko
R. L. Wells
F. A. Westholm
B. D. Whelton
D. M. Williams-Hill
F. S. Williamson
B. J. Wright
G. C. Zolkiewicz
P. A. Benioff
A. S. Boparai
P. Failla
R. H. Huebner
R. P. Larsen
F. Marchetti
R. D. Oldham
R. A. Schlenker
I. Simon
V. C. Stamoudis
K. E. Wilzbach
BIM Division Office (116)
A. B. Krisciunas (20)
ANL Patent Dept.
ANL Contract Copy
ANL Libraries (2)
TIS Files (6)

External:

DOE-TIC, for distribution per UC-48 (156)
Manager, Chicago Operations Office, DOE
U. S. Department of Energy, Washington:
N. F. Barr, Office of Health and Environmental Research
G. D. Duda, Office of Health and Environmental Research
A. P. Duhamel, Office of Health and Environmental Research
C. W. Edington, Office of Health and Environmental Research

M. L. Minthorn, Jr., Office of Health and Environmental Research
D. A. Smith, Office of Health and Environmental Research
G. Stapleton, Office of Health and Environmental Research
J. W. Thiessen, Office of Health and Environmental Research
A. W. Trivelpiece, Office of Energy Research
R. W. Wood, Office of Health and Environmental Research
U. S. Nuclear Regulatory Commission, Washington:
F. J. Arsenault, Office of Nuclear Regulatory Research
J. D. Foulke, Office of Nuclear Regulatory Research
W. A. Mills, Office of Nuclear Regulatory Research
National Cancer Institute:
D. G. Longfellow, Div. of Cancer Cause and Prevention
J. L. Murray, Low Level Radiation Effects Branch
O. F. Nygaard, Low Level Radiation Effects Branch
B. Wachholz, Low Level Radiation Effects Branch
Division of Biological and Medical Research Review Committee:
T. W. Clarkson, U. Rochester
L. Grossman, Johns Hopkins U.
J. W. Osborne, U. Iowa
H. C. Pitot III, U. Wisconsin-Madison
M. Pollard, U. Notre Dame
F. M. Richards, Yale U.
L. R. Almodovar, U. Puerto Rico, Mayaguez
J. W. Baum, Brookhaven National Lab.
V. P. Bond, Brookhaven National Lab.
Brookhaven National Lab., Chairman, Biology Dept.
R. A. Builmette, Lovelace Biomedical and Environmental Research Inst., Albuquerque
L. Bustad, Washington State U.
California, U. of, Library, Davis
N. Cohen, New York U.
Colorado State U. Libraries
Cornell U., New York State Agricultural Experiment Station, Geneva
E. Flaumenhaft, U. Akron
L. S. Gomez, Sandia National Lab., Albuquerque
M. L. Griem, U. Chicago Hospitals and Clinics
Indiana U., School of Medicine Library
Inhalation Toxicology Research Inst., Director, Albuquerque
F. T Kuchnir, Elmhurst, Ill.
Lawrence Berkeley National Lab., Assoc. Dir. for Biology and Medicine Div.
Lawrence Livermore National Lab., Assoc. Dir. for Biomedical and Environmental Sciences Divs.
H. Lisco, Harvard Medical School
Los Alamos National Lab., Director, Div. of Health Research
Mann (Albert R.) Library, Ithaca
M. L. Mendelsohn, Lawrence Livermore National Lab.
Michigan Technological U., Library
Minnesota, U. of, St. Paul Campus Library
New York, State U. of, at Buffalo, Health Sciences Library
W. R. Ney, Nat. Council on Radiation Protection and Measurements, Washington
North Dakota, U. of, Energy Technology Center Library
Oak Ridge National Lab., Director, Biology Div.
A. L. Orvis, Mayo Clinic
Pacific Northwest Lab., Program Mgr., Biomedical Sciences

Pacific Northwest Lab., Mgr., Environment, Health, and Safety Research
Philadelphia, College of Physicians of, Library
E. L. Powers, U. Texas, Austin
B. R. Scott, Inhalation Toxicology Research Inst., Albuquerque
J. M. Smith, U. Utah
St. Louis U., Medical Center Library
R. L. Straube, National Institutes of Health
J. C. Thompson, Cornell U.
R. C. Thompson, Battelle Pacific Northwest Lab.
U. S. Air Force, Brooks AFB, Tex.
A. C. Upton, New York U. Medical Center
Utah, U. of, Director, Radiobiology Div.
H. H. Vogel, Jr., U. Tennessee Center for the Health Sciences
J. Wallach, Howard Beach, N.Y.
Wisconsin, Medical College of, Todd Wehr Lib., Milwaukee
Wisconsin, U. of, Golda Meir Library, Milwaukee
J. Zimbrick, U. Kansas
Comision Nacional de Energia Atomica, Library, Buenos Aires, Argentina
Cancer Institute/Peter MacCallum Hosp., Librarian, Melbourne, Australia
CSIRO, Central Library, East Melbourne, Australia
Institute of Medical and Veterinary Science, Librarian, Adelaide, Australia
J. Myhill, Hughes, Australia
J. M. Debois, St. Norbertus Hosp., Duffel, Belgium
E. DiFerrante, Commission of the European Communities, Brussels, Belgium
A. Heyndrickx, U. of Ghent, Belgium
S. Menezes, Ilha Universitaria, Rio de Janeiro, Brazil
C. Pavan, UNICAMP, Sao Paulo, Brazil
Defence Research Establishment Library, Ottawa, Canada
Guelph, U. of, Library, Guelph, Ontario, Canada
E. G. LeTourneau, National Radiation Protection Bu., Health and Welfare, Ottawa, Canada
Toronto, U. of, Library, Canada
G. Tratt, U. of Alberta, Edmonton, Canada
C. H. Cheng, National Tsing Hua U., Taiwan, Republic of China
Risø National Laboratory, Library, Roskilde, Denmark
P. R. J. Burch, U. of Leeds, England
Y. Cocking, MRC Radiobiology Unit, Harwell, England
W. G. Cunliffe, British Nuclear Fuels Ltd., Salwick, England
J. F. Fowler, Mt. Vernon Hospital, Northwood, England
S. J. Harris, U. Surrey, England
M. T. Herbert, National Radiological Protection Board, Chilton, England
C. R. Hill, Inst. of Cancer Research, Belmont, England
Institute of Cancer Research Library, Belmont, England
D. F. Jackson, U. Surrey, Guildford, England
L. G. Lajtha, Christie Hosp. and Holt Radium Inst., Manchester, England
P. Lindop, Medical Coll. of St. Bartholomew's Hosp., London, England
W. V. Mayneord, Surrey, England
J. S. Mitchell, Addenbrookes Hospital, Cambridge, England
E. Pochin, National Radiological Protection Board, Chilton, England
F. W. Spiers, U. Leeds, England
J. Vaughan, Oxford, England
J. Vennart, MRC Radiobiology Unit, Harwell, England
J. Coursaget, C.E.A., Saclay, France

P. J. P. Fallot, CEN Saclay, Gif-sur-Yvette, France
International Agency for Research on Cancer, Lyon, France
H. Jammet, CEA, Fontenay-aux-Roses, France
H. Franke, Radiologische Univ.-Klinik, Hamburg, Germany
U. Hagen, Gesellschaft für Strahlen- und Umweltforschung, Neuherberg, Germany
A. Kaul, Institut für Strahlenhygiene, Neuherberg, Germany
L. Kemmerich, Kernforschungszentrum Karlsruhe GmbH, Germany
H. Muth, Boris Rajewsky-Inst., U. Saarlandes, Homburg, Germany
M. F. Rajewsky, Universität Essen, Germany
E. L. Sattler, Strahlenzentrum, Giessen, Germany
S. K. Havanur, Tata Inst. of Fundamental Research Library, Bombay, India
Hebrew University, Librarian, Dept. of Botany, Jerusalem, Israel
T. Schlesinger, Soreq Nuclear Research Center, Yavne, Israel
A. Benco, C.C.R. Euratom, Ispra, Italy
G. F. Clemente, CNEN, CSN Casaccia, Rome, Italy
Istituto Superiore di Sanita, Rome, Italy
P. Metalli, CNEN, CSN Casaccia, Rome, Italy
O. Rimondi, Instituto de Fisica, Ferrara, Italy
F. Salvatore, U. Naples, Italy
L. Tommasino, CNEN, CSN Casaccia, Rome, Italy
Institute of Radiation Breeding, Library, Ibaraki, Japan
O. Matsuoka, Nat. Inst. of Radiological Sciences, Anagawa, Japan
National Inst. of Radiological Science, Planning and Information Sec., Chiba-shi, Japan
T. Shegetoshi, Power Reactor Nuclear Fuel Development Corp., Tokai, Japan
Y. Ujeno, Kyoto U., Japan
Korea Advanced Energy Research Inst., Technical Information Div., Seoul, Korea
S. S. Lee, Korea Advanced Institute of Science, Seoul, Korea
M. Bogaardt, Ultra-Centrifuge Nederland, Almelo, The Netherlands
Medical Biological Lab TNO, Director, Rijswijk, The Netherlands
O. Vos, Erasmus U., Rotterdam, The Netherlands
Estacao Agronomica Nacional, Bilioteca, Oeiras, Portugal
T. Caspersson, Karolinska Hospital, Stockholm, Sweden
B. Persson, Lunds Universitet, Sweden
A. Donath, Service cantonal de Controle des Irradiations, Geneva, Switzerland
A. Gunther, CERN Scientific Information Service, Geneva, Switzerland
N. T. Racoveanu, World Health Organization, Geneva, Switzerland
Swiss Inst. for Nuclear Research, The Library, Villigen, Switzerland
O. E. Chaves C, Universidade de Los Andes, Merida, Venezuela
D. Petrovic, Institute "Rudjer Boskovic," Zagreb, Yugoslavia

R. Benioff, Downers Grove, Ill.
R. Bressette, Attleboro, Mass.
M. Chivers, Challis, Ida.
J. L. Dainko, Joliet, Ill.
J. J. Edwards, U. North Carolina Medical School
M. M. Elkind, Colorado State U.
D. Eychner, Cortland, N.Y.
M. A. Fink, Chicago, Ill.
M. L. Garriott, Searle Research and Development Div., G. D. Searle & Co., Chicago
R. Graetzer, Pennsylvania State U.
J. Guzik, Palos Hills, Ill.

M. P. Hagan, Armed Forces Radiobiology Research Inst., Bethesda
R. Haselkorn, U. Chicago
R. M. Hyslop, Illinois Benedictine College
R. E. Jones, Diamond Shamrock Corp., Painesville, O.
S. Kerwin, Broomall, Pa.
D. W. King, U. Chicago
C. H. Lee, Swiftwater, Pa.
J. M. Leonardo, U. Kentucky
L. S. Myers, Armed Forces Radiobiology Research Inst., Bethesda
M. Muraski, Villa Park, Ill.
F. W. Putnam, Indiana U.
D. L. Redman, Cincinnati
L. Sammartano, Hoyt Lakes, Minn.
M. A. Shotola, Villa Park, Ill.
V. Smith, Laurens, N. Y.
J. F. Thomson, Scales Mound, Ill.
C. M. Tompkins, Atlanta
J. E. Trier, Chicago
R. W. Tuveson, U. Illinois, Urbana
D. Venters, Downers Grove, Ill.
R. B. Webb, Hinsdale, Ill.
R. Wechsler, Los Alamos
K. E. Willard-Gallo, Ludwig Institute for Cancer Research, Brussels, Belgium
B. Ryffel, Sandoz Ltd., Basle, Switzerland
R. A. Schwendener, Pharmaceutical Institute ETH, Zurich, Switzerland