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UNIVERSITY OF CALIFORNIA

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HEALTH DIVISION
1954

Wright H. Langham, Group Leader
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HEALTH AND SAFETY

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1. General Comments on Group Activities

This report covers the activities of the Biomedical Research Group (H-4) of the Health Division during the period January 1 through December 31, 1954.

Organizationally, Group H-4 is divided into five sections, namely, Biochemistry, Radiobiology, Radiopathology, Biophysics, and Organic Chemistry. The activities of the Group are summarized under the headings of the various sections. The general nature of each section's program, publications, documents and reports originating from its members, and abstracts and summaries of the projects pursued during the year are presented.

In the last chapter of the report, the proposed research program for the Group during 1955 is presented under the various section headings, insofar as activities for the forthcoming year can be anticipated.

1.1 Principal Group Activities During the Year

Occasionally it is necessary for the members of the various sections to participate in unusual projects which may be looked upon as major or outstanding accomplishments or as concerted group efforts. An attempt is made in this report to list these endeavors under the general comments on the Group's activities as a whole.

1.1.1 Determination of Internal Exposure of Rongelap
Natives Exposed to Radioactive Fall-Out March 1,
1954

Various members of the Radiobiology and Biochemistry Sections collaborated in an attempt to determine fission product exposure of Rongelap natives as a result of the radioactive fall-out of March 1, 1954. Individual and pooled urine samples were obtained from the exposed natives and flown back to Los Alamos. These samples were analyzed for Sr^{89-90} , I^{131} , Ba^{140} , Ru^{103} , Ca^{45} , Ce^{141} , and fissionable material. Gross and detailed gamma and beta assays of the various urines were carried out. It was necessary to modify or develop methods of separation and isolation of the various nuclides under pressure of time, because of the short half-lives of several of the fission products. Most of the beta and gamma activity was accounted for by I^{131} and/or Sr^{89} . Both of these were determined with fair accuracy by extrapolation of the decay curves of whole dried urine samples and in the case of Sr^{89} , of dry-ashed residue of 100-ml. urine aliquots. Small but measurable amounts of Ca^{45} , Ba^{140} , and Ru^{103} activities were found. No fissionable material or Ce activities were detected. Estimates of the body burden and the initial absorbed doses of these various isotopes indicated that only the isotopes of iodine greatly exceeded the acceptable

tolerance values, and the results indicated that total internal contamination was of negligible importance in comparison with the external gamma radiation dose received by the natives as a result of fall-out.

1.1.2 Treatment of a Case of Pu Exposure with Ca EDTA

During the year a case of exposure of a laboratory worker to Pu via a contaminated cut occurred. It was estimated that the employee accidentally received approximately one-half of a maximum permissible amount of Pu. Administration of Ca EDTA on the sixth day increased the rate of Pu excretion 100-fold. A 16-day course of treatment of this individual resulted in the excretion of approximately 25 per cent of her total body burden of plutonium.

1.1.3 Further Studies of the Biological Effects of
Neutrons using Godiva

Studies of the biological effects of fission neutrons using the Godiva critical assembly were continued. Several types of studies were conducted, among which were the following:

(a) The Effect of Massive Rapid Doses of Fission Neutrons on the Behavior of Monkeys. Three monkeys were exposed to high doses of fission neutrons and tested immediately for their ability to perform. Two animals received approximately 2500 rep, one in a 45- μ sec. burst and the other over

a period of approximately 2-1/2 minutes. The third animal received approximately 5,000 rep in a single 45 μ sec. exposure. The animals were tested for their ability to do physical work under the stimulus of electrical shock. These results were distributed in Report LA-1838, "Exposure of Monkeys to High Doses of Fission Neutrons from Godiva," (classified).

(b) The Median Survival Time of Mice following Massive Rapid Doses of Fission Neutrons. By use of the Godiva assembly, the effectiveness of fission neutrons over the dose range of 500 to 30,000 rep was studied. The median survival time of mice was used as the biological indicator. Animals were exposed by operating Godiva as a prompt critical assembly giving 45- μ sec. bursts of radiation and as a reactor operating at a constant power. The respective dose rates under the two different conditions of operation were 2×10^8 and 1.2×10^3 rep/min. At the two dose rates, the relative biological effectiveness of fission neutrons appeared to be approximately 2.

(c) Relative Biological Effectiveness of Fission Neutrons using Lethality and Splenic and Thymic Weight Decrease in Mice. The relative biological effectiveness of the fission neutrons from Godiva was determined using 30-day lethality and splenic and thymic weight decrease as the

biological indicators. When Godiva was operated as a prompt critical assembly, the relative biological effectiveness of the fission neutrons by these two biological methods was found to be approximately 2.

Dosimetry studies with the Godiva assembly using film packs and chloroform chemical dosimeters indicated that the gamma contamination in the radiations from the assembly constituted approximately 5 per cent of the total radiation dose. The physical dose of fission neutrons was carefully measured and calibrated against temperature rise of the assembly and the number of fissions as indicated by sulphur activation analysis. The neutron dose in rep from the assembly was measured by G. S. Hurst of Oak Ridge, using the Hurst Proportional Neutron Detector and the fission and threshold detector systems.

1.1.4 Development of Organic Solution Scintillators

The organic solution scintillator program was continued during 1954 and one of the outstanding accomplishments of this program was the development of a liquid organic scintillator which surpassed the plastic scintillators in light output per equivalent beta excitation. This achievement lies not only in the production of a better scintillation system, but also in the increased ability to measure the light output. A careful relative pulse height study of the best

organic scintillators, crystal, liquid and plastic, has given preeminence to the following three systems:

- (a) Anthracene crystal, relative rating 100;
- (b) 10 g./l. 2-phenyl-5-(4-biphenyl)-1,3,4-oxadiazole in p-xylene, relative rating 70; and
- (c) 36 g./l. p-terphenyl and 0.9 g./l. p,p'-diphenylstilbene in polyvinyl-toluene, relative rating 52.

1.1.5 Design of Large Volume Liquid Scintillator Human Counter

One of the principal accomplishments of the Biophysics Section was the completion of the design of the whole body human counter, designed to employ the principle of organic solution scintillators. A special budget of approximately \$45,000 was obtained for the construction and installation of the human counter and the design was turned over to the Shops Department for construction. As of December 31, fabrication of the major portion of the counter was complete, but installation, mechanical testing, and assembly were not begun. Modifications of the sub-basement of HRL Building for an installation site for the human counter were specified and essentially completed by the end of the year.

A special highly reflecting toluene-resistant paint for coating the inside of the counter tank was developed. The base of the paint was Epon resin 1001 polymerized with diethylene-triamine and pigmented with Glidden's Zopaque anatase TiO_2 .

1.1.6 The Pathology of Monkeys Exposed to Massive Doses of Total Body Gamma Radiation

Although this study promised on paper to be a recapitulation of the well-known morphologic effects of gamma radiation, the fortuitous sequential occurrence of the deaths of the animals enabled the piecing together of the steps in the development of the various well-known lesions and led to the conclusion that several of the commonly accepted concepts of pathology are erroneous. As examples, loss of intestinal epithelium was found to be secondary to mitotic arrest in the crypts, rather than to so-called "radio-necrosis"; time of destruction of lymphocytes was found to be more dependent upon their organic location than upon amounts of radiation; and so-called radiation-resistant organs, such as the pancreas, adrenal, thyroid, and pituitary glands, undergo rapidly progressing and recovering changes once their high thresholds are overcome.

1.2 Changes in Physical Facilities and Capital Equipment

1.2.1 Construction of Monkey Housing Facility

During 1954 there were very few changes in the physical facilities occupied by Group H-4. The principal addition was the conversion of one of the animal rooms to a small but highly efficient monkey housing facility. This housing facility consists of a long narrow corridor constructed of cyclone fencing against the side of which are fastened 12 living cages for monkeys. The cages are approximately 30 x 30 x 30 inches and are held to the side of the corridor by special support clamps. The cages and the corridor are equipped with separate doors to permit the forms to be used in other locations. Three to four rhesus monkeys weighing less than 10 pounds each may be housed per cage, which provides for a monkey colony of from 36 to 48 animals. Total cost of design and construction of this facility was approximately \$4,000.

1.2.2 Capital Equipment

During the year, a Hilger Intermediate Quartz Spectrograph was installed. This is being used in the organic scintillator program to determine the emission spectra of organic scintillators when excited with a 100-mc. Cs¹³⁷ source.

Two additional liquid scintillation beta counters

with coincidence and discriminator circuitry, etc., were obtained from P-Division. Other electronic circuits and numerous pieces of smaller equipment were also obtained during the year.

1.3 Changes in Organization and Personnel

During 1954 there were several personnel and organizational changes in Group H-4. John Storer was still on military leave, and P. S. Harris, who was a military assignee at Los Alamos during 1953, resigned from the Army and became a permanent Staff Member in the Group. Joseph A. Sayeg from the University of California at Berkeley accepted a Staff Member position in the Biophysics Section in the early Fall of 1954.

Donald Ott was hired as a Staff Member in the Organic Chemistry Section early in 1954, and Vernon Kerr was hired as a Research Assistant in the Organic Chemistry Section in the Fall.

John Furchner, who had been on leave of absence to complete his Doctor's degree at the University of New Mexico, returned as a permanent Staff Member of the Group. Camille Finnegan and Edith Lilly accepted Research Assistant and Technician positions, respectively, with the Biochemistry Section.

F. N. Hayes was transferred back to the Organic Chemistry

Section and Betty Rogers transferred from the Organic Chemistry Section to the Radiobiology Section.

During 1954, the animal caretakers, who in the past were on the payroll of the Zia Company and supplied to H-4 on a fee basis, were taken over by Group H-4 and placed on the University of California personnel roll. The transfer of the animal caretakers to the Group has resulted in some improvement and efficiency of operation of the experimental animal facility.

A. R. Ronzio transferred from the Organic Chemistry Section to Group P-8. Kenneth Kohr transferred from the Organic Chemistry Section to Group J-12.

Virgil Koenig terminated his employment with the Biochemistry Section and the Los Alamos Scientific Laboratory.

Patricia Keegan terminated her employment with the Biochemistry Section in January of 1954.

Norma R. Mecaskey, Group Secretary, terminated in July, and was replaced by Bertha Grant.

Donna Faye Turney terminated her employment in the Radiobiology Section and Rita Smith terminated her employment with the Radiopathology Section.

On December 31, 1954, Group H-4 consisted of 45 persons, 21 of whom were Staff Members, 6 of whom were Research Assistants, 15 were SCP's, and 3 were rated as ASC's. Two of the Staff Members were military assignees. The organizational chart of Group H-4 at the end of the year was as follows.

GROUP H-4
BIOMEDICAL RESEARCH

Wright H. Langham, Ph.D., Group Leader
John B. Storer, M.D., Alt. Group Leader (on Military leave)
Ogden S. Johnson, B.S., Asst. Group Leader for Administration

SWITCHBOARD AND CLERICAL

Jeanne Nordberg
Helen Furchner

SECRETARY

Bertha Grant

DISHWASHER-TECHNICIAN

Catherine Aguilar

ANIMAL CARETAKERS

Adiopolito Trujillo
Jose M. Alire
Juan Lovato
Roman Martinez
Leopoldo Ortiz

BIOCHEMISTRY

R. Gordon Gould, Ph.D.,
Leader

Staff Members

Harry Foreman, Ph.D.,
M.D.
Jean C. Sabine, M.D.
Research Assistants
Ted Trujillo, B.S.
Camille Finnegan, B.A.
Technical Staff
Marguerite Magee
Virginia Lotz, B.A.
Helen Miller, M.A.
Edith Lilly, B.S.

RADIOBIOLOGY

Payne S. Harris, M.D.,
Leader

Staff Members

*Maj. S. Rothermel (MC)
Irene U. Boone, M.D.
*Maj. Kent Woodward (MC)
John Furchner, Ph.D.
Technical Staff
Verda Strang
Phyllis Sanders, M.S.
William Schweitzer
Betty Rogers, B.S.

RADIOPATHOLOGY

C. C. Lushbaugh, M.D.,
Ph.D., Leader

Staff Members

John Spalding, Ph.D.
Research Assistants
Julie Wellnitz, A.B.
Dorothy B. Hale, B.S.
Lora B. Hughes, A.B.

BIOPHYSICS

E. C. Anderson, Ph.D.,
Leader

Staff Members

Fred Worman, M.S.
John Larkins, B.S.
James D. Perrings
Joseph Sayeg, Ph.D.
Technical Staff
Louise Larkins, B.S.

ORGANIC CHEMISTRY

W. H. Langham, Ph.D.,
Leader

Staff Members

Arthur Murray, M.S.
Lloyd Williams, M.S.
Donald G. Ott, Ph.D.
F. N. Hayes, Ph.D.
Research Assistant
Vernon N. Kerr, M.A.

SM - 21
RA - 6
SCP - 15
ASC - 3
45

*Military

1.4 Summer Employment Program

During the summer of 1954, Group H-4 had the largest and most successful summer employment program in the Group's history. Two consultants were also in residence at Los Alamos during the summer. They were T. M. Eversole of the Department of Biology, University of New Mexico; and W. D. Armstrong, Department of Biochemistry, School of Medicine, University of Minnesota.

The summer employment program is summarized as follows:

Helen Davison, Charlottesville, Virginia; assigned to the Radiobiology Section as a Technician.

Charles Goodner, University of Utah, School of Medicine, Salt Lake City; assigned to the Radiobiology Section as a Research Assistant.

Arthur J. Hodges, University of Colorado, School of Medicine, Denver; assigned to the Biophysics Section as a Staff Member.

Thomas E. Moore, Jr., Harvard Medical School, Boston, Massachusetts; assigned to the Biochemistry Section as a Research Assistant.

James T. Post, Department of Biophysics, University of California, Berkeley, California; assigned to the Biochemistry Section as a Research Assistant.

Wilfred D. Stedman, University of Colorado, School of Medicine, Denver; assigned to the Radiobiology Section as

a Research Assistant.

Jean D. Van Riper, University of Rochester,
College of Medicine, Rochester, New York; assigned to the
Biochemistry Section as a Technician.

Jo Ann Grometer, University of Colorado, Boulder,
Colorado; assigned to the Biochemistry Section as a Technician.

L. Edward Ellinwood, University of Colorado,
School of Medicine, Denver; assigned to the Radiobiology Sec-
tion as a Staff Member.

2. Progress Reports of the Various Sections

2.1 Biochemistry Section (Gordon Gould, Section Leader)

2.1.1 General Comments on Section Activities

The investigative activities of the Biochemistry Section during 1954 may be grouped under the following general categories.

a. Beta ray determinations on urines from persons exposed to the fall-out of March 1, 1954.

b. Sterol and lipid metabolism in experimental animals and humans, using H^3 and C^{14} , including the effects of X irradiation, hormones, disease, diet, absorbability, and metabolic effects of various T-sterols.

c. Protein metabolism, including the use of filter paper electrophoresis to determine the effects of X irradiation and disease on the concentrations of serum proteins, and to investigate the binding of Pu, Sr, and other fission products with serum proteins.

d. Chelating agents (EDTA, B.A.L., and Perma-kleer), for removal of radioactive metals from humans and experimental animals, including toxicity studies.

e. Strontium metabolism and the comparative rates of turnover in bone of Sr, Ca and Pu.

f. Effect of X irradiation on mineral metabolism (Na, Pu).

g. The determination of cholinesterase titers in a variety of clinical conditions and its significance.

h. Effect of X irradiation on cholinesterase titer in mice.

i. Effects of massive doses of gamma and X rays on cholinesterase in vitro and in vivo.

2.1.2 Publications, Documents and Reports Originating
in the Section

The following list of publications, documents, and reports originated by the various members of the Biochemistry Section includes all material prepared for publication in the open literature, all documents for issue as LASL reports, and all material prepared for distribution to and through outside agencies. All publications, etc., that were in final manuscript as of December 31, 1954, are included.

(1) Biochemical Aspects of Atherosclerosis, R. G. Gould, J. Am. Ger. Soc., 2, 640 (1954).

(2) Absorbability of Dihydrocholesterol and Sitosterol, R. G. Gould, with the technical assistance of L. V. Lotz and K. C. Kohr, Circulation, 10, 589 (1954).

(3) Dietary and Hormonal Influences on Hepatic Cholesterol Synthesis in Chicks, J. Stamler, R. G. Gould, C. Bolene-Williams, Circulation, 10, 612 (1954).

(4) Absorption and Metabolic Effects of Dihydro-cholesterol, R. G. Gould, L. V. Lotz, E. M. Lilly, Federation Proc., 14, March (1955).

(5) Sterol Metabolism and Its Control, R. G. Gould, Nat. Res. Coun. Symposium on Atherosclerosis, to be published in a book or bulletin by the Nat. Acad. of Sci. U.S. (in proof).

(6) Cholesterol in Atherosclerosis, R. G. Gould. (A 1000-word abstract with 25 charts, tables, etc., is being published by the Am. Chem. Soc., in book form.)

(7) Experimental Administration of Ethylene-diamine-tetraacetic Acid in Plutonium Poisoning, Harry Foreman, P. A. Fuqua, W. D. Norwood, A.M.A. Arch. Ind. Hyg. and Occupational Med., 110, 226 (1954).

(8) Cholesterol Metabolism. II. Influence of Dietary Cholesterol on the in vivo Rate of Cholesterol Synthesis, C. B. Taylor, N. E. Eckles, J. S. Hagerman, D. J. Campbell, R. G. Gould. (Accepted for publication by J. Biol. Chem.)

(9) Cholesterol Metabolism. III. The Origin of Plasma Cholesterol and the Interrelations of Liver, Plasma, and Erythrocyte Cholesterol, N. E. Eckles, C. B. Taylor, D. J. Campbell, J. S. Hagerman, P. Keegan, R. G. Gould. (Accepted for publication by J. Lab. Clin. Med.)

(10) Cholesterol Metabolism. IV. The Use of C^{14} -Acetate to Study Cholesterol Metabolism in Man,

R. G. Gould, G. V. LeRoy, G. T. Okita, J. J. Kabara, P. Keegan, D. M. Gergenstal. (Accepted for publication by J. Lab. Clin. Med.)

(11) Cholesterol Metabolism. V. Interrelations of Blood and Tissue Cholesterol, D. J. Campbell, C. B. Taylor, F. B. Kelly, Jr., J. S. Hagerman, K. C. Kohr, P. Keegan, R. G. Gould. (In manuscript.)

(12) Ca EDTA and the Excretion of Radioactive Metals, Harry Foreman, T. T. Trujillo, O. Johnson, C. Finnegan. (In manuscript.)

(13) Clinical Significance of Erythrocyte Cholinesterase Titers. Part I. A Method Suitable for Routine Clinical Use, and the Distribution of Normal Values, Jean Captain Sabine, M. D. (In manuscript form intended for publication in Blood, the Journal of Hematology.)

(14) I. A Laboratory Outline for a Cholinesterase Method Suitable for Routine Clinical Use, and II. Present Status of the Clinical Applications of Cholinesterase Determinations. Prepared by J. C. Sabine at the request of Dr. L. E. Young of the Department of Medicine, University of Rochester.

(15) Chelating Agents and Stability Constants, Harry Foreman, LA-1851, and Proc. of Second Annual Conference on Pu, Ra and MsTh. (June 19, 1954, Salt Lake City, Utah.)

(16) Funds Available for Support of Research

Projects and of Fellowship Programs in the Field of Atherosclerosis. Prepared by R. G. Gould at the request of the Am. Soc. for the Study of Arteriosclerosis.

2.1.3 Progress of Specific Projects

1. Beta Ray Determinations on Urines of Personnel

Exposed to Fall-Out of March 1, 1954 (H-4)

Urines from U. S. personnel and natives on Rongelap and neighboring islands and from Japanese fishermen collected shortly after the March 1, 1954, fall-out were analyzed for I^{131} , Sr^{89} , Ba^{140} , Ru^{103} , Ca^{45} , Ce^{141} , and fissionable material. Total beta activity of dried urine, dry-ashed residue, wet-ashed residue, and the alkaline precipitate was also studied. Most of the activity in the samples was due to I^{131} and Sr^{89} . Ba, Ca, and Ru had slight activity and Ce and fissionable material none. Both I^{131} and Sr^{89} could be determined directly in the dried urine samples by extrapolation of decay curves, and Sr^{89} could be measured in the dry-ashed residues.

A preliminary report including all the procedures used has been completed for future use of H-4, if necessary. A final weapon test (WT) report, combining these results with the gamma ray determinations by the Radiobiology and Biophysics Sections, will be written.

2. Absorption and Metabolic Effects of Dihydro-
cholesterol and Sitosterol (Gould, Lotz, Lilly)

Extension of previous studies on absorption of tritium-labeled sterols has shown that both dihydrocholesterol (DHC) and beta sitosterol are absorbed in very appreciable amounts by experimental animals and man, contrary to all previous literature on the subject. Sitosterol is being fed to humans on an experimental basis by several investigators with the hope that by decreasing plasma cholesterol levels over a long period of time, the incidence and severity of atherosclerosis may be decreased. It is, therefore, of importance to determine how much is absorbed, its metabolic effects and fate in the body. Much help has been obtained from Eli Lilly and Company, who are particularly interested in the absorption of sitosterol. The Biochemistry Section has been in an unique position to carry on such studies, because of the availability of high activity tritium water for labeling and of the liquid scintillation counter.

Two abstracts have been sent in, one of which was published in October 1953, and a detailed paper is in preparation. The results show that beta sitosterol is absorbed about one-fifth to one-tenth as well as cholesterol. However, it does not accumulate in the body, partly because the absorbed sterol is metabolized faster than cholesterol,

and partly because the per cent absorption is lower for larger doses.

The results also show that dihydrocholesterol is absorbed sufficiently to replace a considerable fraction of liver and blood cholesterol at least for a time. After 5 days of feeding a 2 per cent DHC diet to rats, both the plasma and liver total sterol content increased by about 25 per cent, whereas the cholesterol content (determined colorimetrically) decreased by about the same amount, the difference being due presumably to DHC. The absorption of DHC was also demonstrated using C¹⁴- and T-labeled materials and it was shown that some of the DHC was converted back into cholesterol.

This project is to be continued in 1955.

3. Effect of X Irradiation on Metabolic Processes

(Gould, Keegan, Van Riper)

Previously reported results on the increase in the rate of hepatic cholesterol synthesis in rats produced by 2400 r of X radiation were confirmed. An average increase of 500 per cent was observed.

No effect was found in the intestine, an organ that in rats has a more rapid rate of cholesterol synthesis than liver.

The effect in spleen was the same as in

liver; however, it is not certain how much of this effect was due to transfer of newly synthesized molecules from liver to spleen.

No significant effects on fatty acid synthesis were noted.

Protein synthesis in liver, intestine, and various other tissues is now under investigation.

4. Hormonal Effects on Cholesterol Synthesis in Chicks (Gould, Lotz, Stamler, Bolene-Williams, in collaboration with Michael Reese Hospital, Chicago, Illinois)

The effects of estrogens, compound F (Hydrocortisone), thyroid hormone, and cholesterol on the rate of hepatic cholesterol synthesis, the levels of free and esterified cholesterol in liver and plasma, and the distribution of newly synthesized molecules in these four fractions were determined. Both estrogen and hydrocortisone increased the rate of hepatic synthesis. Cholesterol feeding suppressed synthesis almost completely and the addition of thyroid had no effect on the cholesterol-fed chicks. Of most interest was the demonstration that the effect of estrogens was primarily on the distribution of cholesterol between liver and plasma, since it increased the plasma level greatly without affecting the liver level. Hydrocortisone also increased the plasma level without changing that in liver.

5. Effect of Hypothyroidism on Cholesterol
Syntheses in Humans (LeRoy, Gould, Kabara, Bergenstal)

Additional studies on myxedema patients have confirmed the findings previously reported that synthesis of cholesterol is practically completely inhibited in this condition in spite of the greatly elevated plasma level. Various types of therapy have been found to raise the rate to normal. Tri-iodothyronine has a very rapid effect in increasing the rate of synthesis, before any change in level is produced, thus showing that this is a direct effect on the liver.

This work will be published as a collaborative project between Group H-4 and the Argonne Cancer Research Hospital, Chicago, Illinois.

6. The Use of T-Cholesterol and C¹⁴-Acetate in
Studying the Origin of Steroid Hormones and Their Metabolism
(LeRoy, Werbin, Gould, Lotz, Davison)

A series of 7 patients undergoing total adrenalectomy or therapeutic abortions at the Argonne Cancer Research Hospital were studied metabolically by the double labeling technique. Each patient was fed 50 μ c. of T-cholesterol on the first day and 10 μ c. per day for about 1 week before surgery. The patient was then given 100 μ c. of C¹⁴-acetate intravenously while anesthetized, about 1 to 2 hours before removal of adrenals, uterus, etc.

Free or total cholesterol was isolated from all biopsy samples, including liver and plasma, and urine was collected post-operatively for isolation of individual 17-KS compounds, particularly tetrahydrocortisone. All samples were assayed for both C^{14} and T in the liquid scintillation counter. The results are summarized as follows:

(a) Synthesis of cholesterol in the normal human adrenal has been clearly demonstrated.

(b) The conversion of dietary cholesterol into steroid hormones (with the possible exception of estrogens) has been found to occur to a considerable extent.

(c) The rapid appearance of C^{14} in steroid hormones suggested that they are also partly derived by direct synthesis from small fragments, as well as from cholesterol by degradation. However, cholesterol synthesis in the adrenal was so rapid that the pathway

C^{14} -acetate \longrightarrow adrenal free cholesterol \longrightarrow adrenal cortical hormones could not be definitely excluded.

(d) Rapid synthesis of cholesterol in human carcinomas was observed in one case.

(e) A much more rapid rate of cholesterol synthesis was observed in the ovary containing a corpus luteum than in the other ovary.

7. Relative Rates of Synthesis of Cholesterol and Fatty Acids in Various Organs and Tissues with Special Reference to the Aorta (Gould, Keegan, Lotz, Lilly)

The flash labeling method has been applied to the problem of determining the relative rates of synthesis in 10 tissues, by intracardiac injection of C^{14} -acetate of high specific activity 30 minutes before sacrificing the animals. The assumptions made were: (1) equal distribution of the C^{14} -acetate in all tissues; (2) equal dilution by unlabeled acetate in all tissues; and (3) a sufficiently rapid turnover rate of acetate and all the intermediates in sterol and fatty acid synthesis so that maximum incorporation of C^{14} in both will have occurred by the end of 30 minutes.

Results of a typical experiment are given in Table 2.1.1.

Intestine was the most active tissue both per gram and for the whole tissue; liver and spleen had about equal rates per gram, but the spleen's contribution to total body cholesterol synthesis was very small because of its small size. Both skin and residual carcass fractions had considerably slower turnover rates, but because each constituted such a large fraction of the total body mass, their contributions to body cholesterol were of major importance.

Only in the liver was the rate of synthesis

markedly variable; it could be increased 5-fold by X irradiation or decreased to practically zero by cholesterol feeding, as previously described.

Cholesterol synthesis was demonstrated in aorta by this in vivo method in chickens; in rats the results were still of doubtful significance, but indicated a very small, if not negligible, rate of synthesis.

This project is to be continued in 1955.

Table 2.1.1 - SPECIFIC ACTIVITIES OF TISSUE CHOLESTEROL
HALF AN HOUR AFTER INJECTION OF 20 μ c. C^{14} -ACETATE
PER 100 g. BODY WEIGHT

Tissue	Cholesterol S. A. (μ c./g. Cholesterol)	Fatty Acids S. A. (c/m/mg.)
Intestine, free	2.40	2100
Liver, free	0.63	
Plasma, free	0.05	
Spleen, free	1.07	
Kidney, total	0.32	2200
Skin	0.30	300
Lungs	0.26	5600
Testes	0.22	1500
Carcass Residue	0.21	400
Heart	0.05	1400
Skeletal Muscle	0.03	

8. Filter Paper Electrophoresis Studies of the
Effect of X Irradiation on Serum Globulin Levels in Rats
(Magee, Gould)

Preliminary experiments indicated a striking decrease in concentration in the globulin fraction, particularly in the β and γ components, as early as 24 hours post-irradiation with 1000 r of X rays. Concentrations returned to normal by about 1 week. Although this effect must presumably have been observed by others, a cursory examination of the literature has not revealed anything relevant.

9. Binding of Pu, Sr⁸⁹, and Other Fission Products
with Serum Proteins (Magee, Foreman, Gould)

Pu²³⁹ was injected intravenously into rats and blood was taken at 0.5, 2, 6, and 24 hours later, and serum samples separated by filter paper electrophoresis. Preliminary results indicated that Pu was bound to all the globulin fractions but not to albumin. Sr⁸⁹ under similar conditions gave results suggesting that part of it remained at the point of application, suggesting unbound Sr, and part of it moved with globulins.

Dialysis of sera from both Pu and Sr bloods indicated that each was partly dialyzable and partly non-dialyzable. Serum from rats taken half an hour after injection of Pu and dialyzed for 4 hours lost 37 per cent of

the activity. With Sr^{89} , 65 per cent of the activity was lost by dialysis under the same conditions.

10. Application of Filter Paper Electrophoresis to Clinical Laboratory Diagnosis (Magee, Gould)

At the request of Dr. H. B. Ellis, Jr., Los Alamos Medical Center, a number of sera were run to look for cases of agammaglobulinemia. No positives have been found as yet. Both serum and urine from a case of multiple myeloma were studied; a high concentration of a serum component with a mobility between β and γ globulin was found. The urine gave a broad band with about the same mobility as the abnormal serum component.

11. Ca EDTA in Pu Excretion (Foreman, Moss)

An occasion to test the efficacy of Ca EDTA in plutonium contamination in a human arose in a worker who acquired a subtolerance body burden of Pu through a contaminated laceration. It was estimated that the body burden was approximately one-half tolerance and that 16 days of treatment removed 25 per cent of this. The treatment was started on the sixth day after exposure. The maximum effect occurred on the first day of treatment, namely, 100-fold increase in the urine Pu over pre-treatment level.

12. Trial of B.A.L. in Po Excretion (Foreman, Robbins)

B.A.L. (1.7 cc.) was given intramuscularly to an individual who was excreting Po at a level in the urine which indicated a body burden of 0.1 tolerance. Two days of treatment did not alter the urinary excretion rate.

13. Kidney Damage from Ca EDTA (Foreman, Finnegan)

In view of the observation that 16 days of treatment with Ca EDTA was followed by evidence of kidney disturbance, i.e., casts, cells, and albumin in the urine, and unusual kidney findings at autopsy of individuals who had received large doses of EDTA, it was decided to give Ca EDTA to rats in varying doses to see if kidney lesions could be produced and the smallest dose level at which lesions would first occur. Beginning at levels of 250 mg./kg./day given 16 consecutive days, tubular damage was found, first manifest in the proximal tubules and extending to the remainder of the lower nephron as the dose levels increased. The alterations in structure ranged from vacuolization of the tubular epithelial cells and intraluminary secretions at the low dose levels to loss of cellular outlines and complete disruption of cellular structure at the higher dose levels.

There was evidence that the early stages of damage were reversible, which suggested that dosage schedules should provide for intermittent courses of treatment.

Further studies involving the reversibility of the lesions and potential toxicity associated with intermittent treatment will be carried out.

14. Effect of Ca EDTA Therapy on Lung Absorption of Pu (Foreman, Finnegan)

The possibility that Ca EDTA administered intraperitoneally might accelerate the transfer of Pu across the lung alveoli and thereby increase the body burden over that which might occur with untreated case was explored.

Rats were administered Pu intratracheally and then given a course of Ca EDTA intraperitoneally. The drug did not effect any change in transfer of Pu across the lung membranes, since none of the tissues or excreta of the treated animals showed any significant difference in Pu content from the control animals.

15. Effect of Ca EDTA on Absorption of Pu from Intramuscular Deposits (Foreman, Finnegan)

Two groups of rats were administered Pu deep into the muscle of the hind leg. One group was given a course of Ca EDTA. At the end of 4 days the animals were sacrificed and tissues taken for analysis. There was no significant difference between the controls and the experimental group. About 95 per cent of the injected dose was

still at the site of injection. This would suggest that even when Ca EDTA was used, one could still remove a major portion of Pu from a contaminated puncture wound by debridement as late as 4 days and later after an accident.

16. Tests of New Chelating Agents for Removal of Systemically-Bound Heavy Metals (Foreman, Finnegan)

n-Hydroxyethylethylenediaminetriacetic acid (Permakleer 80) and a component known to us only by a trade name Permakleer SP were tested for the ability to affect excretion of Pu. These compounds are both of interest because of their very high solubility in water throughout the pH range. Distribution and excretion studies with Pu, using these agents, showed them to be similar to EDTA in their effect on Pu metabolism when given intraperitoneally. Used orally, they produced much more excretion than EDTA, i.e., between 5- to 10-fold increase over the controls. This was probably due to their solubility at the pH of the stomach. This property makes them of interest and worthy of further study.

17. Strontium Metabolism Studies (Foreman, Finnegan)

(a) Excretion of Strontium. A new approach to the problem of hastening the excretion of Sr from the body

was investigated. About 99 per cent of Ca (and presumably Sr) in the glomerular filtrate was resorbed through the tubules. Alteration of tubular resorption, even to a small extent, could increase Ca and/or Sr excretion manyfold. Two agents, Diamox and Bevenrid, both known to affect tubular resorption, were tested for their ability to alter Sr resorption. No effect was noted. Investigation of this approach will be continued.

(b) Comparative Metabolism of Ca, Pu, and Sr (Finnegan). This study has two objectives: (1) to compare the rate at which Ca, Sr, and Pu are turned over in the skeleton over a long period of time; and (2) to gain information as to the site of deposition of Pu in bone. The approach to the first objective was to inject a triple tracer of Ca^{45} , Sr^{90} , and Pu^{239} in a given ratio and then with serial sacrifice determine the change of the ratio with time. The nature of Pu binding in bone was investigated by determining the per cent of Pu in a given bone, as compared to the per cent of total ash in this bone with respect to the skeleton as a whole.

18. Effect of X Irradiation on Mineral Metabolism
(Foreman, Finnegan, Post)

Sodium turnover in rats after 500, 1000, and 1500 r was studied using the so-called arm counter, a liquid

scintillator 4π detector, which permits the counting of the whole animal. Na^{24} was injected into rats after radiation and followed by counting the animals periodically for several days.

It was found that the rate at which the Na disappeared from the irradiated rats, irrespective of dosage, was not significantly different from the controls, until approaching death (3 to 4 days for the 1000 and 1500 r animals), when the Na content of the body fell rapidly. The Na turnover time for the controls, as well as the irradiated animals, was 3-1/2 to 4 days, when the agonal period was disregarded.

The effect of X rays on the absorption of Pu from the gastrointestinal tract was studied using doses of 500, 1000, and 1500 r. The study was undertaken to simulate conditions in a pile accident or in fall-out of radioactive materials where it is probable that individuals receiving an internal dose of radioactivity, i.e., Pu and fission products, would also receive a concurrent external dose.

It was found that at these dose levels the absorption of Pu was less than 0.1 per cent of the administered dose, the same value that was found in the control animals.

19. The Clinical Significance of Red Cell Cholin-
esterase (Sabine, Miller)

The changes in cholinesterase titer observed in a number of clinical conditions confirmed the earlier finding that hyperactivity of the bone marrow was reflected by the cholinesterase titer of the peripheral erythrocytes more sensitively than by the reticulocyte count. In the present series of cases, the cholinesterase titer was found to have both diagnostic and prognostic value. One or more cases with each of the following clinical conditions were followed: acute and chronic leukemias; congenital hemolytic icterus; secondary anemias; erythroblastosis fetalis; carcinoma; sarcoma; and multiple myeloma.*

Among the earliest changes in a remission of acute leukemia was a rising cholinesterase titer. A case of erythroblastosis fetalis had the usual very low titer of newborns during the first week of life and repeated transfusions were required; during the second week, the cholinesterase titer rose sharply, and no further transfusions were required. (Normal newborns retain the low titer at least 2 weeks.) Among three cases of icterus, one was of hemolytic origin and had very high cholinesterase titers,

* The cases studied were made available through the courtesy of the Medical Staff of the Los Alamos Medical Center.

which persisted for months after therapeutic splenectomy; a child of this patient has a normal hemogram and a cholinesterase titer far above normal, suggesting a subclinical hemolytic condition. The remaining two cases of icterus had abnormally low cholinesterase titers. Both were explored and found to have carcinoma of the head of the pancreas. A case of reticulosarcoma with metastases had a very high titer and moderate anemia when first seen. The titer gradually fell and when it reached the upper limit of the normal range, a rapidly progressing anemia set in. In none of these cases was the reticulocyte count significantly elevated.

It is planned to follow appropriate cases as they become available.

20. Effect of Total Body Irradiation on the Erythrocyte Cholinesterase Titers of Mice (Sabine, Miller)

This is the conclusion of a report of the same title which appeared in the Annual Report of 1953.

Mice subjected to doses of X rays from 25 to 525 r (from 250-KVP machines with various filters) showed significantly increased titers of red cell cholinesterase on the third and fourth days, as shown in Table 2.1.2.

Table 2.1.2 - CHOLINESTERASE TITERS (CHE)
FOLLOWING VARIOUS DOSES OF X RAYS

Dose (rep)	No. Animals per Group	4th Day CHE*
25	36	1.15
50	19	1.18
100	12	1.17
150	11	1.10
200	12	1.30
300	27	1.33
450	12	1.10
525	14	1.19

* The means are referred to the control mean as 1.00

After the fourth day following a dose of 300 r, there was a sharp drop in cholinesterase titer, to values significantly below the normal range, as shown in Table 2.1.3.

Table 2.1.3 - CHOLINESTERASE TITERS (CHE)
AND RETICULOCYTE COUNTS FOLLOWING 300 r X RAYS

Days Post- Irradiation	CHE*	No. Animals per Group	Retics.*	No. Animals per Group
0	1.00	122	1.3(0-4.7)	16
3	1.26	8	0.2	9
4	1.33	27	0.2	19
5	1.18	5		
6	0.91	13	0.4	16
8	0.65	9	0.2	5
9	0.66	8		
12	0.81	6	2.1	7
14	0.86	8	1.5	10
17	1.03	9	3.1	9
20	0.97	6		
21	1.01	8		
35	1.00	16		

* Data are from four groups of mice irradiated at different times. Each value above 1.20 and below 0.80 is significantly different from its own control mean.

About the twelfth day, the titers began to rise and by the seventeenth day, were normal. Injection of plutonium (6.7×10^5 c./m.) produced a picture virtually identical to that shown in Table 2.1.3 during the first 10 days, but there was no recovery. The titers remained at 0.6 for 65 days, the longest period of observation.

The two increases in titer (0 to 4th days and 12th to 17th days) following 300 r are comparable in magnitude, yet one is accompanied by a suppressed reticulocyte count and the other by a small but significant reticulocytosis.

The events during the second week following irradiation resembled those which are seen in certain clinical conditions, whereas the sudden rise and fall during the first week does not resemble anything seen previously. Clinically, a high or rising titer is associated with increased erythropoiesis and it has been shown experimentally that young red cells have a much higher concentration of enzyme than older cells (publications 1940 and 1951). The events during the second and third weeks certainly represent a recovery process; those during the first week cannot be interpreted at present.

Analysis of the fourth-day data makes it appear that the very great increases (up to twice the mean control value) may be the result of a threshold reaction occurring in some individuals and not in others.

The possibility that the increased titer (which is expressed per unit volume of cells) on the fourth day might result from cell shrinkage has been ruled out by appropriate experiments.

The findings presented above indicate that an insult is delivered by doses of radiation too small to alter the conventional red cell hematology and that, with the sub-lethal 300-r dose, recovery from this insult occurs during the second and third weeks post-irradiation. What happens at very low doses and at higher doses is not known, except for the fourth day.

This project has been discontinued. The data on hand will be written up and submitted for publication, since it is believed that evidence of any effect at doses as low as 25 r will be of interest, even though the work is obviously incomplete.

21. Disappearance from the Peripheral Circulation of "True" Cholinesterase Released from Erythrocytes by Hemolysis (Sabine, Spalding, Schweitzer, Miller)

Since larger than trace amounts of this enzyme (which occurs in high concentration only in nervous tissue and erythrocytes) are occasionally found in plasma in clinical conditions, a simple experiment was designed to find out whether the enzyme released by hemolysis remained in the

circulation. A 100-pound sheep was subjected to phlebotomy and 250 ml. of blood were obtained. The blood was hemolyzed by freezing and thawing and re-injected a few days later with 500 ml. of lactate-ringer. Blood samples were taken at 5 minutes after the end of the transfusion and at 24 hours. The 5-minute sample showed bright red plasma containing 0.67 g. of hemoglobin per 100 ml. The "true" cholinesterase activity of the plasma was precisely that of the plasma during the control period. The activity of the hemolyzed blood was such that if the enzyme had remained in circulation in proportion to the hemoglobin, its activity would have been readily detectable. The 24-hour sample had no hemoglobin in the plasma and the same trace of cholinesterase activity.

This procedure was repeated with several injections of india ink during the week preceding the transfusion. The results were precisely the same as before. At autopsy, the liver and spleen were grossly coal black and microscopic examination revealed heavy black deposits.

The results described above are compatible with the well-supported belief of several investigators that the enzyme is firmly attached to the cell membrane and remains attached to the fragments after the cell is hemolyzed.

This project has been discontinued. The results will receive brief mention in a forthcoming clinical paper.

22. Inactivation of Cholinesterase by Gamma Radiation and by X rays: A Possible Biochemical Dosimeter (Sabine, Miller)

Although no inactivation of cholinesterase in vivo has been found with doses up to 60,000 r of gamma radiation of energy 1.3 Mev from a barium-lanthanum source, purified cholinesterase in relatively dilute media is inactivated by such radiation and to a much lesser degree by 250 KVP X rays. In all cases, the relation between dose and effect is linear, not logarithmic. The slope can be varied widely by altering the chemical environment of the enzyme.

With 1.3-Mev gamma radiation, the inactivation was 2 per cent per 1000 r in phosphate buffer, 0.5 per cent if gelatin was added to the buffer. With X rays, the inactivation was 0.13 per cent per 1000 r, if the enzyme was in dilute sheep serum.

Comparison with other types of radiation has been deferred, pending an opportunity to investigate more thoroughly the effects of gamma radiation from a high dose-rate source.

The slope of 2 per cent per 1000 r mentioned above promises a feasible, though technically difficult and exacting, method of measuring gamma radiation from about 1500 to 40,000 r by means of a biological preparation. The sensitivity of such

a dosimeter can be increased if a medium can be found which will give less protection against radiation inactivation and at the same time protect the enzyme from spontaneous deterioration.

23. The Effects of Whole Body X Irradiation on
Intestinal Transport of Tritium Water and Na²² in the Fasted
Rat (Armstrong, Goodner, Moore)

As a part of the program to study the effects of radiation on the physiology of the gastrointestinal tract, studies were conducted concerning the effect of radiation on the transport of water and sodium across the intestinal mucosa. Rather extensive studies, using tritium-labeled water and Na²² as tracers, were carried out on the fasting rats following radiation doses of 2000 r. The Na²² and the tritium in the blood and in the intestinal contents were counted, using an internal liquid scintillation counting system. Both isotopes were counted in the same sample by using pulse height discrimination. Studies were made at 6, 9, 12, 15, and 30 minutes after injection of the radioisotope into the lumen of the gut. The isotopes were placed in the gut at various times after radiation exposure, ranging all the way from a few minutes to several hours. Although no drastic changes were observed, the results indicated rather conclusively that following radiation there is a loss of body fluid and body sodium through the gastrointestinal tract.

2.2 Radiobiology Section (Payne S. Harris, Section Leader)

2.2.1 General Comments on Section Activities

The activities of the Radiobiology Section during 1954 may be summarized under the following general categories:

- a. Studies of relative biological effectiveness of various radiations for the production of response in mammalian systems.
- b. The calibration and radiation characteristics of Godiva for biological studies.
- c. Effects of radiation on gastrointestinal function and physiology.
- d. Median survival time of animals exposed to massive rapid doses of neutrons.
- e. The relation between bacteremia and death of irradiated animals.
- f. Metabolic studies in bacteria using C^{14} -labeled materials.
- g. Analysis of urines of persons exposed to radioactive fall-out of March 1, 1954.
- h. Preparation for participation in 1955 weapon test series (Operation Teapot).

2.2.2 Publications, Documents and Reports Originating in the Section

- (1) Exposure of Monkeys to High Doses of Fission

Neutrons from Godiva, Wright Langham, John E. Pickering (Lt. Col. USAF), D. V. L. Brown (Capt. USAF MC), F. S. Vogel (Capt. USAF MC), C. C. Lushbaugh, LA-1838 (classified).

(2) Harwell Conference on Human Monitoring in the Control of Occupational Health Hazards in Atomic Energy, Wright H. Langham, WASH-490, October 1, 1954.

(3) Isolation of C^{14} -Labeled DPN and TPN by Paper Chromatography from Lactobacillus Plantarum, Irene U. Boone, Donna F. Turney, Kent T. Woodward, Science, 120, 312 (1954).

(4) Biological Effectiveness of 14 Mev Neutrons: Spleen and Thymus Weight Loss in Mice as the Biological Indicator, Payne S. Harris, L. E. Ellinwood, LA-1629, March 1954.

(5) The Biological Effectiveness of Thermal Neutrons on Mice, James T. Brennan, Payne S. Harris, Robert E. Carter, Wright H. Langham, Nucleonics, 12, No. 2, 48; 12, No. 4, 31 (1954).

(6) Studies of the Effects of Massive, Rapid Doses of Gamma Rays on Mammals, Wright H. Langham, Samuel M. Rothermel, Kent T. Woodward, Clarence C. Lushbaugh, John B. Storer, Payne S. Harris, LA-1643, May 1954.

(7) Biological Effectiveness of Thermal Neutrons in Producing Testicular Atrophy in Mice, John B. Storer, Wright Langham, LA-1630, March 1954.

(8) The Chronic Toxicity of Uranium²³³ and Uranium²³⁸: Delayed Effects in Rats following a Single Injection, John B. Storer, Wright Langham, L. E. Ellinwood, LA-1634, April 1954.

(9) Relation between Specific Ionization of Various Radiations and their Relative Biological Effectiveness in Mammalian Systems, John E. Furchner, LA-1849, September 1954.

(10) Measurement of Slow Neutrons and Coexisting Radiations, P. S. Harris, Radiation Research, I, No. 1, February 1954.

(11) Studies on the Mechanism of Action of Thermal Neutrons in Producing Biological Effects. II. Comparative Effectiveness in Producing Intestinal Damage, Lethality, and Marrow Damage in Rats, John B. Storer, John E. Furchner, LA-1848, September 1954.

2.2.3 Progress of Specific Projects

1. Isolation of C¹⁴-Labeled DPN and TPN by Paper Chromatography from Lactobacillus plantarum (Boone, Turney, Woodward)

When Lactobacillus plantarum was grown in media containing C¹⁴-nicotinic acid or amide or when resting cells were suspended in saline containing the labeled materials they converted 10 to 20 times the amount required for maximum growth to DPN and TPN. The labeled DPN and TPN was leached

from the cells by washing with distilled water adjusted to pH 4 and the materials were separated and identified by filter paper chromatography.

This extremely rapid conversion of C^{14} -nicotinic acid to C^{14} -DPN by cells washed free from the growth medium and suspended in saline plus nicotinic acid indicated that all other moities of the DPN molecule (phosphate, ribose, and adenine) must already be present in some form in the "resting cell." That an exchange phenomena between unlabeled DPN and C^{14} -nicotinic acid is involved seems unlikely, since "resting cells" (upon being suspended in saline containing C^{14} -nicotinic acid) convert to DPN 10 to 20 times the amount of nicotinic acid supplied in the medium during their growth phase. Furthermore, the molar specific activity of the C^{14} -DPN, if formed by exchange, would approach only one-half that of the nicotinic acid.

Quantitatively, 100 μ g. of C^{14} -nicotinic acid with a specific activity of 4.07 mc./mmole in the presence of from 50 to 100 mg. dry weight of cells of Lactobacillus plantarum yielded about 250 to 300 μ g. of C^{14} -labeled DPN with equivalent molar specific activity. Some, but not all, of the C^{14} activity remaining in the medium may be removed by further cell absorption or as C^{14} -nicotinic acid or amide by ether extraction at the proper pH.

It is felt that considerable advantage may be offered by this method as a means of obtaining small amounts of pure C^{14} -labeled DPN and TPN with rather high specific activities.

2. Relationship of Bacteremia to Death in Mice following X Rays and Thermal Column Exposures (Boone, Stedman, Grometer, Harris, Woodward)

The purpose of this study was: (1) to attempt to correlate specifically mortality with bacteremia; (2) to determine the time-dose bacteremia relationship in heart blood and various tissues or organs in the LD_{50}^{30} range; (3) to study the incidence of bacteria in heart blood and tissues at very high doses. The data correlating per cent bacteremia at 9 days and lethality at 11 days, following doses of 400 to 800 r of X rays and 480 to 616 rem of thermal column exposure, showed that all mice which died during the first 11 days had bacteremia. The relative potency of the two radiations for the production of lethality and bacteremia was 1.8. At doses of 640 to 950 r of X rays and 480 to 765 rem of thermal column exposure, the percentage of positive tissue and blood cultures was greatest during the days surrounding the median survival time. This was extremely dose-dependent. The lower the dose, the greater the period of time between exposures and appearance of positive cultures. There were no positive blood or

tissue cultures for the first 3 days post-irradiation, except that following 950 r and 765 rem, positive tissue cultures appeared on the third day and continued until death.

The median survival time of animals receiving 3000 r or rem and 10,000 r was between 3 and 4 days, as expected. After the 3000 r or rem dose, blood cultures were essentially negative for the 3 days before death, but by the third day, a large percentage of the mesenteric lymph nodes and spleen cultures were positive. At 10,000 r, some of the tissues and blood cultures were positive by the first day and continued so until the death of the animal. After 23,000 r, with a median survival time of 30 to 36 hours, tissues and heart blood cultures were positive as early as 12 hours after exposure. It is doubtful if there is any direct connection between the presence of bacteria in the tissues and mortality in this high dose range, but the study clearly indicated a rapid, complete collapse of all mechanisms of host resistance.

3. Isoniazid in Pyridoxine-Deficient Rats (Boone, Turney)

Rats fed a diet deficient in pyridoxine and injected daily with the maximum acceptable oral dose of isoniazid showed a greater retardation of growth than rats receiving the same diet plus the known antimetabolite, desoxypyridoxine. The thymus gland had atrophied to the same

proportion of body weight in each of these two groups. In deficient animals receiving either desoxypyridoxine or isoniazid, urinary xanthurenic acid excretion was 20 to 50 times greater after oral tryptophane than in animals receiving over twice as much isoniazid but also 50 µg. of pyridoxine daily.

Acrodynia was very severe in the animals receiving desoxypyridoxine but never occurred in any of the animals receiving isoniazid. The occurrence of convulsions was more pronounced in the animals receiving 50 mg./kg. isoniazid daily and in those receiving over twice as much isoniazid plus 50 µg. pyridoxine daily than in deficient animals receiving no isoniazid.

The animals receiving the B₆-deficient diet and over twice the accepted oral dose of isoniazid daily were protected against severe retardation in growth, thymus atrophy, and excretion of xanthurenic acid in the urine by 50 µg. of pyridoxine daily. This amount of pyridoxine partially protected the animals from convulsions.

It has been shown that some, but not all, the symptoms associated with B₆ deficiency resemble the toxicity of isoniazid. Isoniazid toxicity was enhanced 6 to 12 times in pyridoxine-deficient animals.

It was possible to prevent the convulsions and reverse the symptoms of isoniazid toxicity if a sufficient dose of pyridoxine was given 30 to 60 minutes before

the isoniazid.

4. C¹⁴-Isoniazid Metabolic Study in Pyridoxine-Deficient Rats (Boone, Magee, Turney)

A metabolic study on B₆-deficient rats with C¹⁴-isoniazid was undertaken because of the previously observed relation between B₆ deficiency and isoniazid. A dose of 10 mg. of isoniazid per kilogram of body weight was injected prior to placing the animals in the metabolic cages. Carbon dioxide, feces, urine, and tissues were analyzed in the usual manner by direct plating and wet oxidation. Six B₆-deficient and 6 non-deficient animals were used per point. The rats were sacrificed and analyzed 1, 6, and 24 hours after injection. Very little difference in tissue concentration of the C¹⁴-isoniazid between the two groups was noted. Approximately 85 to 95 per cent of the drug was excreted in the urine at the end of 6 hours. There was no excessive concentration of the drug in any organ. Urinary chromatograms showed that at least 5 metabolites were present. Two bands were identified by their R_f values, unchanged isoniazid and isonicotinic acid. There did appear to be a slight difference in the rate of urinary excretion between the B₆-deficient and the non-deficient rats.

Blood level studies for turnover times and renal clearance of C¹⁴-isoniazid were determined in the two

groups. Plasma levels in animals with the kidneys tied off were nearly twice as high in the B₆-deficient animals as in the controls.

5. Particle Size Radiation Studies of the
Respiratory Tract of the Rat (Boone, Turney, with Anderson,
Worman, Perrings, L. Larkins)

A long-term study to determine the local effects of a radioactive particle in the lungs of rats was undertaken. Small spherical pellets containing varying amounts of alpha and beta activity are to be implanted in the lungs and followed for extensive periods for possible development of carcinomas. At present, the surgical techniques and physical problems have been worked out. It is possible to embed numerous small gold pellets into the lungs without the aid of thoracotomy and then locate them in the lungs by chest X ray. Post-mortem lung X rays with pathological sections of the beaded areas have been studied. As many as 17 gold pellets have been injected into one side of the lung without any harmful effects to the animal during the time of injections. All animals survived the procedure. Post-mortem examination of the lung tissue indicated no reaction to the gold in young animals. The rats have survived from 8 months to over 1 year after the implantation of the gold spheres. On autopsy, the animals showed a diffuse

pulmonary infection. The life-span of a normal Sprague-Dawley rat is from 1 to 2 years. The animals usually die from an endemic pneumonia. As a result of this, it may be necessary to use dogs for the long-term study.

Group CMR-5 has made one attempt at production of a gold-plutonium alloy. When such a gold-plutonium pellet was inserted into the peritoneum of the rat, the plutonium was slowly washed off by the body fluids. A gold-radioactive nickel alloy appears feasible for the beta radiation and CMR-Division is considering the preparation of a U^{233} -gold alloy as well as a PuO_2 -plastic pellet for the alpha radiation studies. The difficulty of production of the spheres is the limiting factor in these experiments at the present time.

6. Uptake of C^{14} -Labeled Isoniazid by Mycobacterium in the Presence of Reported Antimetabolites (Rogers, Boone)

The normal uptake of C^{14} -isoniazid by several strains and species of Mycobacterium was determined. The uptake was determined in the presence of numerous reported antimetabolites of isoniazid, such as pyridoxine, pyridoxal, pyridoxamine, sodium pyruvate, alpha-ketoglutarate, hemin, biotin, and Coenzyme I. No appreciable uptake of C^{14} -isoniazid over that of the controls was noted in the presence

of any antimetabolite except that of pyridoxal. In the H37-Rv-isoniazid sensitive strain of Mycobacterium tuberculosis var. hominis, the uptake of C¹⁴-isoniazid by the cells was increased 200 times over that of the controls. The mechanism of action of this increase in uptake in the presence of pyridoxal is being further investigated. An attempt will be made to identify the adsorbed radioactive compounds.

7. Determination of RBE using AK Mouse Leukemia in Various Strains of Mice (Boone, Turney, Rogers, Grometer, Stedman)

Numerous strains of mice were investigated for possible use in determination of an RBE with the use of AK mouse leukemia. This leukemia kills the mice in 7 to 10 days. After irradiation, mice not susceptible to the leukemia have a percentage of "takes" which is dependent on the dose of radiation the animal is given. The following strains of mice were investigated for possible use in the RBE determinations: CF₁, Strong A, Balb/c, C57 Brown, C57 Black, and Swiss CFW.

As a preliminary screening procedure, the various strains of mice were exposed to doses of X rays ranging from 100 to 550 r. Twenty-four hours after exposure, the mice were injected intraperitoneally with a suspension of AK leukemia. Leukemic "takes" were measured in terms of lethality of the mice following days after injection. The

CF₁, Balb/c, and Swiss CFW strains were unsatisfactory either as a result of high percentage of control "takes" or irregular responses after irradiation. The Strong A and C57 Black and Brown strains proved to be very satisfactory. The probit of the cumulative per cent leukemic "takes" (equivalent to mortality in this case) 11 days after injection is represented by the regression formula of the type

$$Y = a + b \log X$$

where Y = probit of per cent leukemic "takes" (or mortality) and X = dose. Analysis of the data at 11 days gave an LD₅₀ for leukemic deaths of approximately 280 r for the Strong A strain, and between 320 to 350 r for the C57 Brown and Black strains. A preliminary analysis of the data of the C57 Brown mice, exposed to various doses of X rays and thermal neutrons, indicated that the RBE will be close to the expected value of 2.0. The C57 Brown strain will be repeated to check the reproducibility of the data. Strong A and the C57 Black strains will be used, also, to determine the RBE for thermal, fission, and 14-Mev neutrons.

8. Analysis of Urines of Rongelap Natives Exposed to Radioactive Fall-Out of March 1, 1954 (All members of Section)

In collaboration with the Biochemistry Section, urines from the Rongelap natives exposed to the

radioactive fall-out of March 1, 1954, were analyzed for beta and gamma emitting fission products. For a summary report, see Secs. 1.1.1 and 2.1.3.

9. Rate of Weight Loss of Mouse Spleen and Thymus following Neutron Exposure (Woodward, Rothermel)

Weight loss of spleen and thymus has been used repeatedly as a quantitative indicator of biological effects. In all studies of RBE of various radiations by this method, the end-point has been the per cent weight loss of the organs on the fifth day after exposure. The assumption has always been made that the rate of weight loss (and the point of maximum loss) of the spleen and thymus was the same, following neutrons and other radiations, as it was for 250-KVP X rays. Experimental verification of this assumption was considered necessary. Groups of randomized mice were exposed to 300 r of 250-KVP X rays and 300 rem of mixed thermal column radiations. Groups of animals were sacrificed on Days 1, 2, 3, 5, 9, and 21, and the spleens and thymuses weighed. There was no significant difference between the rate of weight loss of the spleen and thymus of mice following neutrons and X-ray exposure. Spleen and thymus weights of animals receiving both radiations fell essentially to the same minimum value on Day 1, and stayed at that minimum through Day 5. It appears from these data that the third

day after irradiation is just as practicable for determining spleen-thymus weight decrease as is the fifth day. Three-day sacrifice of exposed animals will extend the dose range of this biological indicator by permitting sacrifice of the animals prior to the normal 3-1/2 day median survival time of animals exposed to doses in excess of 1000 r.

10. Cataractogenic Effects of Neutrons (Rothermel, Boone, Strang, Harris)

Monthly observations of mice exposed to thermal and 14-Mev neutrons in single and divided doses is still being continued. Some of these animals are now in their second year post-exposure. These data on chronic cataractogenic effects of neutrons will be summarized some time in 1955.

11. Effects of X Rays and Thermal Neutron Exposure on Gastrointestinal Physiology of the Rat (Woodward, Rothermel)

Relatively low doses of whole-body irradiation (50 r or rem) induced gastric retention of barium salts in rats, and the severity and duration increased directly with dose. Body weight decreased linearly with dose at 72 hours after thermal column and 250-KVP X-ray exposure, for doses up to 1200 r. Weight loss after doses from 1200 to 2500 r at 72 hours tended to approximate, but never exceeded,

that of nonirradiated starved and thirsted controls. The relative biological effectiveness of thermal column irradiation as compared to 250-KVP X irradiation was 1.0, using percent starvation weight loss at 72 hours as the biological indicator.

Surgical removal of the pylorus had no effect on gastric retention and is in agreement with Palmer's observations that after irradiation the pylorus is indeed patulous. The absence or diminution of antral persistalsis initiated by small bowel damage, or by direct damage of the stomach after whole-body irradiation, could explain the observed lengthy delay in gastric emptying after pylorectomy. This does not explain the failure of the stomach to empty after other stresses, including irradiation, which does not primarily involve the gastrointestinal tract.

Gastric retention had little or no direct effect on the food intake and on consequent body weight changes as totally gastrectomized animals lost comparable or greater body weight. The irradiated small bowel, unreceptive to food, might account for the diminished food intake, although the barium meal was observed to have a relatively normal time of transit from the esophago-duodenal anastomosis to the colon. Consequent body weight loss after superlethal doses paralleled and approximated that seen from starvation and water deprivation.

It was not possible to demonstrate any significant effect of adrenalectomy on gastric retention. That the animals were adrenalectomized was demonstrated by markedly enlarged thymus glands and normal spleens at post-mortem, which is in contrast to the usual state of lymphoid tissues after irradiation in the doses utilized.

12. Therapeutic Attempts at the Intestinal Effects of Radiation (Woodward, Rothermel, Schweitzer)

The 3- to 4-day death of irradiated animals has been ascribed to intestinal damage and has been produced by irradiating the intestine alone. Diarrhea is one of the pronounced signs of radiation damage. The role of fluids and electrolytes in the acute intestinal syndrome was investigated by treating the animals with a variety of fluids. Saline-glucose in the water, glucose-saline, and Ringer's lactate solutions were given to mice exposed to 1200 r of 250-KVP X rays. Multiple doses were given by intraperitoneal injection. The effects of all of these methods of treatment were of questionable significance. Survival of the animals that received physiological saline alone following 1200 r of radiation was increased by 20 per cent.

13. The Relative Biological Effectiveness of
14-Mev Neutrons for the Production of Lethality in Mice
(Woodward, Rothermel, Harris, Sanders, Schweitzer, Strang)

In the 1953 Annual Report, a preliminary value for relative biological effectiveness of 14-Mev neutrons was reported. Previous exposures of CF_1 mice to 14-Mev neutrons produced by deuteron bombardment of a tritium target in the Cockcroft-Walton Accelerator gave a rem value of 1×10^8 n./cm.² for spleen-thymus weight loss and 1.3×10^8 n./cm.² for lethality. These data were reported in the Annual Report for 1953; however, the data showed a considerable scatter and the study was repeated in 1954. It was possible to use the same group of randomized animals for concurrent experiments of lethality and spleen-thymus weight loss. Previous studies with thermal neutrons indicated a slightly lower rem value for the spleen-thymus weight loss than for 30-day lethality, as did the data already obtained on the Cockcroft-Walton for 14-Mev neutrons. A repeat of the data for 14-Mev neutrons gave a rem value of 1.1×10^8 n./cm.² for spleen-thymus weight loss and 1.7×10^8 n./cm.² for 30-day lethality.

14. Relative Biological Effectiveness of Fission
Neutrons from Godiva using Spleen-Thymus Weight Loss and
Lethality in Mice (Harris and Entire Section)

Approximately 900 mice were exposed in lucite cages at different distances from the Godiva assembly during a prompt burst of approximately 1×10^{16} fissions. Three different types of biological studies were made on the exposed animals. These were: (a) spleen-thymus weight loss; (b) 30-day lethality; and (c) median survival time.

By comparing the biological results obtained at Godiva with those obtained with 250-KVP X rays, it has been possible to show that the RBE of the fission neutrons indicated by each of the three test systems was approximately 2.

The median survival curve obtained with fission neutrons was essentially the same shape as that obtained with Ba-La gamma rays. Below doses of 1000 rem, median survival time was dose-dependent, followed by a region (approximately 1000 to 10,000 rem) over which the median survival time was 3.5 days and independent of dose. Above approximately 10,000 rem, median survival time was again dose-dependent. An exact evaluation of the RBE of fission neutrons from the Godiva assembly cannot be established until the neutron dose is adequately measured by physical means and until the inherent gamma radiation dose is also evaluated.

15. Calibration of the Godiva Assembly using
Biological and Physical Evaluation of Neutron Dose (Hurst
et al., ORNL; Harris et al., LASL)

The importance of physical dose measurements to the assessment of the true relative biological effectiveness of fission neutrons from Godiva led to a cooperative experiment between the Health-Physics Group of ORNL and the Radiobiology Section of Group H-4. The physical dose (in rep) delivered per neutron and the associated gamma dose were measured when Godiva was fired under burst conditions and when it was operated at a constant power level.

The neutron measurements were made using the fast neutron proportional counter method and the activated foil system. Films and chloroform-phase dosimeters were used for the gamma radiation measurements.

The proportional counter was used as the basic measuring device, as it had been most studied and was considered to be reasonably accurate. The foil system was checked against proportional counter results.

The studies were made for two reasons; first, the foil system was precalibrated against the proportional counter using Godiva as the source in preparation for use at Operation Teapot. Godiva was an ideal source for this purpose because of the neutron spectrum, ease of operation both

as a reactor and for prompt bursts, and because of the low gamma contaminant (if high, the gamma could negate proportional counter results); and, secondly, the results as found by the two systems were applicable to the animal runs made previously on Godiva and would allow an accurate calculation of effectiveness, since both the amount of energy deposited and the biological response were known.

The proportional counter method utilized the Bragg-Gray cavity theory, in that the collecting chamber was made of polyethylene filled with ethylene gas with walls thick enough so that there was secondary equilibrium established for the maximum path of an hydrogen nucleus scattered by a bombarding fast neutron. By utilizing a proportional system, gamma rays (if the photon/neutron ratio was not too great) could be biased out without a great loss of neutron sensitivity (~ 5 per cent). Also, the biasing system allowed pulse-height selection. Since an integral alpha source was used for calibration, the total energy from the collected pulses was determined, and using single collision theory for neutrons interacting with tissue, the energy deposition per gram of tissue (or rep) was determined.

The foil system utilized the principle of threshold detection to determine neutrons in a particular energy interval. The particular foils used were Au, Pu²³⁹,

Np^{237} , U^{238} (depleted in U^{235}), and S. Thermal neutron effect on Pu and Np and the low energy resonances in Pu were eliminated by shielding the foils with B^{10} . Thus, it was possible to produce a Pu response with a threshold of about 100 ev, a Np response at 0.75 Mev, and a U^{238} response at 1.5 Mev. Gold with and without Cd was used to determine the thermal flux, and S was used for neutrons above its threshold of 3 Mev. Utilizing the flux information in the various defined energy intervals, the rep was calculated from the single collision curve for tissue. This method was certainly practical for field use, since no elaborate set-ups were necessary and there was no dependence on rate of delivery of the flux and no saturation effect (as occurred in the proportional system). The proportional counter and the threshold detector system were in good agreement with regard to dose measurements in rep per fission from Godiva. There is still some disagreement between LASL measurements of rep per fission and those made by ORNL. Interpretation of these data and the determination of the true RBE of fission neutrons from Godiva will be made in early 1955.

16. Measurement of Associated Gamma Irradiation from Godiva (Harris, LASL; Lt. Sigoloff, USAF-SAM)

Using film badges and chloroform-phase dosimeters, attempts were made to measure the gamma ray

contamination associated with the neutron flux obtained from Godiva. These measurements indicated that the inherent gamma contamination is in agreement with theoretical calculations and amounts to approximately 5 per cent of the total rep radiation dose.

17. Relative Biological Effectiveness of 14-Mev Neutrons for the Production of Bone Marrow Damage (Harris, Rothermel, Schweitzer, Woodward)

The effect of 14-Mev neutrons on the uptake of Fe^{59} by the red blood cells of rats was determined. These data indicated that 1.4×10^8 14-Mev n./cm.² produced an effect equivalent to 1 r of 250-KVP X rays.

18. Relative Biological Effectiveness of Fission Neutrons from Godiva for Production of Bone Marrow Damage (Harris, Schweitzer, Rothermel, Woodward)

Rats were exposed to fission neutrons from Godiva and the effect of the neutron irradiation on uptake of Fe^{59} by the red blood cells of rats was determined. When compared with the effects produced by 1 r of 250-KVP X rays, the relative biological effectiveness of the fission neutrons was approximately 2. A more precise value for the RBE will be obtained when the data on physical dose measurements made by Hurst and Harris are interpreted.

19. Effect of Massive Rapid Doses of Fission Neutrons on the Behavior of Monkeys (Entire Radiobiology and Biophysics Sections)

Three animals were trained to a high performance level in a test situation in which the animals were required to climb up and down on a bar 1-1/2 feet off the floor of the exposure cage under the stimulus of shock avoidance. These monkeys were exposed to high doses of fission neutrons from the Godiva assembly, and their performance level checked immediately after, and 1, 4, 8, 12, 18, and 24 hours afterwards. The results of these studies were distributed in documentary form as LA-1838 (classified). For some additional discussion of these studies, see Sec. 1.1.3 (a).

20. The Relative Biological Effectiveness of Fission Fragments (Harris, Furchner, Ellinwood, Sanders, Strang, Schweitzer)

An attempt was made to study the relative biological effectiveness of fission fragments by determining the effect of plutonium fission inside the body on the rate of uptake of Fe^{59} by the red blood cells of rats. Four groups of rats were injected with equivalent activities of Pu^{238} and Pu^{239} . The first two groups were injected with Pu^{238} and the other two groups with Pu^{239} . One group that received Pu^{238} and one group that received Pu^{239} were exposed to a known

flux of thermal neutrons from the Los Alamos Homogeneous Reactor. The Fe^{59} uptake by the red blood cells of the animals in the four groups was determined after the neutron exposures. Since the thermal neutron fission cross-section for Pu^{238} is insignificant compared to that of Pu^{239} , a comparison of the difference in uptake of Fe^{59} by the various groups of animals should be an indication of the biological effect produced by the fissions of Pu^{239} inside the animal. The skeletons of the animals were analyzed after the blood samples were taken and the results normalized on the basis of Pu content of the skeleton. As of December 31, 1954, the results of these studies were not completely analyzed, but preliminary results indicated that the RBE of fission fragments under these conditions is low.

21. Studies of Cell Surface Adsorption of C^{14} -
Labeled Compounds (Rogers, Boone)

The adsorption of utilization of C^{14} -nicotinic acid by Lactobacillus arabinosus has been re-evaluated. With proper correction for self-adsorption and the elimination of the loss of cells by washing the cells with saline rather than distilled water, growing cells retain 80 to 85 per cent of the originally available nicotinic acid, at the end of 18 hours of growth. This is in the form

of Coenzyme I and II and is essentially all of the 15 to 20 per cent of the remaining nicotinic acid in the medium.

Cells grown in medium containing unlabeled nicotinic acid and then suspended in saline with C^{14} -labeled nicotinic acid have been found to require the presence of glucose to convert the nicotinic acid to Coenzyme I. Approximately 80 to 90 per cent of the C^{14} -nicotinic acid is adsorbed on the cell after 1 hour incubation at $37^{\circ}C$, if glucose is present.

22. The Pharmacology of Quaternary Salts of Oxazoles (Sanders, Ott, Hayes, Lushbaugh)

The methyl p-toluenesulfonate (methyl tosylate) salts of the following compounds were synthesized by the Organic Chemistry Section and tested for the ability to suppress the thermo-regulatory mechanism in mice.

2-(1-naphthyl)-5-	Phenyloxazole
2-(2-naphthyl)-5-	"
2-(2-chlorophenyl)-5-	"
2-(3,4-dichlorophenyl)-5-	"
2-(3-methoxyphenyl)-5-	"
2-(4-methoxyphenyl)-5-	"
2-(3,4-methylenedioxyphenyl)-5-	"
2-(4-tolyl)-5-	"
2-furyl-5-	"
2-thienyl-5-	"
2-cyclohexyl-5-	"
2-(4-biphenyl)-5-	"
2-(ω -styryl)-5-	"
2,5-di(1-naphthyl)-oxazole	
2-phenylbenzoxazole	
1,4-di[2-(5-phenyloxazolyl)] benzene	
2,5-diphenylthiazole	

Groups of mice were injected intraperitoneally with various concentrations of water solutions of the oxazole quaternary salts. Body temperatures were measured to note both the duration and magnitude of the inactivation thermo-regulatory system of the animals. Three environmental temperature conditions were employed, namely, room temperature, 8 to 12°, and 36°. The tosylate salt 2-(1-naphthyl)-5-phenyloxazole has given the most interesting results. Table 2.2.1 gives a typical set of data showing the effect of a dose of 1 mg./10 g. body weight on the body temperature of mice as a function of time after injection.

Table 2.2.1 - EFFECT OF TOSYLATE SALT OF 2-(1-NAPHTHYL)-5-PHENYLOXAZOLE ON THE BODY TEMPERATURE OF MICE

Time after injection (min.)	Body Temperature (°C)
0	37
20	32
40	28.5
60	26
100	25
200	24
300	25
400	29
500	31.5
600	33

23. Calibration Studies for Participation in
Operation Teapot, 1955 (Entire Section)

Initial experiments were conducted on the response of the NBS film packet to neutrons from Godiva. A single series of exposures were made. In this series, the film packets were exposed in three different assemblies: (a) the bare packet; (b) the packet surrounded by 6-inch lead; and (c) the packet surrounded by 1/8-inch lithium metal. There was no difference between the bare packet and the lithium-shielded packet, which indicated that, as could be predicted from foil results, the number of thermal neutrons emitted from Godiva was not enough to increase the density of the film. However, the results of the packet surrounded by lead, as compared with the bare packet, indicated a reduction in roentgen equivalent reading by a factor of 3. This result could have been due to (a) reduction in gamma exposure by prompt fission gammas; (b) reduction in gamma exposure from residual fission gammas; or (c) change in the neutron sensitivity of the film due to the interposition of the lead shield.

If it were assumed that the lead eliminated all gamma photons and that neutron processes in the lead did not appreciably influence the film response to neutrons, a first approximation indicated that about 6×10^9 n./cm.² caused a density change in film equivalent to 1 r. These

studies were continued in order to determine the film response more accurately and account for the time function of the film response to prompt and delayed gamma rays.

24. Pre-Test Biological Calibrations for Operation Teapot, 1955 (Effect of Cold Stress) (Furchner, Schweitzer)

A comparison of the effect of radiation alone and radiation preceded by cold stress on the weight changes in the spleens and thymuses of CF_1 mice was made, in order to establish the possibility of eliminating expensive heating of exposure containers during Operation Teapot. The mice were placed in cages and exposed to cold in an aluminum test exposure device overnight. After exposure to cold, the mice were exposed to X irradiation. The weight changes in the spleens and thymuses of these mice were compared with relevant weight changes in mice exposed to X irradiation only. Changes in body weight were also compared. These results suggested that it would be feasible to eliminate heating elements from the exposure containers during the field operations.

2.3 Radiopathology Section (C. C. Lushbaugh, Section Leader)

2.3.1 General Comments on Section Activities

The activities of the Radiopathology Section during 1954 may be summarized under the following general categories:

- a. Research on the morphological changes following high doses of gamma radiation in rats and monkeys.
- b. Research on the cytological effects of radiation, particularly upon the dividing nucleus.
- c. Research on biochemical and enzymatic changes possibly related to the fundamental nature of "radiation damage."
- d. Research on non-mammalian, simple biologic systems that might be applied to the assessment of relative biological effectiveness of various kinds of ionizing radiation.
- e. The provision of service to other sections and groups requiring pathological interpretations and preparation of experimentally and clinically obtained materials.

2.3.2 Publications, Documents, and Reports Originating in the Section

- (1) Comparative Study of Experimentally Produced Beta Ray Lesions and Other Skin Lesions in Utah Range Sheep,

J. F. Spalding, C. C. Lushbaugh, presented at the meeting of the Physiological Section of the American Society of Animal Production, November 1954, in Chicago, Illinois.

(2) Effects of Massive Doses of High Dose Rate Gamma Radiation on Monkeys, Phase V, Chapter 5, Pathology, C. C. Lushbaugh, Carl Hoak, USAF, in Phase V Report by Langham et al., in manuscript as LA-document.

(3) The Pathology of Monkeys Exposed to Massive Doses of Total Body Gamma Radiation, C. C. Lushbaugh, Carl Hoak, Federation Proc., 14, No. 1, 411-412; Part II, 495 (1955).

(4) Radiopathology of Islets of Langerhans in Rats, J. F. Spalding, C. C. Lushbaugh, Federation Proc., 14, 420; Part II, 495 (1955).

2.3.3 Progress of Specific Projects

1. Gross and Microscopic Pathology of Dermal Lesions of Nevada Range Sheep, as Compared with Those Produced Experimentally with a Sr⁹⁰-Y⁹⁰ Applicator (Lushbaugh, Spalding, Wellnitz, Hale)

All experimental work on this project was terminated after the progression of the post-irradiation scarring was documented and studied by obtaining sequential biopsies of the wounds up to 12 months after inception. The conclusion that dermal radiation damage and the skin disease of the range sheep are dissimilar remained valid.

2. The Effect of Lethal Doses of X Rays on the
Maturation of the Rat and its Modification by Gonadotropism
(Spalding, Wellnitz)

In line with other morphologic studies in the Radiopathology Section, the results showed that many of the ovarian changes post-irradiation are not primarily from ovarian radiation damage but are secondary to changes in other organs, in this case the pituitary. It also showed for the first time that ova in primary follicles are relatively radioresistant in comparison with ova in other situations.

3. The Pathology of Monkeys Exposed to Massive
Doses of Total Body Gamma Radiation (Lushbaugh, Hoak,
Wellnitz, Hale)

See Sec. 1.1.6.

4. The Fava Bean Root as a Test System for
Studying Biological Effectiveness of Ionizing Radiations
(Spalding, Langham, Anderson)

The growth retarding effects of X irradiation in air and beta particle exposure from absorption of tritium as HTO was measured under carefully controlled environmental conditions. The new scintillating mediums enabled an actual determination of the internal dosimetry that occurred with this type of beta particle exposure. The resulting LD₅₀

figures indicated that the RBE of the beta particle was only about 1.5 times that of X rays. The ease with which this system can be analyzed dosimetrically gives hope that the RBE of the alpha particle can be similarly re-evaluated and "permissible tolerances" thereby given a new factual basis.

5. Histochemical Changes following Radiation Damage (Wellnitz, Lushbaugh)

This study is still in its infancy and has not progressed beyond that previously reported concerning alkaline phosphatase, lipin, and esterase in irradiated skin. Time was used this year in a 2-week study of techniques by Mrs. Wellnitz in Dr. G. Gomori's laboratory at the University of Chicago, and in obtaining a more efficient freezing-dehydrating technique and apparatus. Techniques are being worked out for determining the locus of activity of cathepsins and ATPase.

6. Determination of the Mitotic Dynamics of the Fava Bean Root (Hale, Spalding, Lushbaugh)

The method of Knowlton and Widner for determining mitotic index of mammalian tissues in vivo with X irradiation was applied to the bean root. So far with exposures of 400 and 500 r, the method appears applicable and indicates that a root mitosis spends about 30 minutes

dividing (visibly). The prolonged depression in mitotic rate after these doses indicates that the continued post-irradiation growth of the bean root is due to the process of cellular elongation which apparently takes several days to cease.

7. Determination of the Mitotic Dynamics of Normal and Malignant Tissues after Death of the Host or after Their Removal from the Body (Hale, Lushbaugh)

Following death, the mitotic rate declines progressively in normal mouse skin apparently as the result of (a) failure of cells to begin prophase (early prophase arrest), and (b) proteolytic enzyme activity evidenced by pyknotic damage to the metaphase chromosomes and spindle. In the neoplasia studied so far (2 rats and 4 humans), prophase arrest does not occur and pyknosis (or proteolytic freeing of nucleic acid) is not significantly increased up to 6 hours after death, even with incubation at 37°. An experiment was begun, but is not completed, to study any alterations imposed upon these phenomena by in vitro X irradiation.

8. The Effect of Ionizing Radiations upon Achromobacter fischeri (Hughes, Lushbaugh)

Much experience was obtained in handling,

culturing, and storing this luminescent bacterium. A method was developed for measuring the light output by growing and stable cultures. An approximate LD₅₀ for the bacterium was obtained on the basis of how much radiation was required to extinguish 50 per cent of the light. This number was not reproducible in subsequent experiments. In investigating why it was not, it was found extremely easy to vary the population sensitivity by repeated exposures to X irradiation, producing apparent resistant strains, one of which was immune to 10,000 r. These observations are being extended.

9. Changes in *in vivo* Enzymatic Systems Associated with Growth and Mitotic Division Post-Irradiation (Hughes, Lushbaugh)

The previous study of profound alterations in serum alkaline phosphatase and sulphhydryl levels after total body irradiation was extended to adenosinetriphosphatase in the serum and spleen of rats. Spleen ATPase activity was found to vary inversely with spleen weight, so that post-irradiation atrophy gave rise to an apparent increase in ATPase activity. An attempt is now being made to study this phenomenon histochemically since the constancy of total spleen ATPase activity does not account for or explain the concomitant fall in this enzyme in the serum. Since

serum phosphates are rate-modifiers for proteolytic enzyme systems, it was quite logical to find that serum proteolytic enzyme inhibitors are profoundly disturbed following total body X irradiation. This disturbance in serum proteinases and their inhibitors is being studied further.

2.4 Biophysics Section (E. C. Anderson, Section Leader)

2.4.1 General Comments on Section Activities

The activities of the Biophysics Section during 1954 may be summarized under the following general categories:

- a. The design of a liquid scintillation counter for the measurement of the natural radioactivity of human beings.
- b. The development of physical dosimetry techniques.
- c. Theoretical and experimental evaluation of the hazards associated with acute exposure to high concentrations of tritium gas.
- d. Assistance to other sections with instrumentation, dose measurements, radiation exposures, and theoretical calculations and considerations.
- e. Preparation for participation in Operation Teapot during 1955.

2.4.2 Publications, Documents and Reports Originating
in the Section

(1) K-9: A Large 4π Gamma Ray Detector, M. A. Van Dilla, E. C. Anderson, R. L. Schuch, Nucleonics, 12, No. 9, 22 (1954).

(2) A Theoretical Consideration of the Hazards Associated with Acute Exposure to High Concentrations of Tritium Gas, E. C. Anderson, W. H. Langham, LA-1646, April 1954.

(3) Gamma Ray Spectrometry, E. C. Anderson, RCL Counter, 1, No. 2 (1954).

2.4.3 Progress of Specific Projects

1. Construction and Study of Large Volume
Scintillation Detectors for Biological Use (Anderson,
Perrings, with M. A. Van Dilla, University of Utah)

(a) Human Counter. See Sec. 1.1.5.

(b) K-10. An improved version of the dog counter (K-9) built for the University of Utah has been designed. It is hoped that a counter capable of detecting effectively the Bremsstrahlung from nuclides such as Sr^{90} can be developed. Since this will involve looking at pulses of 100 kev or less, light collection efficiency is very important. The design being worked on is an all-lucite tank, made toluene-resistant by a coating of Epon 1001 resin.

It is hoped that total internal reflection will trap an appreciable fraction of the light with high efficiency and that additional reflectors outside the system will return some of that escaping. The transparent system will also make possible a study of the effect of phototube placement.

(c) Monkey Counter. This is the old El Puercoespín, built as a pilot model for the neutrino program, now modified with a thin wall stainless steel insert to convert it to a well-type system. Shielded with 2 inches of Pb and installed in the H-5 counting room in the basement of HRL, the background is 120 c/s with a K^{40} counting efficiency of 15 per cent. It will be used to study the metabolism of fission products in the monkey.

2. Eye Counter and Study of Turnover Rates in the Anterior Chamber of the Rabbit's Eye (Anderson, with Rothermel and Woodward)

A scintillation counter for the measurement of turnover times of beta and gamma emitters in the eye has been developed, using a small anthracene crystal. The loss of Na^{24} from the anterior chamber of the rabbit's eye has been studied and a half-time of about 80 hours measured. This value can be decreased to about 50 minutes by administering pilocarpine. The instrument appears to be too insensitive to permit the injection of the activity into the

rabbit's circulatory system and the measurement of the rate of appearance of activity in the eye. The dilution factor involved necessitates the use of excessively large amounts of radioactivity.

3. Preparation for Participation in Operation
Teapot, 1955 (Anderson, Sayeg, Larkins, Worman)

A large amount of the Section's effort during November and December was directed toward cooperation with Dr. H. H. Rossi of Columbia University, in preparation for tissue-equivalent ion chamber measurements during the 1955 weapons test series. The tissue-equivalent chambers designed by Rossi and constructed by Frank Shonka of Chicago were calibrated and studied using 250-KVP X rays.

A charger-reader for the chambers was designed and constructed by Dick Hiebert of P-1. This equipment was checked and used for the chamber studies and proved to be an excellent piece of electronics.

4. Tissue-Equivalent Ion Chambers (Sayeg,
J. Larkins)

The study of the use of tissue-equivalent materials for the measurement of radiation dose to biological systems in rep (rad) has been accelerated by the assignment of a new Staff Member to work full-time on this problem.

Several chambers have been built and tested with various gases, in order to establish the validity of the Bragg-Gray principle for the systems used.

5. Boron Analysis of Biological Materials using a Sigma Pile (J. Larkins)

Several systems of detecting the alpha particles emitted by small amounts of boron under neutron bombardment in a small graphite pile, using a Po-Be neutron source, have been investigated. Both proportional counters and pulse ion chambers failed to discriminate sufficiently against the high background. A thin ZnS screen and photomultiplier tube look very promising, but the problem is as yet unsolved.

6. Theoretical Consideration of the Hazards Associated with Acute Exposure to High Concentrations of Tritium Gas (Anderson, Langham)

Theoretical consideration of the hazards associated with acute exposure to high concentrations of T_2 gas indicates that a lethal radiation dose would result only from explosive oxidation of very rich T_2 -air mixtures. If the gas remains as T_2 , the dose rate to a 1-micron layer of the respiratory surfaces would be very high (thousands of rep per second), but the effect of doses of this magnitude

on a very thin layer of these surfaces is completely unknown. Other hazards considered were whole-body irradiation from dissolved T_2 in body fluids; biological oxidation of T_2 and subsequent absorption of the tritium water produced; auto-oxidation of T_2 by the tritium beta rays and absorption of the tritium water; and Bremsstrahlung radiation of tissue surfaces. The orders of magnitude of these hazards are a few tenths of rep per second of exposure.

7. Experimental Studies of the Hazards Associated with Inhalation of High Concentrations of Tritium Gas (Trujillo, Anderson, Langham)

In order to study the direct effect of tritium beta irradiation on a 1-micron layer of respiratory surfaces, mice were exposed to inhalation of high concentrations of T_2 . The animals were placed in a closed all-glass system and were supplied oxygen on demand through a sensitive demand valve from a respirometer. After the animals were maintained in the closed system for a few minutes, enough pure T_2 was added to the cage to provide a 1 per cent or 10 per cent tritium concentration in the gas volume of the cage. The animals were allowed to breathe these tritium concentrations for periods of time ranging from a few minutes to 3 hours. The animals were removed from the cage and observed for signs of radiation damage. Urine samples were collected and

analyzed for tritium, to determine the amount of tritium converted to tritium water by auto-oxidation and biological oxidation and absorbed by the animals.

In all experiments, appreciable concentrations of tritium water were found in the body fluids of the animals, and whole-body irradiation in the body fluids was the limiting factor in survival. The LD_{50}^{30} concentration of tritium water in the body fluids of the animals was found to be 1.58 mc./cc., regardless of whether the tritium water was obtained through inhalation or by intraperitoneal injection. These experiments are being continued in order to evaluate the various hazards associated with inhalation of high concentrations of tritium gas.

2.5 Organic Chemistry Section (Wright H. Langham, Acting Section Leader)

2.5.1 General Comments on Section Activities

The general activities of the Organic Chemistry Section in 1954 were as follows:

- a. Syntheses of labeled compounds.
- b. Preparation of manuscript for the book "Organic Syntheses with Isotopes."
- c. Preparation and testing of new scintillation solutes and solvents.
- d. Development of liquid scintillation detectors

for low energy, high sensitivity, multiple isotope, and 4π counting.

e. Syntheses of oxazolium salts as drugs for inhibiting body temperature control.

An exhibit entitled "Organic Scintillation Detectors" was presented at the Eighth National Chemical Exposition of the American Chemical Society on October 11-16, in Chicago, Illinois.

2.5.2 Publications, Documents and Reports Originating in the Section

(1) Liquid Solution Scintillators, F. N. Hayes, Betty S. Rogers, LA-1639, May 1954.

(2) Photomultiplier Testing for Low Energy Counters, F. N. Hayes, LA-1728, September 1954.

(3) Argon Treatment of Liquid Scintillators to Eliminate Oxygen Quenching, F. N. Hayes, D. G. Ott, Jay E. Hamme, John F. Kephart, LA-1837, October 1954.

(4) Photovoltaic Scintillation Detector for High Dose Rates, R. L. Schuch, R. D. Hiebert, F. N. Hayes, Nucleonics, 12, No. 2, 16 (1954).

(5) 2,5-Diaryloxazoles and 2,5-Diaryl-1,3,4-Oxadiazoles, F. N. Hayes, Betty S. Rogers, D. G. Ott, J. Am. Chem. Soc., 77, 1850 (1955).

(6) Photocathode and Reflector Effects on Relative Pulse Height Measurements, F. N. Hayes, Betty S. Rogers, submitted for publication to Nucleonics.

2.5.3 Progress of Specific Projects

1. Preparation of Manuscript on the Subject of "Organic Syntheses with Isotopes" (Williams, Murray)

A major effort during 1954 was the preparation of a manuscript that will make a major publication, concerning the syntheses of organic compounds labeled with isotopes. In this manuscript, an attempt has been made to include all the syntheses of isotopically labeled organic compounds appearing in the literature through 1953. In addition, information concerning isotopic work, techniques, and major references have been included which might be of value to any program involving isotopic syntheses, e.g., mechanism studies, rate studies, and exchange reaction investigations. This compilation, over 2,000 pages, was prepared primarily to summarize existing literature on the subject for the benefit of the Group H-4 isotopic labeling program and it already has been of considerable value to the Organic Chemistry Section.

It is hoped that the manuscript will be published by a commercial publisher. If arrangements with a commercial publisher cannot be made by June 1955, attempts will be made to have it published by the Government Printing Office, or distributed as an LA-document.

2. Synthesis of Acetic-1-C¹⁴ Acid (Williams)

A 20-mc. quantity of acetic-1-C¹⁴ acid was prepared from barium carbonate-C¹⁴ via the Grignard reaction with methyl magnesium iodide.

3. Preparation of an Absolute C¹⁴ Standard for Liquid Scintillation Counters (Williams)

For the determination of efficiency in the scintillation method of counting disintegrations an absolute standard is essential. Since it has been shown that C¹⁴ dioxide is not counted very satisfactorily when dissolved in the liquid scintillator solution, an organic compound which is soluble in toluene and is easily prepared from carbon dioxide, in high yield, is desirable. Benzenecarboxylic-C¹⁴ acid (benzoic acid) for this purpose from C¹⁴ dioxide via the Grignard reaction has been prepared. The isotopic ratio of the C¹⁴ dioxide was determined on a Consolidated-Neir Model 201 Mass Spectrometer, using half the total sample.

Since only one sample of the C¹⁴ dioxide was assayed with a single mass spectrometer, it appeared desirable to have a second sample of C¹⁴ dioxide assayed in several laboratories. Consequently, a quantity of C¹⁴ dioxide (approximately 5 per cent isotope) was generated from barium carbonate-C¹⁴ and divided into four equal portions. One of these portions was reserved for preparation of the benzoic

acid standard. The other three portions were analyzed mass spectrometrically in different laboratories with the results shown in Table 2.5.1.

Table 2.5.1 - STANDARDIZATION OF C^{14} DIOXIDE

Laboratory	Instrument	Atom per cent C^{14} Isotope	Number of Assays
National Bureau of Standards	Consolidated-Neir Model 21-103	5.97 ± 0.04	20
GMX-2 Los Alamos	Consolidated-Neir Model 21-103	5.92 ± 0.01	24
CMR-9 Los Alamos	Consolidated-Neir Model 201	5.88 ± 0.06	--

Standard C^{14} -benzoic acid will be prepared from the remainder of each of the samples analyzed, as well as the portion reserved for that purpose. This procedure will afford a more complete check on the standard.

4. Micro-Analytical Laboratory (Williams)

A micro-analytical laboratory was set up for the analysis of organic compounds for carbon, hydrogen and nitrogen content. The Dumas-type nitrogen apparatus has given excellent results. The carbon-hydrogen apparatus gave good results on compounds not containing nitrogen, in preliminary runs. An absorption tube for the elimination of

oxides of nitrogen is yet to be added to the train, so that compounds containing nitrogen can be analyzed for carbon and hydrogen.

5. Scintillation Solutes: Oxazoles and 1,3,4-Oxadiazoles (Hayes, Ott, Rogers, Kerr)

Additional syntheses in the 2,5-diaryloxazole series have brought the number of oxazoles prepared in this laboratory to 48, most of which were previously unreported. Likewise, the total number of 1,3,4-oxadiazoles is now 24. These further syntheses in established series have greatly added to existing information on the influence of substituent groups on scintillation efficiency in the polyaryl series.

This project is being continued with special emphasis on polyoxazoles and on compounds with various degrees of steric hindrance interfering with continuous resonance in the molecule.

6. Scintillation Solutes: Representative Polyaryls (Hayes, Ott, Rogers, Kerr)

Several new series of heterocyclics have been added to the list of potential scintillation solutes. These involve the thiophene, pyrrole, pyrazine, pyridazine, imidazole and indole ring systems in combination with benzene rings to give molecules wherein all the rings join to give a continuous resonance system.

In its future phases, this project will include poly-condensed aromatics such as diphenylnaphthalenes and phenylphenanthrenes.

7. Syntheses of Oxazolium Salts (Ott, Hayes)

Twenty new oxazole quaternary salts were synthesized for pharmacological investigation. Oxazoles such as those derived from the scintillation solute program reacted readily with methyl tosylate to give the 3-methyl-oxazolium tosylates. A metathetical reaction sequence allowed these to be converted to perchlorate, iodide, and then chloride salts. The tosylate salts could be converted directly into chloride salts by a simple anion exchange resin procedure.

The salts were shown to be quite stable in neutral or acid solution, but were readily hydrolyzed by bases even as weak as dilute aqueous ammonia.

Further work on these compounds will involve ultraviolet and infrared absorption spectra, as well as mercury arc-excited fluorescence spectra.

8. Spectroscopy of Organic Scintillators (Ott, Hayes)

The ultraviolet absorption spectra of scintillation solutes in cyclohexane solution have been

obtained for 40 representative compounds.

A Hilger Intermediate Quartz Spectrograph has been outfitted to operate either with mercury arc excitation or with radioactive source excitation, using 100 mc. of Cs^{137} .

The nature of the spectra will be studied, not only as related to the emitting solutes, but also as to mode of excitation, concentration of solute, and light path in the solution. Scintillation spectra will be compared with absorption spectra and a significant contribution to theory is expected.

9. Relative Pulse Heights of Organic Scintillators

(Hayes, Ott, Rogers, Kerr)

Relative pulse heights as a function of concentration in toluene have been obtained for all the scintillation solutes. A photomultiplier tube with an S-4 type cathode and an aluminum reflector were used throughout. Some curves were also obtained with a titanium dioxide reflector. Each solute at 3 g./l. or at its maximum concentration, if less, had its ratio of relative pulse height run as derived from titanium dioxide and aluminum reflectors. This TiO_2/Al value was shown to be related to the wavelength distribution of the scintillation spectrum.

Procedures for best pulse height evaluation of secondary solutes have been found and such measurements will occupy a large part of the future of this project.

10. Liquid Scintillation Counting (Hayes)

The potentialities of liquid scintillation counting have been explored in relation to the great number of instrumental variables offered in the Los Alamos Model 530 Coincidence System. A method of operation has emerged which involves best use of the little structure available in a beta spectrum. The discriminators are set relative to the pulse spectrum such that the highest and lowest parts are excluded. An optimum counting efficiency can be found by varying high voltage or amplifier gain. This peak position contains elements of stability which are highly desirable in this complicated counter.

Na^{22} , C^{14} , and H^3 counting have been studied, singly and in mixtures. Counting efficiencies over 50 per cent are unavailable only for H^3 , with its very low energy spectrum. The energy dependence of the liquid scintillator allows accurate analysis of mixtures of the above three isotopes.

Routine counting of tritium-containing urine samples with efficiencies of 8 to 10 per cent and very little quenching interference has been successfully accomplished.

The necessity for extreme refrigeration ($< 0^{\circ}\text{C}$) has been shown to be nonexistent where reasonably good photomultipliers may be selected. Experience has been obtained in counting at room temperature which has shown that a refrigerator is needed only when the highest possible efficiencies in tritium counting must be realized. Thermionic emission is a serious problem here.

Accurate standards for Na^{22} , C^{14} , and HTO have been obtained. Future work in this line will be directed toward the accurate tritium standard in a toluene-soluble form.

New ideas in electronic circuits will be explored with featuring of doubly-clipped and slightly slower amplifiers and discriminators with much less power dissipation.

11. Quenching of Liquid Scintillators (Hayes, Ott)

Quenching by various functional groups in organic compounds has been noted in an earlier solvent study. A program to explore this field more thoroughly was initiated by studying the effect of various anions with acid salts of tertiary amines. A notable absence of quenching for chloride and bromide ions, whereas these atoms cause considerable trouble when covalently bonded in organic compounds, allowed optimism for a general ability to engage in liquid scintillation counting in the presence of inorganic salts.

Dissolved oxygen quenching at Los Alamos versus that at sea level has been investigated and a reliable method for its removal has been found. This method, bubbling with argon, is not practical when many samples per day must be run, but may be extremely useful in large detectors whose filling remains the same over long periods of time.

A very inclusive program is being started in which the goal is to establish the quenching characteristics of all types of compounds which may be encountered in the use of liquid scintillation methods.

12. Large Volume Liquid Scintillators (Hayes, Ott)

Work is continuing in collaboration with other sections and groups to improve on and supply liquid scintillator fillings.

A detector with 50-l. capacity will be used in running some relative pulse heights to show how to extrapolate small detector measurements to give applicable values for giant detectors.

13. Supernatural C¹⁴ (Ott, Hayes)

Successful development of a method for converting camphor into p-cymene will allow contemporary natural C¹⁴ values to be measured with great accuracy and related to specific recent years. Formosa camphor samples harvested in

1948 and in 1954 have been obtained. An attempt is being made to locate a sample which is many years older. A large quantity of "dead" p-cymene has been prepared for use in the background measurements.

3. Proposed Program for 1955

As in the past, the programmatic effort of the Biomedical Research Group (H-4) will be determined by the needs and interests of the Los Alamos Scientific Laboratory, the weapons program, and the AEC Division of Biology and Medicine.

Those projects, both basic and programmatic, that are being continued from 1954 and those anticipated in the near future are listed by title under the Section headings to provide a general outline of the Biomedical Research program for 1955. Obviously, it is not possible to anticipate all problems that will constitute the 1955 program, and it is not possible to state which of the projects listed below will be pursued to completion or dropped because of loss of interest during the coming year.

3.1 Biochemistry Section (Gordon Gould, Section Leader)

a. Effect of X Irradiation on Protein Synthesis in Intestine, Liver, Spleen and Other Tissues, and on Individual Plasma Protein Levels and Turnover Rates (Gould, Lotz, Magee).

b. Investigation of the Binding of Pu, Sr⁸⁹, and Other Fission Products with Serum Proteins using Filter Paper Electrophoresis (Magee, Foreman, Gould).

c. Continued Studies on Absorption and Metabolic Effects of Dihydrocholesterol and β -Sitosterol (Gould, Lotz, Lilly).

d. Determination of the Rates of Oxidation of C^{14} - β -Hydroxybutyric Acid in Tissues of Normal and Diabetic Animals (Gould, Lotz, Lilly).

e. A Continuation of the General Investigation of Factors Controlling Cholesterol Synthesis, Its Distribution between Liver and Plasma, and the Mechanisms Regulating the Plasma Cholesterol Level, including the Effects of Tri-iodothyronine, etc., on Hepatic Synthesis in vivo and in vitro; Cortisone, Estrogens; Replacing Plasma by Delipidized Plasma from the Same Species; and Blocking or Destroying the R. E. Cells (Gould, Lilly, Lotz).

f. Comparison of the Metabolism of Pu, Sr, and Ca (Foreman, Finnegan).

g. Metabolic Studies with Sr, including Attempts to Alter Tubular Resorption, and Investigation of the Excretion of Sr in the Bile and via the Gut (Foreman, Finnegan).

h. Further Investigation of the Effect of Permakleers in Excretion of Metals (Foreman, Finnegan).

i. Assay of Necropsy Specimens for Pu, U, and Be (Foreman, Milligan).

j. Investigations of a Pulmonary Function Test using Radioactive Krypton (Foreman, Finnegan).

k. Acute Toxicity Studies of Special Materials of Interest to LASL (Sabine, Miller).

1. The Clinical Significance of Cholinesterase Titers
(Sabine, Miller).

3.2 Radiobiology Section (Payne S. Harris, Section Leader)

a. Participation in 1955 Weapons Test Series, Operation
Teapot (Entire Section).

b. Continued Studies of the RBE of Fission Neutrons
using Godiva (Harris, Furchner).

c. Further Calibration of Godiva as a Standard Source
of Fission Neutrons for Biological Investigations (Harris,
Anderson, Sayeg).

d. Effect of Dose Rate on the Biological Effectiveness
of Fission Neutrons from Godiva (Harris, Furchner, Sanders).

e. Additivity of Fission Neutrons and X or Gamma Radia-
tion (Furchner, Harris, Schweitzer, Sanders, Storer).

f. Determination of RBE of Fission, Thermal and 14-Mev
Neutrons using Effect of Radiation on Incidence of AK Leukemia
in Various Strains of Mice (Boone, Rogers, Harris).

g. RBE of Fission Fragments (Harris, Furchner, Sanders,
Schweitzer).

h. Studies of Cell Surface Adsorption of C^{14} -Labeled
Compounds (Boone, Rogers).

i. Uptake of C^{14} -Labeled Isoniazid by Mycobacterium
(Boone, Rogers).

j. Effect of Radioactive Particles in the Lungs of Mammals (Boone, Rogers, Shultz (LAMC), Langham).

k. Metabolism of Fission Products and Other Gamma Emitters in Monkeys (Woodward, Rothermel, Johnson, Langham).

l. Follow-up Examinations of Los Alamos Plutonium Exposure Cases (Langham, Shipman, Hempelmann (University of Rochester)).

3.3 Radiopathology Section (C. C. Lushbaugh, Section Leader)

a. Pathogenesis of the Post-Irradiation Loss of the Mucosa of the Intestinal Villus (Lushbaugh, Wellnitz).

b. Cause of Rapid Death from Massive Amounts of Rapidly Delivered Radiation (Lushbaugh, Wellnitz).

c. Possibility of Using Enzymatic Debridement to Hasten the Development and Healing of Dermal Wounds caused by Beta Irradiation (Lushbaugh, Spalding, Hale).

d. Investigation of Proteolytic Enzymes Being the Active Healing Principle in Aloe vera Leaves (Lushbaugh, Hughes).

e. The Effect of X Irradiation upon the Quantity of Proteolytic Enzyme Inhibitor in the Serum of Rats as a Function of Time after Irradiation and Time of Death (Lushbaugh, Hughes).

f. Histochemical Determination of the Locus of ATPase Action in the Irradiated and Non-Irradiated Spleen (Wellnitz, Lushbaugh).

g. Histochemical Determination of the Locus of Proteolytic Enzyme Action in Normal, Irradiated and Neoplastic Tissues (Wellnitz, Lushbaugh).

h. Extension of the Comparison of the Effectiveness of X Irradiation and Beta Particle Exposure on the Growth of the Fava Bean Root to include a Comparison of the Similar Effectiveness of Alpha Particles and Neutrons (Spalding, Langham).

i. Cytology of the Mitotic Dynamics of the Fava Bean Root after X Irradiation (Hale, Lushbaugh).

j. Cytological Changes attending Death of Normal, Neoplastic, and Irradiated Tissues (Hale, Lushbaugh).

k. The Applicability of Achromobacter fischeri and its Luminescent System of Light Production to Radiobiological Problems and in Particular to the Problem of Assessing Radiobiologic Effectiveness (Hughes, Lushbaugh).

l. Study of the Effect of Tritium Beta Particles upon the Pulmonary Alveolar Wall, and Post-Irradiation and Post-Mortem Changes in Mammalian Chromosomes (Wellnitz, Kalmus, Lushbaugh).

3.4 Biophysics Section (E. C. Anderson, Section Leader)

a. Assembly, Testing and Application of the Human Counter (Anderson, Perrings, J. Larkins, Worman).

b. Physical Dosimetry of Neutrons and Correlation of Dose with Biological Effects (Sayeg, Larkins, Anderson).

c. Physical Dosimetry of Neutrons from Atomic Detonations during Operation Teapot (Sayet, Anderson, Larkins, Perrings).

d. Dose and Effect of Beta Radiation to Respiratory Surfaces as a Result of Inhalation of High Concentrations of Tritium Gas (Anderson, Trujillo, Langham).

e. Analysis of Boron in Biological Materials using the Sigma Pile (Larkins, Perrings, Anderson).

f. RBE of the Beta Particles from Na^{24} .

g. Further Studies of the Characteristics and Biophysical Applications of Large Volume Liquid Scintillation Detectors.

h. Theoretical Instrumentation and Radiation Services and Assistance to Other Sections (All members of the Section).

3.5 Organic Chemistry Section (Wright H. Langham, Acting Section Leader)

a. Syntheses of Labeled Compounds.

(1) Preparation of N-methyl- C^{14} -di(2-chloroethyl)-amine (Nitrogen Mustard).

(2) Preparation of Isonicotinic Acid- C^{14} Hydrazide (Isoniazid).

(3) Purification of 2,4,6-Triethylenimino-1,3,5-triazine-2,4,6- C_3^{14} (TEM).

(4) Preparation of Acetic Acid- C^{14} .

(5) Preparation of Benzenecarboxylic Acid- C^{14} Standard.

(6) Preparation of N, N', N''-Triethylene-1-C¹⁴-thiophosphoramidate and N, N', N''-Triethylene-1-t-thiophosphoramidate (Thio TEPA).

(7) 1,3-Dichloro-2-isopropyl-N-diethylcarbamate-C¹⁴ (Compound 738).

(8) Preparation of 5,5-Diphenylhydantoin-4-C¹⁴ (Dilantin).

(9) Preparation of 3-Hydroxybutyric Acid-1-C¹⁴.

(10) Preparation of 3-Methyl-2-butenic Acid-1-C¹⁴.

(11) Preparation of Pyridoxin-C¹⁴ (Vitamin B₆).

(12) Preparation of 10-[1-(3-Dimethylaminopropyl-3-C¹⁴)]-2-chlorophenothiazine Hydrochloride (Thorazine).

(13) Preparation of a Tritium Standard for Liquid Scintillation Counters.

(14) Preparation of Tritium-Labeled Steroids.

b. Syntheses of New Organic Scintillation Solutes.

c. Spectroscopy of Organic Scintillators.

d. Relative Pulse Heights of Organic Scintillators.

e. Quenching of Liquid Scintillators.

f. Liquid Scintillation Counting.

g. Supernatural C¹⁴.

h. Fundamental Properties of Scintillator Solutions in Large Detectors.

- i. Isolation and Characterization of Metabolites of Labeled Compounds from Urine of Experimental Animals.
- j. Tracer Studies of Organic Reactions.
- k. Preparation of Final Manuscript on the Subject of "Organic Syntheses with Isotopes."