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THE ACUTE RADIATION SYNDROME

A Study of Ten Cases and A Review of the Problem

VOLUME II

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HEALTH AND BIOLOGY

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INTRODUCTION

(To Volume I)

"Acute radiation syndrome" is the name applied to the group of signs and symptoms that characterize the injury produced by exposure of the entire body or a large part thereof to ionizing radiation. This is an acute generalized disease, lasting only a matter of weeks. The syndrome differs in this respect from the chronic form of radiation injury caused by repeated or prolonged exposure of the body to radiation. Such injuries manifest themselves as gradually developing, lingering illnesses. The acute radiation syndrome is also distinct from the most usual type of radiation injury, i.e., that produced by irradiation of a limited part of the body. These localized radiation injuries may be either acute or chronic in nature. Erythema and x-ray burns of the skin are a well-known example of the acute localized reaction, while the atrophy, hyperkeratoses and epitheliomata of the skin are characteristic forms of chronic localized radiation injuries.

In this report, ten cases of the acute radiation syndrome are presented. These cases occurred several years ago at the Los Alamos Scientific Laboratory as a result of two accidents of a unique nature involving fissionable material. Because of the limited number of similar cases of acute radiation injury that have appeared in the medical literature, the case histories are presented in considerable detail, and the complete autopsy protocols of the two fatally injured patients are included. The essential findings in each case are summarized at the end of each case presentation.

The report comprises ten sections, each dealing with a specific category of the acute radiation syndrome. For ease of publication, the report is divided into three separate volumes. This present volume includes three sections: I, The Nature of the Radiation Accidents; II, Radiation Doses, and III, Presentation of the Cases. Volumes II and III will discuss: The Biological Basis for the Clinical Response; Clinical Signs and Symptoms; Hematology; Chemistry of the Blood and Urine; Induced Radioactivity; Discussion of Pathological Findings; Reconsideration of the Calculated Radiation Doses; Autopsy Protocols; Bibliography and Acknowledgments.

This report is intended primarily for the clinician who is interested in radiation injuries and, therefore, emphasis has been placed on the correlation of clinical and pathological changes with the type of cytogenetic change known to be produced by ionizing radiation. To keep reference to fundamentals from being too repetitive for specialists in the radiation field, discussions of this type are confined, in so far as is possible, to Sections IV, VIII, and X. The present cases are contrasted with those instances of uncomplicated acute radiation syndrome observed in the Japanese victims of the atomic bomb explosions. The clinical and pathological responses are also compared with those in commonplace injuries such as thermal burns. In the discussion of these cases, an effort is made to review the pertinent literature in the fields of radiology and radiobiology.

TABLE OF CONTENTS

	Page
INTRODUCTION	5
IV. THE BIOLOGICAL BASIS FOR THE CLINICAL RESPONSE SEEN IN THE ACUTE RADIATION SYNDROME	11
A. Physical Conditions	11
1. Nature of Ionizing Radiation	11
2. The Relative Biological Effectiveness (R.B.E.) of Various Ionizing Radiations	11
3. The Units of Radiation Dosage	12
4. Penetration of Matter by Ionizing Radiation	13
B. Biological Considerations	13
1. Cell Damage Produced by Ionizing Radiations	14
a. Radiation-Induced Chemical Changes	14
b. Mechanism by which Radiochemical Changes Cause Cell Injury and Death	15
(1.) Inadivation of enzymes	15
(2.) Modification of genes and chromosomes	15
c. Types of Radiation-Induced Cell Death	17
d. Effect of Radiation of the Cell Environment	17
2. Tissue Damage Caused by Radiation	18
a. Delayed Manifestations of Tissue Damage in Animals Subjected to Whole-Body Radiation	18
(1.) Latent period preceding tissue damage and rate of tissue destruction	18
(2.) Other factors influencing tissue damage	19
b. Variation in Tissue Radiosensitivity	19
c. Regeneration of Radiation-Injured Tissue	20
d. Contrast of Radiation-Induced Tissue Damage with that Caused by Other Toxic Agents	20
3. Systemic or Constitutional Reaction of the Irradiated Individual	20
a. Toxic Catabolic Products Given Off by Dying and Dead Cells	20
b. Disturbance of Organ Function Produced by Radiation	21
c. Factors Secondary to Organ Damage in Radiation Injury	21
d. Response of the Individual to Total-Body Irradiation	21
4. Variation in the Response of the Individual to Total-Body Irradiation	22
C. Summary	22
V. CLINICAL SIGNS AND SYMPTOMS	23
A. Hand Injuries	23
1. Edema of the Hands	23
2. Extent of the Dry Gangrene of Case 1	24
3. Absence of Erythema	24
4. The Swelling of the Upper Arms	24
5. Treatment	25
6. Summary	26
B. Reaction of the Skin and its Appendages	26
1. Skin Reaction of Case 1	26

482 4

	Page
2. Skin Reaction of Case 3	26
3. Skin Reaction of Survivors	27
4. Summary	27
C. Systemic Response	27
1. Initial Reaction of the Patients	28
2. Toxemia	29
3. Gastro-Intestinal Symptoms	29
a. General Features	29
(1.) Phases of response	30
(2.) Reasons for phasic response	31
b. Unusual Features of Gastro-Intestinal Response	31
(1.) Gastric retention	31
(2.) Radiostomatitis	32
(3.) Gastro-intestinal secretions	32
4. Cardiovascular System	33
5. Jaundice	33
6. Genito-Urinary System	33
7. Weight Loss	34
8. Hemorrhagic Lesions	34
9. Delayed Weakness and Fatigability	35
10. Absence of Symptoms in Cases 2 and 6 through 10	35
11. Treatment	35
a. Consideration of Specific Drug and Harmone Therapy	36
b. Physiological Considerations	36
c. Practical Considerations in the Treatment of Patients with the Acute Radiation Syndrome	36
12. Summary of the Systemic Reaction	37
D. Addendum - Changes in the Eye of Case 4	37
VI. DISCUSSION OF HEMATOLOGICAL FINDINGS	39
A. Methods	39
B. Changes in the Blood Count	39
1. Lymphocyte Response	40
a. Development of Lymphopenia	40
b. Recovery of the Lymphocyte Count	41
c. Cause of Radiation Lymphopenia	41
2. Total Leucocyte and Neutrophil Count	42
a. Response of the Patients	42
b. Initial Increase in Neutrophils	42
c. Neutropenia	43
3. Eosinophil Count	44
4. Monocyte Count	44
5. Basophil Count	45
6. Erythrocyte Response	45
7. Reticulocyte Count	46
8. Platelet Count	46
9. Summary	46
C. Cytologic Changes in the Blood Cells	47
1. Lymphocytes	47
2. Neutrophils	48
3. Monocytes	48
4. Platelets	48
5. Summary	48

Page

D. Bone-Marrow Response as Revealed by Sternal Cavity Aspirations 48
E. Disturbance of the Blood-Clotting Mechanism 49

VII. CHEMISTRY OF THE BLOOD AND URINE 51

A. Chemistry of the Blood 51
1. Quantitative Changes in Blood Chemistry 51
a. Sodium and Chloride Ions 51
b. Calcium 51
c. Non-Protein Nitrogen and Urea 54
d. Uric Acid 54
e. Total and Fractionated Serum Proteins 54
2. Electrophoretic Pattern of the Plasma Proteins 55
3. Summary 55
B. Chemistry of the Urine 57
1. Corticoid-like Fraction 57
2. Urobilinogen 63
3. Porphyrins 64
4. Urorosein and other Pigments 64
5. Amino-Aciduria 68
6. Potassium 69
7. Summary 69

VIII. INDUCED RADIOACTIVITY 71

A. Interaction of Fast Neutrons and Tissue 71
B. Interaction of Slow Neutrons and Tissue 71
1. The (n, p) Reaction 71
2. The (n. γ) Reaction 71
3. The Neutron Capture Reaction 71
C. Use of Neutron-Induced Activity in Calculating Radiation Dosages 72
D. Contribution of Radiation from Induced Tissue Activity to Total-Body Dose 72
E. Unusual Observations Related to Induced Radioactivity 72
1. The Constancy of the Specific Activity of Serum Sodium 78
2. The Inequality of the Specific Activity of Serum and Urine Phosphorus 78
3. The Unexpectedly High Specific Activity of Urine Sodium in Case 3 79
4. Relative Specific Activities of Serum Sodium and Phosphorus Compared with the Known Neutron Capture Cross Section of These Elements 79
F. Summary 79

IX. DISCUSSION OF PATHOLOGICAL FINDINGS 81

A. Methods 81
B. General Features of the Tissue Reaction 81
1. Suppression of Mitoses 81
2. Cell Damage and Death 82
a. Nuclear Changes 82
(1.) Unusual type of nuclear disintegration 82
(2.) Unusual or chromosomal type of nuclear damage 82

	Page
b. Cytoplasmic Changes	82
(1.) Evidence of cytoplasmic dysfunction	82
(2.) Cellular differentiation	83
3. Interstitial Tissue Response	83
4. Blood Vessel Changes	83
5. Cellular Reaction	83
a. Inflammatory Cells	84
b. Mast Cells and Their Probable Relationship to the Anticoagulant in the Circulating Blood	84
C. Skin	86
D. Hematopoietic Tissue	87
E. Reproductive Organs	87
F. Gastro-Intestinal Tract	88
G. Heart	88
H. Adrenals	89
I. Liver	89
J. Kidneys	89
K. Summary	
X. RECONSIDERATION OF THE CALCULATED RADIATION DOSES IN TERMS OF THE OBSERVED BIOLOGICAL RESPONSE OF THE PATIENTS	91
A. Reliability of the Radiation Dose Calculations	91
1. Doses in Terms of Energy Units	91
2. Doses in Terms of X-Ray Equivalent	92
B. Reconsideration of the Characteristics of the Radiation Mixture and its Biological Implications	92
1. Uneven Distribution of the Radiation in Cases 1, 3, and 4	92
2. The Depth Dose of the Radiation	92
3. Dose Rate of the Incident Radiation	93
4. Specific Effect of Neutron Irradiation	93
C. Consideration of the Quantitative Aspects of Selected Tissue Response	94
1. Lethal Doses of Total-Body Radiation in the Human	94
2. Epilation	94
3. Sterilization	94
4. Delayed Damage to the Lens of the Eye	95
5. Initial Reaction or Radiation Sickness	95
6. Bone Marrow Damage	95
D. Summary	96

IV. THE BIOLOGICAL BASIS FOR THE CLINICAL RESPONSE SEEN IN THE ACUTE RADIATION SYNDROME

The pathogenesis of the acute radiation syndrome can be appreciated more completely if one understands the way in which ionizing radiations produce biological damage. Therefore, before discussing the medical aspects of this disease, a brief resume of some of the pertinent fundamental facts and concepts underlying the biological actions of radiation will be presented. Only those phases of the subject that relate to the present study are discussed in this section. The reader is referred to the book by Lea (1947), to the group of articles published in the British Medical Bulletin (Spear, 1946; Gray, 1946; Catchside, 1946), and to an article by Zirkle (1949) for a more thorough discussion of the actions of radiation on tissue.*

A. Physical Considerations

Since this report is written primarily for the physician, it seems worth while to define some of the more important physical terms and to explain their significance as well as the limits of their application in this study.

1. Nature of Ionizing Radiations

The ionizing radiations of interest in this study are neutrons, gamma rays, beta particles, and x-rays. They have the common property of dissipating energy in matter by causing ionization of atoms. Ionization, in this sense, means the ejection of an electron from its mother atom with the resultant production of two ions -- one, the negatively charged electron; the other, the heavier positively charged atom. The electron usually comes to rest at some distance from the atom, making recombination of the two unlikely. Thus, the ion pair exists for a finite time before it is neutralized by coming in contact with particles of opposite charge. The amount of energy expended by radiation in producing an ion pair is not constant, since the electron may be ejected from its orbit with varying degrees of violence. In fact, many instances of incomplete separation of the electron from the atom occur. This process of knocking an electron from an inner to an outer orbit is known as excitation. Taking excitation into account as well as ionization, it has been found that the average amount of energy expended per ion pair is 32.5 electron volts. This means that a gamma ray or a beta particle with an energy of one million electron volts will produce 30,785 ion pairs before dissipating all its energy.

The ability of ionizing radiation to produce ion pairs in matter distinguishes it from other types of radiation, such as ultraviolet light, which lose energy largely by excitation of atoms. This unique property of these radiations is of utmost practical importance, since ionization of tissue is believed to be responsible for all its biological effects (Lea, 1947), while ionization of air forms the basis for radiation-dose measurements.**

2. The Relative Biological Effectiveness (R.B.E.) of Various Ionizing Radiations

All forms of ionizing radiation produce the same type of tissue reaction. However, although the observed effects are essentially the same, the efficacy in producing cytological damage varies with the nature of the radiation. Thus, a dose of

*In most instances, in this as well as in succeeding sections, reference is made to recent review articles rather than to each original paper.

**Recently, it has been shown that, in certain radiochemical reactions as exemplified by the dissociation of organic chlorides, excitation plays a more important role than ionization (H.A. Andrews, personal communication). It seems quite possible that reactions of this type, although as yet undemonstrated, may take place in irradiated protoplasm.

neutrons will cause more damage per unit energy absorbed by the tissue than will the identical physical dose of gamma or beta rays. The difference in biological effectiveness of these radiations is due to the spatial distribution of the ions produced as a result of the interaction of the radiation with the tissue. It has been found that the more dense the ion tracks produced by the radiation, the greater the biological effect per unit energy dissipated in the tissue (Zirkle, 1935). Neutrons produce dense clusters of ions in an indirect manner as described in Section VIII, while gamma rays cause the formation of relatively widely spaced ions. The former are more effective biologically than gamma rays, but are not as damaging as alpha particles which cause even more dense ionization (Zirkle, 1935).

The term "relative biological effectiveness (R.B.E.)" has been introduced to take into account the quantitative difference per unit energy absorbed in the biological action of ionizing radiation (Zirkle, 1943). The R.B.E. is a convenient term to use in comparing the biological damage produced by different radiations. For example, the R.B.E. per unit ionization of fast neutrons, as compared to gamma rays, is 4 to 10 for most acute biological reactions in mammalian tissues (Gray, et al., 1940; R.D. Evans, 1947), and is even higher in the case of chronic exposures to these two radiations. It should be emphasized at this time that the R.B.E. for neutrons and x-rays is not constant but may vary for different effects even in a single tissue, or for the same effect in different tissues (see Section X-B). This means that it is impossible to use a single value for the R.B.E. of all the biologic effects produced in animals exposed to two types of radiation.

Despite differences in effectiveness and in the mechanism of causing tissue damage (see Sections VIII and X), the biological actions of the different types of ionizing radiation are sometimes directly additive. Thus, if an animal receives half a lethal dose of fast neutrons and half a lethal dose of gamma rays, death will occur in the same manner as it would if a lethal dose of either radiation had been given singly (Zirkle, et al., 1945). However, other experiments indicate that the actions of the combined radiations are less effective than either applied alone (Mitchell, 1947).

3. The Units of Radiation Dosage

The units of ionizing radiation used in this report are the roentgen, the roentgen equivalent physical (rep), the roentgen equivalent man (rem), and the gram roentgen.* It has been stated previously that ionization of air forms the basis of radiation-dose measurements. Thus, the units of dosage may be expressed in terms of the number of ions produced per cubic centimeter of air under standard conditions. Since the amount of ionization in air is dependent upon the energy dissipated by the radiation, dosage may also be expressed in terms of the energy loss per unit mass of air. The latter method of expression is particularly convenient in dose computations.

A roentgen of x-rays is the amount that will produce 2.08×10^9 ion pairs in a cubic centimeter of air under standard conditions. It may also be defined as the amount of radiation that dissipates 83.9 ergs per gram of dry air. It is important to realize that this term does not refer to the rate at which the radiation is administered or to the amount of tissue exposed to the x-ray beam, and to understand that, strictly speaking, the roentgen can be applied only to x-rays and gamma rays.

Other units have been formulated to define the dose of beta rays, neutrons, and other types of radiation. Of these, the roentgen equivalent physical (rep), perhaps the unit most frequently used in the United States, is also based on the ionization of

*The N unit, an empirical unit of neutron dosage frequently seen in radiobiology literature, is based on measurements using a certain type of ionization chamber. It may be defined as the neutron dose that gives the same reading with the Victoreen Thimble Chamber as one roentgen of gamma rays. The value differs slightly for the 25- and 100-r chambers.

482 9

air. The rep may be defined as the amount of any ionizing radiation (beta rays, neutrons, etc.) that dissipates the same amount of energy per gram of air (or produces the same number of ion pairs in air) as a roentgen or x-rays or gamma rays. Therefore, the rep, like the roentgen, is a unit of physical measurement. As has been pointed out, the biological efficacy of a given amount of ionization varies with the type of radiation. In an attempt to take the relative biological effectiveness into consideration so that doses of different kinds of radiation can be compared directly, the term roentgen equivalent man (rem) has been introduced. A rem of any type of radiation -- neutrons, for example -- is defined as the amount that is equivalent in its biological action on man (assumed from effects on other higher organisms) to one roentgen of hard x-rays. This value for any type of radiation can be computed by multiplying reps by the R.B.E. of the radiation.

Since the terms "roentgen", "rep", and "rem" do not take into account the amount of tissue that has been irradiated, the gram roentgen, another radiation unit, has appeared (Mayneord, 1940, R.D. Evans, 1947). This unit is obtained by multiplying rep by the mass of tissue that has been irradiated. One gram roentgen, then, may be defined as the energy lost in one gram of air by one rep. As this unit integrates the total energy dissipated in a mass of tissue, the radiation dose to which it applies is called the integral or volume dose. When the amount of energy lost by radiation passing through the entire body is considered, the gram roentgen unit is too small for practical usage. Therefore, the term megagram roentgen (or gram roentgens multiplied by 1,000,000) is applied to doses of this magnitude.*

4. Penetration of Matter by Ionizing Radiation

Ionizing radiations differ in their ability to penetrate tissue. Fast neutrons, energetic gamma rays, and hard x-rays have great powers of penetration in tissue. Thus, radiation of this type originating from a source outside the body has the potentiality of reaching and injuring all the organs in the body. It has been pointed out previously (Section II, Vol. I) that neutrons are much less penetrating in tissue than x-rays or gamma rays of comparable energy. Soft x-rays have limited powers of penetration; therefore, they cause damage primarily to the skin and superficial tissues when delivered to the exterior of the body. The range of beta rays in tissue, in contrast to that of the penetrating radiations, is not more than a few millimeters. Thus, beta rays from a source outside the body can injure only the skin and skin appendages. The effect of beta rays from radioactive materials deposited within the body is similarly confined to the cells in the immediate vicinity of the radioactive materials.

The term "depth dose" is frequently used to describe the penetrating power in tissue of a beam of radiation that originates outside the body. It refers to the fraction of the radiation dose that reaches a given point within the body, and is usually expressed in terms of percentage of the surface dose. The depth dose depends upon the type and energy of the radiation and the size of the beam. This term is particularly useful in the radiation therapy of internally placed tumors.

B. Biological Considerations

In order to translate the physical phenomenon of ionization into the ultimate clinical picture of the acute radiation syndrome, it is necessary to consider each stage in the sequence of events leading to the response of the body as a whole. Thus, the biochemical changes induced by radiation in protoplasm must be viewed at the level of cellular activity; similarly, the reaction of injured tissues should be evaluated

*All the radiation doses discussed thus far are based on the energy absorption in air. However, the energy absorbed from a given radiation is not the same in air and tissue. Thus, the energy loss by one roentgen of hard x-rays is 83.8 ergs per gram of air and 93 ergs per gram of wet tissue. This must be taken into consideration in dose computations.

in terms of damage to the component cells; and, finally, the clinical response of the individual can be appreciated only when the damage to each organ system is taken into account.

In discussing the fundamental biological reactions, it will be noted that conclusions concerning radiation effects in man are drawn from observations on numerous biological materials ranging from simple unicellular organisms to complex tissues of other animal species. The tendency to oversimplify biological reactions and to explain all observed phenomena in terms of those mechanisms that we understand is great. However, the extrapolation of these diverse data to man, and oversimplification of cellular dynamics appear justifiable since they provide a stepping stone to a workable concept of the nature of the acute radiation syndrome. However, one must constantly keep in mind the realization that the true state of affairs may be much more complex.

1. Cell Damage Produced By Ionizing Radiations

The fundamental lesion in the irradiated individual is damage to the cell. The cytogenetic response of the individual irradiated cells is incomplelety understood, but discussion of experimental observations and of the present concepts of the mechanism of cell damage add immeasurably to our understanding of the biological response in man.

a. Radiation-Induced Chemical Changes

Irradiation of cells or tissues leads to chemical changes in the molecules that are ionized. The chemical reactions that occur in protoplasm are of two general types depending upon the type of molecule that has been affected. Thus, there may be radiochemical changes due to ionization of either organic molecules or water molecules (discussed by Lea, 1947). In the first instance, an organic molecule that has been ionized may undergo either chemical modification or disruption. For example, a protein molecule may be denatured, or broken into smaller peptide units, as a result of ionization. It is easy to see how such a biochemical change may lead to cell injury, particularly if the affected molecules are important to the physiology of the cell. In the second type of chemical change, that involving water molecules, it is believed that an ionized water molecule may undergo one of a series of chemical transformations which can result in the formation of highly reactive radicals or compounds. Indirect evidence suggests that ionized water molecules may be converted into free (uncharged) OH radicals, atomic hydrogen, or perhaps hydrogen peroxide (Lea, 1947). It has also been suggested that irradiation of water containing organic material leads to the formation of stable organic peroxides (Barron, Flood and Gasvada, 1949-c). Thus, it becomes apparent that the formation of "activated" products in the aqueous medium of protoplasm can cause cell damage by the interaction of these products with the organic constituents of the cell.

Such primary radiochemical reactions in irradiated aqueous media are limited quantitatively by the number of atoms that have been ionized. Usually one (or not more than two or three) molecule undergoes a chemical change for every ion produced (Lea, 1947). In some organic compounds, the yield of chemical reactions is much lower. Since ionization occurs indiscriminately throughout protoplasm, it may be calculated that two or three ionizations and subsequent chemical modification occur in the aqueous medium for every one in the constituents of protoplasm. It is impossible to say, however, which type of radiochemical reaction is more important biologically. Undoubtedly, many of the organic molecules, modified directly by irradiation, are unimportant to cell function, and such modification does not interfere with normal cell activity. These ionizations are wasted in so far as contribution to cell damage is concerned. In the case of the activated products of irradiated water, as has been pointed out, the damage to the cell takes place in an indirect manner. Labile groups of nearby molecules compete for the highly reactive water products which, being small, can move easily in protoplasm and act over a finite range. It seems likely that the more labile of the chemical groups will attract the activated

products and will undergo more chemical reactions than will the less labile groups. For example, it is believed that sulfhydryl radicals of a certain kind are particularly prone to being oxidized in this manner (Barron and Dickman, 1949-a). Thus, there appears to be a certain degree of selectivity in radiochemical reactions in protoplasm, although not in the same sense that ultraviolet radiation is selectively absorbed by specific chemical groups.

b. Mechanism by Which Radiochemical Changes Cause Cell Injury and Death

It is important to realize that the number of ions formed per cell is not great, and, therefore, only a small fraction of the molecules within a cell undergoes chemical changes as a direct result of the action of radiation. Thus, only one in every ten million ~~cells~~ Molecules is modified by exposure of a tissue to 1000 roentgens of x-rays (Zirkle, 1949). This means that 1000 roentgens will cause chemical changes in about ten million molecules of a cell of average size, i.e., containing approximately 10^{14} molecules. It is obvious, then, that the primary radiochemical changes that occur indiscriminately in relatively few molecules must be greatly amplified by intracellular mechanisms to account for the microscopically visible structural changes seen in cells injured by radiation. Although there may be other methods of amplification of primary radiation effects, the following two mechanisms have been observed experimentally: (1) inactivation of enzyme molecules and (2) modification of certain key genetic units within the cell nucleus.

(1.) Inactivation of enzymes: Inactivation of a single enzyme molecule by radiation may influence many thousands of other molecules in the reaction system. This is illustrated by the accumulation of ribonucleic acid in the cytoplasm of irradiated cells as demonstrated by ultraviolet photomicrography (Mitchell, 1942). It has been calculated that at least 1000 to 100,000 nucleic acid molecules are indirectly affected for every molecule directly ionized by radiation (Mitchell, 1943, 1946). The actual amplification must be much greater than this, since only relatively few of the molecules directly modified by the radiation are concerned with nucleic acid metabolism. It is believed that this effect on nucleic acid metabolism is mediated through damage to the enzyme system. Thus, it has been shown that irradiation of rats diminished the ribonuclease activity in the spleen, (Carter, 1949). Further direct proof of the action of radiation on intracellular enzymes is shown by the selective inhibition of the sulfhydryl oxidative enzymes in various tissues demonstrable after exposure of an animal to 100 roentgens or more of x-rays (Barron, Wolkewitz and Muntz, 1947).*

(2.) Modification of genes and chromosomes: A far more effective method of amplifying primary radiation effects is through destruction or modification of key intranuclear molecules that control important cellular processes. Two main types of such intranuclear changes have been shown to be responsible for severe damage or death of cells. The first, and presumably the simplest, is the gene mutation. A mutation of this sort can be produced by a single ionization, or at the most, a cluster of two or three ionizations within a gene

*Indirect experimental evidence suggests that the action of radiation on the easily damaged -SH enzymes occurs as a result of the inactivation of the molecule by "activated" water (Barron and Dickman, 1949-a). The effect of radiation on the more resistant non-SH containing enzymes is believed to be a result of denaturation of the enzyme molecule. The great radiosensitivity of the former type of compound is shown by the fact that significant inhibition of adenosine triphosphotase can be achieved by irradiation of a pure solution of the enzyme with one roentgen of x-rays (Barron, et al., 1949-b).

unit (Lea, 1947). The yield of mutations is directly proportional to the dose of radiation. The second type of intranuclear damage is due to the production of structural changes in the chromosomes. Breakage of chromosomes may occur if an ionizing particle passes through a chromosome or in its immediate vicinity (Lea, 1947).* As few as twenty ionizations within a chromosome may be sufficient to cause a chromosome break (Gray, 1946). The number of simple chromosome breaks, like the yield of gene mutations, is directly proportional to the dose. It has been estimated that 2048 roentgens of x-rays would be required to cause an average of one break per chromosome of irradiated grasshopper neuroblast cells (Carlson, 1941). Most chromosome breaks undergo restitution, but abnormal structural rearrangements occur frequently. The latter abnormalities can be easily visualized during mitosis when the chromosomes become large and readily stainable. Examples of bizarre chromosomal changes can be seen in the photomicrographs of lymphoid and intestinal tissues of Case 3.

Gene mutations and chromosome structural changes may lead to death of the cell. Lethal gene mutations have been convincingly demonstrated in bacteria and viruses (Lea, 1947). Recessive lethal gene mutations, which may not become manifest for many generations, have been observed in fruit flies (drosophila melanogaster) (Lea, 1947). In irradiated mammals, there are relatively few examples of experimentally produced gene mutations in germ plasm (Hertwig, 1939, Charles, 1949). In contrast to lethal gene mutations, which can be more easily demonstrated in lower organisms than in higher animals, chromosomal aberrations -- or chromosome mutations, as they are sometimes called if mitosis is successful -- have been shown to be an important, if not one of the primary causes, of cell death in many biological materials, including mammalian tissues (Lea, 1947). Chromosomal aberrations, such as failure of a broken chromosome to undergo restitution, or the formation of chromosomal bridges, may render a cell incapable of dividing for obvious mechanical reasons. Presumably, this type of chromosomal damage is responsible for the delayed cell death during mitosis, discussed later in more detail. Even though a cell with chromosomal structural changes may complete mitosis successfully, the genetic imbalance in the daughter cells may be incompatible with life or continued reproduction.**

*The concept that ionization occurring within a specific and often submicroscopic portion of a cell leads to cytogenetic change of a definite type is known as the target theory (Lea, 1947). This theory has been used for some time to explain the quantitative aspects of radiation-induced gene mutations and chromosome breaks. The comparatively recent demonstration that chemicals such as the nitrogen mustards can also produce mutations and chromosome breaks make it clear that other mechanisms besides that implied in the target theory may play a role in the production of radiation-induced cytogenetic changes (Zirkle, 1949). One of the obvious other mechanisms is that mediated through "activated" water. It has been stated recently that the target theory is encountering more and more difficulties in explaining radiation effects, even within relatively stable biological systems (Koller, 1949). It is evident from the opinions expressed in the current literature that increasing emphasis is being placed on the indirect or radiochemical mechanisms as a cause of cytogenetic effects in irradiated tissues.

**The consideration of non-lethal gene or chromosome mutation in germ plasm with transmission of altered characteristics to the offspring of an individual is beyond the scope of this discussion. The reader is referred to a recent report by Dr. Donald Charles (1949) and to other sources (Glucksmann, 1947) for a consideration of the effect of acute doses of radiation on reproductive tissues.

c. Types of Radiation-Induced Cell Death

Dismissing for the moment cell death due to environmental factors, it appears that two main types of cell death are caused by radiation. The first is observed in cells during their resting or intermitotic stage. This mode of death occurs soon after exposure and is caused only by large radiation doses. The exact mechanism responsible for prompt cell death is not clear, but it seems likely that it is similar to the usual processes of cytolysis seen in normal tissues. The second kind of death does not occur until the fatally injured cells attempt mitotic division. Such delayed cell death, produced by relatively small doses of radiation, is probably due to chromosomal damage (Lea, 1947). Cell death during mitosis apparently occurs only rarely in normal tissues, since the characteristic cytogenetic changes are not seen in histological sections of untreated animals.

These two types of cell death are illustrated by an experiment with tissue cultures (Laznitski, 1940, 1943). Exposure of a culture of chick fibroblasts to 2500 roentgens of x-rays results in the degeneration of a substantial number of resting cells within a few hours. In contrast, irradiation of the tissue cultures with 100 roentgens has no immediate effect on the resting fibroblasts. Later, however, when many of these cells try to divide, they either degenerate during the act of mitosis or produce non-viable daughter cells.

Delayed cell death during or after mitosis can be demonstrated indirectly in vertebrates. Irradiation of an animal (tadpole, mouse, etc.) produces a prompt decrease in the mitotic activity of proliferating tissues (Spear and Glucksmann, 1938; Knowlton and Hempelmann, 1949). This is due to the fact that cells in the pre-mitotic phase are rendered temporarily unable to enter mitosis.* After a lag period, the length of which depends upon the size of the dose of radiation and the nature of the tissue, the cells regain their ability to divide. Careful studies correlating the mitotic activity with cell degeneration after irradiation with a few hundred roentgens of x-rays indicate that, in tadpoles and in the developing rat retina, degeneration of cells does not occur until mitotic activity is resumed (Spear and Glucksmann, 1938; Tansley, Spear, and Glucksmann, 1937). There is an excellent correlation between the numbers of degenerating and mitotic cells. This is strong circumstantial evidence that these tissue cells do not die until they attempt to divide.

Experimental study also indicates that animal cells are particularly sensitive to the chromosomal type of lethal radiation damage just before they enter mitosis (Marshak, 1942-a). The premitotic radio-sensitivity is not so pronounced with neutron irradiation as with x-rays (Marshak, 1942-b; Spear and Tansley, 1944). This observation, which suggests that the resting cells are more severely damaged by neutrons than by x-rays, is an example of one of the few demonstrable qualitative differences between the biological actions of these two types of radiations. In contrast to these observations in animal tissues, it has been possible to show that in plant cells (Trillium) irradiated with x-rays, chromosomal damage is greatest in the metaphase and anaphase stages of mitosis (Sparrow, 1944). Such chromosomal abnormalities do not become manifest until the following cell division.

d. Effect of Radiation of the Cell Environment

While cellular damage due to intracellular changes is undoubtedly very important in the death of irradiated unicellular or simple organisms and of cells in tissue culture, recent experiments have shown that the effects of radiation of the environmental media

*The cause of the delay in cell division is not definitely established, but it may be related to the temporary disturbance in the desoxyribonucleic acid metabolism shown to occur in radiation-injured cells (Hevesy, 1945; Lea, 1947). Recently it has been suggested that the action of radiation on the high sulfhydryl content of dividing cells is responsible for suppression of mitosis (Barron and Dickman, 1949-a).

are also important in determining the response of simple organisms (T. C. Evans, 1947; Dickey, Cleland and Lotz, 1949). In the complex body tissues, it is obvious that the action of radiation on the cell environment must play an even greater role in determining the response of the cell. Thus, cell injury or death may result from damage to the blood supply, from the injurious effects of toxic products given off by adjacent dead or dying tissue, or from other environmental factors as yet unknown. The dry gangrene and ischemic necrosis of the hand tissues of Case 1 are outstanding examples of the first of these three environmental factors. The relative importance of direct cellular damage and environmental changes in producing tissue death is not clearly understood for the dose range usually involved in the acute radiation syndrome. In the higher dose ranges, such as are used in radiation therapy, it seems highly probable that both factors contribute to tissue death (Koller, 1947; Lasnitzki, 1947).

2. Tissue Damage Caused by Radiation

Injury of tissues exposed to ionizing radiation is obviously the result of death and damage of its component cells. Since the clinical response in patients with the acute radiation syndrome is dependent upon tissue damage of individual organ systems, it is important to consider carefully the manner in which such changes take place.

a. Delayed Manifestations of Tissue Damage in Animals Subjected to Whole-Body Radiation

If the tissue reactions in humans and animals are considered in the light of data just presented, one would expect the "delayed" type of cell death to predominate as a result of the doses of total-body radiation usually responsible for the acute radiation syndrome (50 to 1500 rem). This is borne out by clinical and histological observations. An obvious clinical example of delayed tissue damage is found in the case of skin heavily treated with x-rays; here, epidermolysis does not occur for a week or more after exposure. Histological evidence of cell destruction is also seen only after a period of delay that varies from tissue to tissue. For instance, in the case of lymphoid tissue, the delay in cell death is only 30 minutes in rabbits given 800 roentgens of x-rays, while in the epidermis of mice exposed to 5000 rep of beta rays, marked histological damage is delayed for 10 days (Bloom, 1948).

(1.) Latent period preceding tissue damage and rate of tissue destruction: There are no direct experimental data to explain the nature of the variability in the latent period before cell death and in the rate of tissue destruction in different organs. However, it seems logical to assume that the period of delay in cell death and the rate of tissue destruction can be correlated with the normal life span of the component cells. It may be reasoned that, following irradiation, a tissue composed of cells with a short life span should undergo rapid destruction after a brief delay, while tissues whose cells are long-lived should undergo less-rapid degeneration after a longer latent period. Indeed, this seems to be the case both in so far as the period of delay in cell division and the rate of cell destruction are concerned. In the rat, for example, thymic lymphocytes, whose life span is short, possibly of the order of two days,* undergo rapid lysis

*This estimate of the life span of the tissue lymphocyte is based on the time of turnover of nucleic acid in lymphoid tissues of rats (Andreason and Ottesen, 1945). The value is somewhat longer than the average duration of life of the blood lymphocyte which, though debatable, is believed to be twelve hours or less (Drinker, Kent and Yaffey, 1941; Lawrence, Dowdy and Valentine, 1948). The concept of the two-day life span is in conflict with a recent computation based on experimental results of a different nature, which gives a value of 580 ± 10 hours for the intermitotic period of all cell types in the lymph node of a mouse (Knowlton and Widner, 1950).

after a short latent period. Similarly, the epithelium of the small intestine, whose intermitotic time in the rat is said to be 1.35 to 1.57 days (Leblond and Stevens, 1948), undergoes prompt and rapid destruction. In contrast to these tissues, consider the more slowly proliferative human skin, the life span of whose basal epithelial cells has been calculated to be 129 days (Hoffman, 1949). Clinically and histologically, epidermolysis develops slowly and lasts for a number of weeks. Finally, examples of very slowly proliferating or non-proliferating tissues are provided by the liver, muscles, and nerve cells. Death of cells due to chromosomal damage, if the above assumptions are correct, should be very rare or should not occur at all in these tissues. Histological studies indicate that cell death is unusual in these radioresistant tissues except with very high doses.*

(2.) Other factors influencing tissue damage: Although it is tempting to try to simplify the concept of radiation injury by explaining all phenomena on the basis of inability of the injured cells to divide successfully, it must be stressed again that environmental changes contribute to the destruction of tissues. Delayed cell death can well be due, in part, to the late effect of radiation on the blood vessels and on other factors in the cell environment. Also, it should be pointed out again that extremely large doses of radiation can cause prompt cell death even in non-proliferating tissues. Thus, extensive cell destruction was noted in all types of tissue, including muscle and brain of rabbits at the end of a three-hour exposure period during which 50,000 roentgens of x-rays were administered (Henshaw, 1944-b).

b. Variation in Tissue Radiosensitivity

The wide variation in the degree of injury suffered by different organs of an animal exposed to radiation has been the subject of much discussion. In the early days of study of the biological effects of x-rays, a law was formulated to be used as a guide in predicting the response of normal and tumor tissues to radiation. This postulate, called the law of Bergonie and Tribondeau (1906) states, in effect, that the radiosensitivity of a tissue is directly proportional to the mitotic activity and inversely proportional to the degree of differentiation of the cells. In practice, there are many exceptions to this law.

While it is true that there are great differences in radiosensitivity, as, for instance between a nerve cell and a lymphocyte, it is important to bear in mind that there are no absolute and clearly defined categories of radiosensitivity. It seems advisable, therefore, to follow an arbitrary classification such as has been used in the study of the radiosensitivity of neoplasms (Warren, 1941). In this scheme, tumors are placed in the following broad and ill-defined groups: (1) radiosensitive, (2) radioresponsive, and (3) radioresistant.

If such a classification is applied to normal tissues, the blood-forming organs and reproductive tissues fall into the first or radiosensitive group; the epithelium of the gastro-intestinal tract, the skin and connective tissues can be placed in the radioresponsive group; and the radioresistant group is composed of the cells in the remainder of the organ systems of the body.

As mentioned previously in this section, the sensitivity to the actions of radiation is undoubtedly the result of many factors inside and outside the cell itself. One factor that would appear to determine tissue radiosensitivity is the proportion of cells in the premitotic phase, during which radiation injury, particularly in the case

*In Cases 1 and 3, the muscle fibers of the abdomen show waxy degeneration of the cytoplasm which is in keeping with the heavy irradiation of these tissues.

of irradiation with x-rays, is readily produced. Another factor is the vascularity of the tissue. Since, however, neither of these easily measurable characteristics gives a satisfactory guide to radiosensitivity, it may be assumed that there are other unknown factors that are important in determining the response of cells to the damaging actions of radiation.

c. Regeneration of Radiation-Injured Tissue

Regeneration of a tissue injured by ionizing radiation may occur before cessation of tissue destruction or at a variable time thereafter. The degree to which a tissue regenerates depends upon the number of surviving stem or mother cells and upon the inate ability of the surviving cells to proliferate. It also depends upon the damage sustained by the vascular supply of the tissue. Thus, the atrophic nature of the skin covering an old x-ray burn undoubtedly is a reflection, in part, of the scarring of the underlying tissues and the poor vascular supply. What factors govern the time at which regeneration starts is not at all clear. Healing of the skin in an acute x-ray ulcer may be delayed for months and then may occur suddenly and rapidly. Waves of destruction and regeneration have been described in some tissues (see Section V). These are even more difficult to understand.

d. Contrast of Radiation-Induced Tissue Damage with That Caused by Other Toxic Agents

It should be emphasized that the delay in the occurrence of tissue death differentiates radiation injuries from these produced by other physical agents, such as heat or electricity. The latter cause immediate cell death, presumably by coagulation of cell proteins. The clinical picture in the acute stage of thermal burn injuries, for example, represents the reaction of the body to localized and usually superficial dead tissue that was killed at the time of the injury. The clinical response in acute and subacute radiation injuries, on the other hand, is a reaction to dead and dying cells that persist in selected tissues throughout the body for a matter of weeks. The prolonged period of tissue destruction in this type of injury is not unlike that in certain chemical poisonings and bacterial infections.

3. Systemic or Constitutional Reaction of the Irradiated Individual

The response of the individual to irradiation of his entire body, or a large part thereof, is the result of the total damage inflicted on all his organ systems. Besides direct damage to the exposed tissues, there are other factors that determine the clinical reaction of the individual. The similarity in symptomatology of the acute radiation syndrome to other diseases in which tissue destruction is prominent (thermal burns, intestinal obstruction, etc.) suggests that the indirect factors are extremely important in shaping the clinical reaction.

a. Toxic Catabolic Products Given off by Dying and Dead Cells

In a damaged tissue, the dying and injured cells give off products of cell catabolism that are abnormal in quantity and perhaps in quality. This material, which is liberated into neighboring tissues and into the body system at large, constitutes the so-called "toxic" products of cell breakdown.* Thus, the blood adenyl compounds

*In order to avoid introducing too many complicating factors, the assumption is made that all toxic products in damaged tissues come from cytolysis of dead cells or from distorted catabolism of injured and dying cells. It should be pointed out that bacterial toxins have not been considered in many experimental studies of this type. Toxins, such as those derived from clostridium Welchii and other anaerobes, are important in producing experimental traumatic shock in animals (Aub, 1944). Thus, infection may well play an important role where the bacterial barriers are broken.

have been shown to increase sharply in rabbits with fatal cutaneous thermal burns (Stoner and Green, 1949). Experimental evidence suggests that much of the toxic material liberated as a consequence of thermal burns originates in the injured cells at the margin of the dead tissue (Leach, Peters and Rossiter, 1943; Peters, 1945). Similarly, studies of yeast cells injured by ultraviolet radiation and by other means point to the fact that damaged cells elaborate and discharge large quantities of materials that have growth-promoting properties (Loofbourow, 1948). Other information suggests that radiation-injured tissues show accelerated degradation of protein, while retaining much of their ability to synthesize protein (Hempelmann, et al., in press). All these data suggest that tissues injured by ionizing radiation give off toxic materials that may influence the neighboring tissues and the animal as a whole.

b. Disturbance of Organ Function Produced by Radiation

The specific function of tissues may be altered by radiation. Thus, the ability of the body to form antibodies is depressed after exposure of an animal to moderately large doses of irradiation (Craddock and Lawrence, 1948). Similarly, the decreased secretion of saliva caused by irradiation of these tissues is well known. The effect of radiation on the activity of the adrenal gland has been demonstrated (Engelstad and Torgersen, 1937; Patt, et al., 1947). This dysfunction may be responsible for many of the constitutional reactions of the irradiated animals (see the discussion of the general adaptation syndrome in Section VII and of the role of the adrenal in radiation death of the individual, Section IX). Thus it may be assumed that the function of all organ systems is disturbed to some degree (depending upon the radiosensitivity of the tissue and upon the dose received) by the exposure of an animal to a large or moderately large dose of ionizing radiation.

c. Factors Secondary to Organ Damage in Radiation Injury

Many factors, secondary to organ-system damage, play a prominent role in determining the acute reaction in the radiation-injured animals. For example, invasion of the body by intestinal bacteria is unquestionably responsible for some of the terminal clinical response and death of animals (Lawrence and Tennant, 1937; Bennett, et al., 1949-b). Such invasion is secondary to loss of the intestinal epithelium, destruction of myelopoietic tissues, diminution in the ability of the body to form antibodies and other factors that lower the resistance of the body to infection. Similarly, dehydration, unless successfully remedied, may influence the clinical response. This reaction is also secondary to damage of the intestines. Malnutrition subsequent to loss of appetite, and lowered absorption ability of the damaged intestines may be prominent in a prolonged illness of this type. These and other factors, some of which lend themselves to treatment, are important in determining the systemic reaction of an irradiated individual.

d. Response of the Individual to Total-Body Irradiation

As has been emphasized, the acute response of the individual to large or lethal doses of ionization is the result of the total damage inflicted on all organ systems. It seems appropriate, therefore, to summarize briefly what has already been said about the effect of radiation on the organs and their component parts.

Radiation damage to each organ system is the result of two types of tissue injury: (1) the direct injury and death of cells by intracellular chemical changes or by modification of the cell environment; and (2) the indirect injury to the tissue caused by: (a) the products liberated by dead and damaged cells in the same or other organs, and (b) the disturbed function of other interrelated organs. The extent of the damage of each organ system depends upon the radiosensitivity of the cells. The rate of tissue destruction varies from one tissue to another in such a manner that destruction and repair may go on simultaneously in different organ systems. The damage and degeneration of the tissues of various organ systems are directly responsible for

many of the aspects of the clinical response. In addition, factors secondary to organ damage influence the systemic reaction. Infection is undoubtedly one of the most important of the secondary effects but adrenal disfunction and disturbed fluid and electrolyte balance as well as malnutrition play prominent roles in the over-all body response. The clinical picture in the acute radiation syndrome, then, is one of a generalized systemic disease with evidence of massive tissue breakdown in selected organ systems throughout the body, complicated by infection and other factors. The symptoms due to damage of individual organ systems occur at varying times in the course of the disease as a result of difference in the time of maximum tissue destruction in individual organs.

4. Variation in the Response of the Individual to Total-Body Irradiation

There is a great difference in the sensitivity of various plant and animal cells to the actions of radiation. Lethal doses for the extremes in sensitivity throughout nature differ by a factor of 10,000 (Spear, 1946). Even in mammals, there is a great species variation in radiosensitivity. Thus, the thirty-day median lethal dose for guinea pigs exposed to 200-kv x-rays is 200 roentgens (Henshaw, 1944-a; Ellinger, 1945), while the life span of bats similarly irradiated is shortened only after doses of 15,000 to 16,000 roentgens (Smith, Svihla and Patt, 1949). There is also variation, although less marked, in the amount of radiation required to kill individuals of the same species. This can be illustrated by an experiment in which groups of highly inbred mice (Carsworth CF_1) of the same age and sex were subjected to various doses of hard gamma rays at a rate of 30 roentgens per minute (Henshaw, Riley and Stapleton, 1947). No deaths occured in those animals receiving less than 500 roentgens. Eight percent of the animals exposed to 500 roentgens were killed. In those mice receiving larger amounts of radiation, the percentage killed varied with the size of the dose. One hundred percent mortality did not occur until a dose of 1200 roentgens or more was administered This means that some animals survived more than twice the minimum lethal dose. The median lethal dose (MLD), which, by definition, is the dose that kills 50 percent of the individuals within a specified time (usually 30 days) after exposure, is 840 roentgens for mice of this strain irradiated with gamma rays of this energy and intensity.

Variable and graded responses are also noted when limited portions of the body are irradiated. Thus, the amount of the skin erythema and epilation produced by exposure to a single dose of x-rays will vary from person to person. This variation in individual response, noted in localized tissue reactions as well as in the response of the body as a whole, is comparable to that observed in the toxicity of pharmacologic action of drugs and chemicals.

C. Summary

The topics discussed in this section can be found in the Table of Contents. An effort is made to define the physical terms used in this report for the benefit of the reader not versed in radiation physics. The body damage is discussed in terms of injury to its component cells and tissues and of the consequent disturbed organ function. The variability in the response of the individual is also considered. Certain aspects of the biological action of radiations have been fitted elsewhere into the text of this report. Thus, the phasic nature of the tissue response is discussed in Section V, and the role of the adrenal in the acute radiation syndrome is considered in Sections VI, VII, and IX.

482 19

V. CLINICAL SIGNS AND SYMPTOMS

The outstanding aspect of the clinical picture presented by these ten patients is their close conformation to the pattern of radiation injuries previously described in humans and animals. The complexity of the incident radiation, the short duration of the exposure and the magnitude of the doses to limited parts of the body appear to have produced no basically new clinical phenomena. The clinical, laboratory, and pathological data will be considered in detail, however, since these injuries are unique in certain aspects. Other accounts of the acute radiation syndrome in humans can be found in the reports of the atomic bomb medical casualties in Japan (Medical Report of Joint Commission), and in articles dealing with a case of acute mesothorium poisoning (Hamperl and Roemheld, 1936), and with several instances of acute x-ray injury caused by teleoroentgen therapy (den Hoed, Levie and Straub, 1938). Recent physiological studies in animals exposed to total-body radiation have been summarized in a report by Prosser, et al., (1947-a).

To simplify discussion of the clinical signs and symptoms, this section will be divided into three parts--the reaction of the hands and upper extremities, the reaction of the skin and its appendages, and the systemic response.

The symptomatology of the illness of these patients will be compared with that in patients with thermal burns in an effort to establish a point of reference with a well-known and somewhat similar type of injury.

A. Hand Injuries

The reaction of the hands of Cases 1 and 3 were much more sudden and severe than that in other previously described radiation injuries. However, the nature and sequence of clinical events were similar, except in degree, to those in acute radiation burns of the usual type. Edema, pain, exudative epidermolysis followed by dry gangrene usually occur in this order in severe localized radiation injuries. Severe pain requiring large doses of morphine is the outstanding symptom during the acute stage of a severe x-ray or gamma-ray burn (Brown, McDowell, and Fryer, 1949). Four features of the reactions of these cases, however, are so extreme as to deserve comment. They are: (1) the promptness of the appearance of edema of the hands, (2) the extent of the dry gangrene of the right hand of Case 1, (3) the absence of erythema of the skin of the more severely injured hands, and (4) the marked inflammatory response of the upper arms of Cases 1 and 3. The treatment of the injuries (5) is also discussed.

1. Edema of the Hands

Swelling of both hands of Case 1 was pronounced within thirty minutes after exposure, while that of Case 3 appeared in a period of three hours. This tissue response was much more rapid than that usually produced by x-rays or gamma rays. Even beta radiation, which provokes prompt evidence of skin damage, has not been reported to cause visible skin reactions in humans in such a short period of time (Robbins et al., 1946; Knowlton et al., 1949). The explanation for the explosive nature of the response of the hand tissues undoubtedly lies in the magnitude of the dose of radiation to which these tissues were subjected, although it must be admitted that the soft radiations to which the hands were exposed may have contributed to the acceleration of the tissue response. Similar development of massive edema of subcutaneous tissues has been observed at the end of a three-hour exposure period in rabbits given 50,000 roentgens of x-rays (Henshaw, 1944-b). No morphologic changes were noted in the blood vessels of these animals at the end of the exposure, but marked damage and destruction of cells were observed in all other tissues including the radioresistant muscle and brain.

The edema which occurred in the hands of the two patients was undoubtedly the result of at least two factors -- one, the increased permeability of the injured capillaries, and two, the greatly increased osmotic pressure of the extravascular fluids created by the rapid destruction of protoplasm (Borak, 1942). The increase in osmotic pressure of these tissues must have been very great to account for the ligneous character of the hand edema of Case 3 and the tense destention of the blisters of Case 1. Such a marked increase in tissue osmotic pressure and the consequent edema is compatible with the severe cell destruction which must have occurred in all soft tissues of the hands in these cases.

The analogous edema occurring in tissues adjacent to thermal burns serves to illustrate the magnitude of the fluid shift which can result from even superficial tissue injury. Calculations indicate that for every one percent of the total-body surface burned, 150 cc of fluid (containing proteins and electrolytes) are lost from the vascular system into the interstitial spaces around the wound and onto the wound surface (Cope and Moore, 1947). Although this value may be high for hand burns (because of the limited tissue distensibility), it shows that the flow of fluid into the interstitial spaces adjacent to severely injured tissues can be massive and may seriously affect the fluid balance of the body. In contrast to the edema of thermal burns which reaches its maximum 36 to 48 hours after injury (Cope and Moore, 1947), the edema and loss of fluid from the wound surfaces in Case 1 did not reach its peak until the second week following exposure. The continuation of fluid loss from the wound surfaces during this two-week period indicates the persistence of blood flow into the damaged tissues.

2. Extent of the Dry Gangrene of Case 1

Gangrene often develops in the dermal tissues of severe superficial x-ray burns (MacKee and Cipollaro, 1946). The sloughing of the necrotic dermal tissue in such an injured area results in the formation of the characteristic acute radiation ulcer. The gangrene observed in the fingers and palms of Case 1 undoubtedly represents the same process. The superficial white necrotic tissue noted in the dermis of the dorsum of the right hand of Case 1 (Figure 21) is more characteristic in appearance to the type of tissue death usually seen in x-ray or beta-ray burns than is the blackened dry type of gangrene observed in the fingers. The blood vessel changes, which presumably are largely responsible for this type of late tissue death are similar to those seen in frost-bite and immersion foot (see Section X).

3. Absence of Erythema

The absence of erythema in the skin of the more seriously injured hands can be explained on the basis of edema of the superficial tissues which obscured the vascular reaction. Edema of the deeper tissues does not decrease the blood flow even through the extracellular osmotic pressure is markedly elevated (O. Cope, personal communication). That the peripheral circulation was disturbed soon after injury in both cases is shown by the early cyanosis. The late deep cyanosis of the hands and forearm of Case 3 was obviously largely due to the refrigeration of those tissues.

4. The Swelling of the Upper Arms

An intense inflammatory reaction is usually found around the gangrenous tissue in an acute radiation (MacKee and Cipollaro, 1946). A reaction of this nature is primarily due to the direct injury of the tissues by radiation. Secondarily, the response of these injured tissues to the nearly dead and dying cells undoubtedly contributes to the inflammatory process. It seems likely that both factors played a part in producing the marked swelling of the forearms and upper arms of Cases 1 and 3. The inflammatory reaction of the injured tissues of the arms probably contributed to the toxemia of the patients. This is analogous to the constitutional reaction in thermal

burns where experimental evidence suggests that some, if not a large share, of the toxic materials come from the injured but living cells at the edge of the burned coagulated tissue rather than from the dead tissue itself (Leach, Peters and Rossiter, 1943; Peters, 1945).

The possible role of infection of damaged tissues by anaerobic bacteria was pointed out in Section IV-B.

The discussion of the response of the hands should not be concluded without emphasizing the fact that prompt death of resting cells undoubtedly occurred in the tissues that were exposed to the heaviest doses of radiation (Section IV-B). The cell destruction, even in the tissue of the hands must have been almost comparable in rapidity and extent to that described in rabbits given 50,000 roentgens of x-rays (Henshaw, 1944-b). This fact must be kept in mind when one compares the clinical reactions of the hands of these patients with that of other humans and animals which have been irradiated with smaller doses of ionizing radiation.

5. Treatment

The treatment of the injured hands of Case 1 was patterned after that used in thermal burns, while the treatment of Case 3 represents an attempt to "amputate" by refrigeration, the hopelessly damaged tissue. The pressure dressing, vaseline gauze treatment often used in thermal burns, was modified somewhat in Case 1, so that the progress of the hand lesions could be followed. The use of frequent dressings and debridement in this case afforded an opportunity to observe the clinical condition of the wounds. In retrospect, however, it is evident that this treatment did not control the pain or prevent the absorption of toxic breakdown products. The experience gained from the study of the hands of Case 1, however, permitted a realistic evaluation of the nature and extent of tissue injury in Case 3 at the time of the development of the first blister. Effective removal of the dying tissues with minimum trauma to the patient, i.e., by refrigeration seemed to be the treatment of choice.

It should be emphasized at this point that prolonged refrigeration of the skin below 15 to 18°C causes an inflammatory type of tissue reaction (Lewis, 1942), while chilling tissues below -6°C even for a very short time causes tissue necrosis (Lake, 1917). Therefore, drastic measures such as were used in Case 3 can be employed early in the course of a radiation injury only when the extent of subsequent radiation injury can be anticipated. Accurate clinical predictions can be made in the early stage of the injury in the absence of good data about dosage only if the nature of damaging radiation is known. For example, the prompt swelling and erythema of the skin caused by beta radiation have a much better prognosis than does a comparable early reaction due to gamma rays. Prolonged refrigeration below 18°C of a beta-ray injury which shows early swelling and blistering would therefore be contraindicated.

The use of less extreme refrigeration in the treatment of a local radiation injury has been suggested because of certain experiments reported in the literature prior to the occurrence of these accidents. Drastic temperature reduction (to about 5°C) of newborn rats before exposure increases their ability to survive large doses of x-rays (Lacassagne, 1942). Lowering the skin temperature in humans before exposure, for example, does not definitely diminish the effectiveness of x-rays in producing erythema (Evans and Kerr, 1943), but refrigeration of newborn rats raises the skin erythema dose by a factor of from 2.2 to 5.0 (Evans et al., 1941). Although refrigeration in all of these experiments had been employed before exposure, it was suspected that lowering the temperature (and the metabolism) of an irradiated tissue after exposure might also prove beneficial. Experimental evidence which has accumulated since the accidents, however, indicates that temporary chilling of adult mice and rats and prolonged chilling of frogs has no effect on the mortality rate of the

animals (Hempelmann, unpublished data; Patt and Swift, 1948-a). Refrigeration of thermal burns in humans is reported to decrease edema formation and diminish infection, but it also delays healing. It is not recommended, therefore, for general use in the treatment of thermal burns, except as a temporary measure to control pain, because the dangers of prolonged refrigeration are not thoroughly understood (Langohr et al., 1949).

It would seem, then, that there is little evidence to support the belief that lowering of the temperature after exposure has a specific beneficial effect on a radiation injured tissue. Nevertheless, as has been pointed out, refrigeration of irradiated or thermally burned tissues (of limited volume) is of great assistance in controlling pain. It also diminishes bacterial growth and has the theoretical advantage of decreasing the rate at which the products of cell disintegration enter into the general circulation. Not too extreme chilling (above 18°C) of the injured tissues, therefore, might help the patient survive the period of acute systematic reaction. If large volumes of tissue are chilled, it must be remembered that there is danger of reducing the body temperature of the individual (Langohr et al., 1949).

6. Summary

In summarizing the discussion of the clinical response of the severely injured hands in Cases 1 and 3, it can be said that the reactions are compatable with the prompt and extensive destruction in all soft tissue which unquestionably was caused by the large radiation doses. The speed with which cell death occurred differentiates these injuries from the more slowly progressing injuries caused by smaller doses of radiation. Tissue death due to impairment of blood flow appears to have been delayed for more than a week. Refrigeration of the injured tissues was helpful in controlling pain and may have diminished the constitutional reaction in Case 3.

B. Reaction of the Skin and its Appendages

The response of the skin of the face and abdomen in Case 1 and of the skin of the abdomen in Case 3 conforms much more closely in time and character to the response observed when limited areas of the body are treated intensively with x-rays. The appearance of erythema of the abdomen of Cases 1 and 3 on the third day is not unusual for a third degree x-ray reaction. The gradual development of epidermolysis of the abdomen in Case 1 over a period of three weeks suggest that death of the epidermal cells of the abdomen was for the most part, delayed in character, in contrast to the immediate cell death which occurred in the tissues of the hand (Section III, B).

1. Skin Reaction of Case 1

Although less striking in appearance than the hand injuries, the skin reaction of the torso of Case 1 present some interesting features. First of all, the severity of the skin reaction in the groins agrees with the clinical observation that the skin of different parts of the body shows varying degrees of radio-sensitivity. It has been shown that the skin of the groin and axilla is the most sensitive to radiation, while the skin of the palm of the hand and sole of the foot is the least sensitive (Hermann and Pack, 1944). Secondly, the epilation of the hair of the temples and the beard leaving the eyebrows relatively undamaged reflects a similar variation in the sensitivity of the hair follicles to the action of radiation. This has long been known to clinical radiologists. Thirdly, the perifollicular pigmentation noted around the edges of the burned area in Case 1 is an unusual type of skin reaction which has been observed before in a somewhat different form (MacKee and Cipollaro. 1946).

2. Skin Reaction of Case 3

The skin reaction of the abdomen of Case 3 was unusual only in so far as

the degree of the edema and inflammatory reaction of the underlying tissues is concerned. This is consistent, however, with the large dose of radiation delivered to this part of the body. The gradual change in color of the torso from the intensely pigmented (sunburned) and unpigmented shades above and below the waist to the uniform deep red color which was present just before death was spectacular but not unexpected.

3. Skin Reaction of the Survivors

The survivors showed two unusual skin reactions, i.e., hypoesthesia of the left arm of Case 4 and the increased rate of beard growth of Case 5. Changes in sensation are frequently observed in skin which has recovered from a third degree x-ray reaction. Its occurrence in skin which shows no gross evidence of radiation injury is not of clinical importance (R. R. Newell, personal communication). It is this reaction in Case 4 that seems to have been more pronounced than usual, possibly due to the character of the incident radiation or to the method of exposure; it is also possible that the disturbed sensation may be linked to the bizarre type of histological skin changes produced by this type of radiation described in Case 1. The apparent increased rate of beard growth reported by Case 5 also is an important observation. Abnormal hair growth was reported in six-female patients with severe thermal burns as a result of over activity of the adrenal glands (Aub, et al., 1943). The disturbance in adrenal function, known to have occurred in this patient, as well as in the burned patients, was, in all probability, responsible for the abnormal hair growth.

The localized epilation shown by the survivors was not very remarkable when one considers the type and depth dose of the incident radiation (see Section X). The transverse ridging of the nails of Case 4 is a usual consequence of any injury to, or nutritional disturbance of, the nail bed. Its occurrence only in the more heavily irradiated left hand is consistent with the knowledge that ionizing radiation can disturb nail growth (Knowlton, et al., 1949).

4. Summary

The discussion of the reactions of the skin and skin appendages of the more heavily exposed patients may be summarized by stating that they conformed, for the most part, to the well-established pattern of acute x-ray reactions. The most noteworthy responses, observed only in the survivors, consist of the hypoesthesia of the arm of Case 4 in the absence of external evidence of skin damage and the increased rate of beard growth of Case 5. The latter can be explained on the basis of adrenal dysfunction.

C. Systemic Response

The clinical response of these patients is similar to that which has been described in the Japanese injured by radiations from the atomic bomb explosion (Medical Report of Joint Commission). The symptomatology of these Japanese cases has been divided into the following four chronological phases: Phase I, the initial reaction consisting chiefly of prostration and gastro-intestinal reaction; Phase II, a period of relative well-being; Phase III, gradually developing toxemia lasting several weeks and complicated by diarrhea, ulcerations of the oropharynx infection, hemorrhagic manifestations and epilation; and Phase IV, death or prolonged convalescence with eventual recovery. The first phase usually lasted one day, but sometimes persisted for two or more days after exposure. The duration of the second phase variable and in general was inversely proportional to the size of the dose. In the more severely injured patients, the toxic or febrile phase often began within ten days after exposure with severe diarrhea as its most obvious manifestation. In the less seriously injured group, epilation on about the twentieth day initiated the febrile phase.

This was soon followed by the appearance of purpura and oropharyngeal lesions. Diarrhea was not as prevalent in this group of patients as the other symptoms. Statistical analysis of the Japanese data shows that epilation and purpura were more highly associated than other pair of lesions with oropharyngeal lesions as the third most common symptom. Although there was evidence of marked individual variation, the severity of the response in the Japanese cases was, in general, dependent upon the size of the dose they received. As a rule, those cases which had been fatally injured died before the end of the sixth or eighth week after exposure.

The discussion of the systemic response of the present patients will deal first with the initial reaction of the patients, then with the toxemia and the subsequent reactions of individual organ systems.

1. Initial Reaction of the Patients

The initial prostration and gastro-intestinal symptoms are discussed together because they constitute the syndrome known as "radiation sickness", which is so common in patients receiving intensive x-ray therapy. The severity of this syndrome shows marked variation from one individual to another and cannot be correlated with the integral or volume dose. This is demonstrated in the present series of cases by the fact that the initial reaction of Case 1 which was much more severe than that of Case 3 despite the lower radiation dosage of the former. The reaction bears some resemblance to primary shock. There was, for example, a transient fall in blood pressure in the more severely injured patients of this series. Transient hypotension, is also observed in animals, particularly in rabbits, given large doses of total-body radiation. (Prosser, et al., 1947-a). The slight hemoconcentration noted in Cases 1 and 3 does not occur in animals. It was probably due to the dehydration which resulted from repeated vomiting. The cause of the initial reaction has not been clearly established, as can be seen by referring to the sixteen distinct theories of the etiology of radiation sickness listed in a recent article (Shorvon, 1949).

The latent period of relative well-being, from the second to the fifth or sixth day, noted in those patients who showed an initial systemic reaction, was striking in its contrast to the illness of the first day. Although all patients were kept in bed to rest and did not exert themselves during the first few days after exposure, it was apparent that the more seriously injured patients were not well during this period. Weakness and exertional dyspnea were most notable in Case 1. This failure to recover completely from the initial illness was also noted in the Japanese (S. Warren, personal communication). Although many seriously injured Japanese are reported to have walked many miles and to have performed work of other kinds, it is known that they exerted themselves only because of necessity. The latent period of relative well-being in the injured Japanese lasted from a few days up to three or four weeks. It was observed that the longer the latent period, the better was the prognosis in these patients. In Cases 1, 3 and 4, however, the period of well-being lasted less than one week despite marked differences in severity of illness.

2. Toxemia

Toxemia, as judged by fever and tachycardia, occurred only in Cases 1, 3, and 4. Comparison of the temperature and pulse charts of these patients suggests that the toxic phase of the illness began about five or six days after exposure. Cases 1 and 3 had a mild elevation in temperature dating back to the time of admission to the hospital; Case 4 did not run a significant early fever. On the sixth post-exposure day, Case 1 showed a gradual rise in temperature and an abrupt increase in heart rate. Cases 3 and 4 showed a distinct rise in temperature and pulse rate on the fifth and sixth days respectively. These observations suggest that the absence of fever during the second week after exposure to ionizing radiation is perhaps a favorable prognostic sign.

Despite the more rapid progress of the illness of Case 3, his fever was never so high as that of Case 1. This may have been due to the fact that the chilling of the upper extremities, a substantial proportion of the total-body volume, prevented the temperature of Case 3 from reaching its full height. That refrigeration of the extremities can influence body temperature is shown by an experiment with thermally burned dogs. In those dogs, all four burned legs of which were chilled, the fall in body temperature was sometimes so extreme as to jeopardize the life of the animals (Langohr, et al., 1949).

Data about fever in the Japanese patients is meager, particularly for the period immediately after exposure (Medical Report of Joint Commission, Section 2). Nevertheless, there is a record of temperature elevation in a few patients during the first few days. Fever was prominent in the patients who died in the first week or so after exposure. A step-wise increase in temperature until death is reported in many of the fatally injured patients, but all types of fever curves were seen. In the less severely injured patients, fever occurred during the toxic phase of the illness several weeks after exposure. It is not clear from the Medical Report of the Joint Commission whether or not these patients had fever during the second phase of their illness.

The etiology of the fever and toxemia toxicity of patients and animals with the acute radiation syndrome appears to be clear. Early in the disease, exogenous causes such as infection are not clearly present.* It seems apparent, therefore, that the early fever in patients with radiation injury as well as that noted in persons with severe thermal burns most likely represents a response on the part of the body to the products of tissue breakdown. That tissue degradation occurred promptly in these patients is suggested by the early appearance of amino-aciduria. Later in the disease, however, infection, usually by coliform bacilli, plays an increasingly prominent role in the acute radiation syndrome. Thus, the blood cultures of dogs given fatal doses of x-rays over the abdomen were not positive until the fourth day after exposure (Warren and Whipple, 1923). After the fourth day, bacteremia was usual in a series of irradiated dogs (Lawrence and Tennant, 1937). A clear correlation between infection and mortality was observed in irradiated dogs that lived more than two weeks after irradiation (Bennett, et al., 1949; C.P. Miller and C. Hammond, quoted by A. Brues, personal communication). In the fatally injured Japanese, evidence of bacteremia was often found at autopsy (Medical Report of Joint Commission, Section 2). Bacterial lesions (pneumonia, anginal lesions in the oropharynx, and skin postules) were frequent in the group of patients who died between the third and the sixth week after exposure (Liebow, Warren and DeCoursey, 1949).

In the cases reported in this report, there was no evidence of overwhelming septicemia even in the face of agranulocytosis, except possibly in the terminal stages of the illness of these cases. It seems quite possible, therefore, that the fever and toxemia, even in the latter part of these illnesses, was largely endogenous rather than infectious in origin.

3. Gastro-Intestinal Symptoms

a. General Features

(1.) Phases of response: The two distinct phases of gastro-intestinal symptoms seen in Cases 3 and 4 were also observed in the injured Japanese. The first phase occurred as a part of the initial reaction immediately after exposure, while the second, a more generalized reaction of the alimentary tract, became

* In animals, as has been pointed out, rapid multiplication of anaerobic bacteria in injured tissue results in the elaboration of toxins which plays an important role in the development of experimentally produced traumatic shock (Aub, 1944). This may conceivably have occurred in the present cases.

evident after an asymptomatic period of several days. In the Japanese, the second phase was often associated with diarrhea, bleeding from the bowel and necrotizing anginal ulcers in the oral cavity. Cultures of the diarrheal stools showed only a few instances of pathogenic bacteria of the dysentery type and gross blood was present in one-sixth of the cases of one series of patients.

The gastro-intestinal reaction of Case 1, unlike that in the Japanese, may be divided into three phases, even though the second and third phases are not entirely distinct. The first phase was not unusual except for its severity. The second phase, during the second week of illness, was remarkable because of the radiostomatitis which was associated with a moderate degree of paralytic ileus and an essentially non-bloody diarrhea. Radiostomatitis, although common in people receiving therapeutic x-ray treatment for malignancies in the mouth region, was not observed in the injured Japanese, except possibly in a few patients who died ten to fourteen days after exposure (Medical Report of Joint Commission, Section 2). It should be mentioned at this time that the dryness of the mouth experienced by Case 4 two weeks after exposure undoubtedly represented a very mild form of radiostomatitis. The second phase of the reaction in Case 1 showed definite improvement before it merged with the third phase, the necrotizing ulcerations in the buccal cavity of an entirely different character. Although the abdominal distention was limited to the second phase, the diarrhea persisted throughout the latter part of the illness.

(2.) Reasons for phasic response: Although the mechanism responsible for the periodicity in the gastro-intestinal response is not clearly understood, it is interesting to consider which of the three following possible explanations, alone or in combination, might be responsible for the clinical manifestations.

(a.) Direct damage to different portions of the alimentary tract: The variability in the latent period between exposure and response of different tissues has been stressed (Section IV). It is not unreasonable to assume, therefore, that the different types of intestinal epithelium should respond at varying intervals after exposure. Indeed, the autopsy protocols of Cases 1 and 3 indicate that the most intense response of the latter case, 10 days after exposure, occurred in the jejunum and ileum while the only evidence of gross damage in the former instance (24 days post-exposure) was found in the colon. Obviously, a conclusion cannot be drawn from two cases but the possibility of damage of different parts of the intestines occurring at varying time intervals after exposure cannot be excluded as a factor which was responsible, in part, for the repetitive nature of the intestinal response.

(b.) Indirect damage to the gastro-intestinal system: Gastro-intestinal reactions as an indirect manifestation of bodily damage are well-known. Thus, it is possible that reflex mechanisms similar to those which cause nausea and vomiting in appendicitis, pneumonia, and other severe infections may have contributed to the initial phase of the intestinal reaction. Admittedly, such an explanation is unnecessary in Cases 1 and 3 in view of the severe epithelial damage of the small intestine which must have occurred during the first day and thereafter as a direct result of the exposure. However, in the case of patients who experience radiation sickness after localized irradiation of parts of the body other than the abdomen, reflex mechanisms may well play a role in the causation of early nausea and vomiting. Another example of the role of indirect factors in producing damage of the gastro-intestinal tract is the development of the oropharyngeal lesions already mentioned. Such injury is believed to be the result of a number of factors which lead to lowered tissue resistance to infection. Direct local tissue injury is one factor; agranulocytosis and diminished antibody production also are undoubtedly important in the development of these lesions. A third example of possible indirect factors contributing to the gastro-intestinal reaction is the non-specific intestinal manifestations which can occur in any kind of injury. For example, acute duodenal (Curling's) ulcers have been described in patients with thermal burns (Harkins, 1942). Similarly, in a series of

burned patients, one third showed guaiac positive stools (up to 3 plus) on the fifth day after injury (Aub, et al.,1943). Such reactions, which are probably related to the blood flow in the intestines during injury, may have participated in the development of the intestinal lesions in these cases.

(c.) True periodicity of response in radiation-injured tissues: Since the possibility of periodicity in the gastro-intestinal response has been mentioned, it is pertinent to refer to the true cyclic phenomena observed in certain other tissues injured by ionizing radiations. Periodicity has been described in the reaction of the skin and buccal mucosa of humans, and in the liver and marrow response of animals. Thus, three distinct waves of erythema occur in the skin at different time intervals after exposure. The suggestion that the reaction of the blood vessels is rhythmical in nature was first made twenty-five years ago (Miescher, 1925). It is now postulated that the periodicity of skin erythema is due to the action of radiation on different parts of the peripheral vascular system (Borak, 1942). The first erythema is said to be a capillary response; the second, an arterial reaction; and the third, a venous effect. Furthermore, it has been claimed that tumor tissue as well as the mucosa of the oral cavity shows a cyclic response to the action of radiation, although there is not good agreement among investigators as to the interval between periods of retrogression (Coutard, 1935; Friedman and Rosh, 1939). Two waves of degeneration also have been described in irradiated liver tissue (Pohle and Bunting, 1932). Recently, recurring periods of cell destruction and regeneration have been observed in the bone marrow (Bloom, 1947). The causes of the cyclic tissue reactions have not been established. It is not inconceivable, however, that these waves of reaction may be related, in part, to the variations in blood flow resulting from the action of radiation on the blood vessels.

b. Unusual Features of Gastro-Intestinal Response.

There are several features of the gastro-intestinal response which deserve further discussion. The gastric retention of Case 3, the radiostomatitis of Case 1 and the observations of the gastro-intestinal secretions will be considered individually.

(1.) Gastric retention: Extreme gastric retention, presumably due to paralytic ileus during the acute stage of radiation as seen in Case 3, has not been reported previously although intestinal obstruction as a late complication of irradiation of the gut is known (Martin and Rogers, 1942). It is reasonable to assume that this unusual response is the result of the intense irradiation of the abdomen of this patient.

(2.) Radiostomatitis: Stomatitis associated with dryness of the mouth and loss of taste, such as was observed in Case 1, is common in patients who have received intensive x-ray therapy for lesions in the oral cavity. However, the mucosal reaction produced by x-rays of the energy usually employed in this kind of treatment (200-kv peak voltage) is almost always accompanied by a comparable epidermal reaction. High voltage x-rays or gamma rays from radium, in contrast, produce a more severe mucositis than skin response (Friedman and Rosh, 1939). This is explained by the fact that strong tissue ionization occurs further below the skin surface in the case of the x-rays of higher energy. In the present situation, where neutrons of limited range caused the major portion of the tissue damage, the mucosal reaction cannot be blamed on the depth dose. Nor is it possible to explain the reaction by assuming increased scattering of the neutrons by the teeth.

As will be shown later (Section VII), irradiation of the adjacent tissues by the neutron induced radioactivity in the teeth and other bony structures cannot be ignored but probably did not contribute significantly to the total mucosal dose. On the other hand, the intense radiation from the highly radioactive gold inlay in Case 3 caused a localized tissue reaction.

The brevity of the exposure to intense neutron radiation may explain this reaction, since it has been found that the relative intensities of the skin and mucosal response to neutrons depends upon the time interval at which the tissues are irradiated (Stone and Larkin, 1942). Thus, when mouth lesions were treated with high energy cyclotron neutrons (21-Mev maximum energy) more severe reactions occurred in the buccal mucosa than in the overlying skin, provided the fractionated doses were given within a period of twelve to fourteen days. If treatment was protracted, however, the skin effect was more extreme than the mucosal reaction.

(3.) Gastro-intestinal secretions: The presence of combined acid in the gastric secretions of Case 3, on the sixth day after exposure, and the lack of mucus in the stools of all patients are noteworthy. Irradiation of the abdomen of patients and animals with large doses of x-rays is known to cause a diminution of the free and combined acid in the gastric secretions (Desjardins, 1931). The fall in gastric acids occurs before the pepsin content is decreased. In a small group of Japanese patients, the incidence of anacidity and hypoacidity was not unusual. The normal acid content of the vomitus of Case 3 is somewhat surprising therefore in view of the large radiation dose delivered to the abdomen. The absence of increased amounts of mucus in the stools of these patients on the other hand, is distinctly unlike the findings in irradiated animals, where fecal mucus is extremely prominent (Prosser, et al., 1947-a).

4. Cardiovascular System

The circulatory collapse noted in Case 3 is in all probability related to the severe adrenal damage. This will be described in greater detail in Section IX, where the similarity between the pathological findings in the adrenals of this case and in acute infections will be stressed. The circulatory collapse also calls to mind experimental work which emphasized the resemblance between the reaction to radiation and the circulatory collapse observed after intestinal obstruction and thermal burns (Moon, Kornblum and Morgan, 1941). The prostration, vomiting, diarrhea, early leucocytosis followed by agranulocytosis, the hemoconcentration and absence of early fever are similar in dogs injured either by radiation and by ligation of the bowel. Hemoconcentration, however, a prominent early finding in Case 3, does not appear in irradiated animals for several days. The severity of the systemic reaction in experimental animals is said to parallel the amount of damage to the intestinal epithelium (Moon, Kornblum and Morgan, 1941).

The arrhythmias and pericarditis of Case 1 call attention to the possible action of ionizing radiation on the heart. The heart is believed to be very resistant to radiation (Desjardins, 1931). However, functional disturbances in heart action were observed in some of the Japanese patients (Medical Report of Joint Commision, Section 2). Thus, hemic murmurs and accentuation of the second pulmonary sound were reported as well as disturbances in the electrocardiographic pattern (low T waves, depressed ST segments and left axis deviation). Electrocardiographic changes in the T waves are also seen terminally in those irradiated dogs which developed myocardial hemorrhages (Prosser, Painter, and Swift, 1947-b). Extrasystoles but an otherwise normal electrocardeograms were recorded in the case of acute mesothorium poisoning (Hamperl and Roemheld, 1936). Such symptoms are, for the most part, nonspecific and of the same nature as the abnormality of the heart sounds noted in Case 3. The fever and toxemia may well have been responsible, in part, for these functional changes.

Paroxysmal auricular tachycardia and pericarditis, on the other hand, have not been described in humans or animals with lethal radiation injuries. Arrhythmias of this nature, however, are common in persons with Wolff-Parkingson-White syndrome, a defect in the myocardial conductive system, which Case 1 was known to have (White, 1944). It seems probable then that the added strain of the severe illness of Case 1 precipitated the disturbance in rhythm. Therefore, this episode probably

does not represent a direct effect of the radiation. The discussion of the nature and etiology of the pericarditis of Case 1 will be deferred until Section IX.

5. Jaundice

Although there is laboratory evidence of liver damage in animals seriously injured by radiation, no instances of frank liver failure or of gross icterus of blood serum and tissues have been reported. No cases of clinical jaundice were noted in the Japanese even though one instance of elevated serum icteric index was reported (Medical Report of Joint Commission, Section 2). However, biliverdin in amounts which caused a green discoloration of the urine has been observed in dogs which had received lethal doses of plutonium (Schwartz, et al., 1947-b).

In contrast to these observations in irradiated humans and animals, jaundice is sometimes observed in patients with severe thermal burns (Harkins, 1942). The absence of jaundice and other signs of severe liver damage indicates that the functions of the liver cells are not seriously disturbed as a result of exposure to lethal doses of total-body radiation. Perhaps the prolonged period of tissue death with the consequent gradual, rather than sudden, release of degraded products of protoplasm into the circulation allows the liver cells to keep up with the required rate of detoxification. Also, the absence of early hemolysis in the radiation injured individual means that the body is not flooded with bile pigments.

The rapid development of jaundice in Case 3 during the terminal hours of life is a significant observation. It was probably the combined result of liver failure and hemolysis just before death. (The evidence for increased hemolysis is presented in the Section VI). That liver damage existed in Case 3 is shown by the porphyrin excretion studies, by the decreased prothrombin level during the last two days of life, and by the histological observations, see Section IX. The large dose of radiation delivered to the abdomen must have been contributed to this severe degree of liver damage, although liver injury secondary to tissue damage elsewhere in the body undoubtedly was an important factor. It should also be mentioned that Case 1, who received a similar high abdominal exposure, also showed bile in the urine passed during the last day of life.

6. Genito-Urinary System

Studies of the kidney function in these cases were limited to urine output, urinalysis and nitrogen retention determinations. They indicate that a serious interference with renal function occurred only in Case 3. These findings are similar to those noted in the Japanese in whom few disturbances in kidney function were observed (Medical Report of Joint Commission).

Detailed study of the kidney physiology in irradiated dogs by direct (phenol red, creatinine and urea clearances) and indirect methods (blood N.P.N. and water excretion) are indicative of an enhanced renal function during the febrile or toxic phase of the illness (Prosser, Painter and Swift, 1947). It has been suggested that this alteration may be due to increased blood flow, general increase in catabolic processes or to the increase in the plasma: cell ratio. In the terminal stages of the illness of fatally injured animals, kidney failure often occurred.

The testicular damage including the loss of organ weight and aspermia of Cases 1, 3, 4 and 5 will be discussed in Section IX.

7. Weight Loss

The loss of weight noted in the severely injured patients is also observed in animals exposed to large doses of total-body radiation. In dogs, for example, the body weight is decreased when the dose exceeds one third the median lethal dose (Prosser, Painter and Swift, 1947). The weight loss is not rapid during the first week following exposure but is accelerated during the second and third weeks.

Fatally injured animals which survive for several weeks may lose as much as twenty-five percent of their pre-exposure weight. The weight loss cannot be explained solely on the basis of dehydration resulting from diarrhea and vomiting. As a matter of fact, studies of the fluid balance in rats show water retention rather than loss (France, 1946). In the past, the weight loss resulting in "radiation cachexia" was attributed to intestinal injury, as it had been observed in dogs after irradiation of isolated loops of gut (Martin and Rogers, 1942). Recent experiments indicate that decreased food intake and enhanced catabolism are probably the most important factors in the decrease in weight. Thus, dogs have decreased appetite and go into a negative nitrogen balance (Prosser, Painter and Swift, 1947). Protein degradation, as shown by studies in mice with C^{14}-labeled alanine, appears to exceed protein synthesis (Hempelmann, et al., in press). Furthermore, the basal metabolism is elevated in irradiated rats, a factor which presumably contributes to weight loss (Kirchner, Prosser and Quastler, 1949).

In the present series of cases, the amino-aciduria in the more severely injured patients and the high potassium excretion in Case 4 may be explained, in part, by assuming that tissue breakdown was enhanced. Patients with severe thermal burns also lose weight and go into a negative nitrogen balance of a mild degree (Aub, et al., 1943). It is hard to attribute the increased nitrogen output to excretion of degraded tissue protein since the potassium balance of the patients is usually unchanged. The marked elevation in the basal metabolism of thermally burned patients may also contribute to weight loss (O. Cope, personal communication).

8. Hemorrhagic Lesions

Spontaneous hemorrhagic manifestations are such a characteristic finding in humans and animals fatally injured by radiation that their absence in these cases, except in Case 3, deserves comment. In the animals irradiated during the Bikini tests, extravasation of blood which occurred in almost every tissue of the body was the earliest and most consistent gross lesion (Tullis and Warren, 1947). Gingival bleeding followed by hemorrhages into the skin and bowel were observed in many of the injured Japanese who survived for three weeks or more (Medical Report of Joint Commisssion, Section 2). Similar bleeding unquestionably would have occurred in Case 3 had life been prolonged. Studies of the clotting time in the survivors of the second accident indicate that subclinical disturbances in the clotting mechanisms of the blood were present. It is not known whether these patients showed increased bleeding tendency or enhanced capillary fragility. The disturbances in blood clotting will be discussed further in Section VI.

It is interesting to note that the three patients in whom epilation occurred (Cases 1, 4 and 5) did not develop hemorrhagic manifestations, since, as has been pointed out, these two symptoms were highly associated in the Japanese patients. The absence of bleeding in all of these epilated patients suggest that the radiations which were responsible for the damage in these cases had a greater effect on the hair follicles than on the blood clotting mechanism. This was undoubtedly due to the difference in character of the radiations in the two situations.

9. Delayed Weakness and Fatigability

The delayed weakness and fatigability of Cases 4 and 5 after a period of well-being was a notable symptom. Whether the development of weakness was delayed for two weeks or whether it merely became manifest when the patients overtaxed their strength upon leaving the hospital, is not clear. It is difficult to believe that these patients would have misjudged their strength and endurance so completely had weakness not become more pronounced at this time. The prolonged weakness was also noted in the Japanese survivors some of whom had not recovered their sense of well-being by the end of the third month after exposure (Medical Report of Joint Commission, Section 2). This weakness is analagous to the diminution in physical

endurance observed in the mice which survive a median lethal dose of fast neutrons (Stapleton and Curtis, 1946). The loss of vitality in these animals was progressive throughout their lives even though they appeared to be normal in other respects.

Although Case 6 received almost as great a dose as Case 5, he displayed neither loss of hair nor loss of weight and strength. This is another demonstration of the variation in the reaction of individuals to almost the same dose of radiation.

The correlation between weakness and the disturbances in the corticoid-like substance in the urine as well as the need for salt noted by Case 4 and the enhanced beard growth in Case 5 strongly suggests that the weakness in these cases was due to adrenal dysfunction. This is supported by the fact that the abnormality in the corticoid fraction of the urine of Case 5 was still present 30 days after exposure at which time the acute reaction had subsided. More will be said about the significance of the corticoid-like substance in Section VII. The adrenal damage will also be mentioned in Section IX.

10. Absence of Symptoms in Cases 2 and 6 through 10

It is important to emphasize the almost complete lack of symptoms in Cases 2, 6, 7, 8, 9, and 10. One might have expected clinical evidence of injury in Cases 6 and 7, since their radiation dosage differed from that of Case 5 by less than a factor of two. Again, this is an example of the previously mentioned wide individual variation in the response to radiation injury. Previous observation indicates that some patients can tolerate as much as 100 roentgens of whole-body radiation given therapeutically without experiencing symptoms of any sort (Cantril, et al., 1947). The energy absorption of Case 5 is equal to that of 94.6 roentgens of gamma rays (see Section X) indicating that this patient's reaction compared favorably with that of the more resistant patients in the previous series. Since all of these patients were aware of the potential seriousness of their injuries, the fact that none of the patients experienced serious psychosomatic reactions is a tribute to their courage and emotional stability.

11. Treatment

Treatment of the systemic reaction of these patients was entirely supportive, directed at assisting the body to survive the period of acute injury. As in thermal burns, there is no specific therapy for fatally injured tissues. Unlike the situation in thermal burns, there is no surgical or other means of helping to repair injured tissues, except where the radiation injuries are superficial and localized (Brown, McDowell, and Fryer, 1949). Regeneration of an injured tissue is, therefore, completely dependent upon the number of cells which survive, their capacity for regeneration, and the adequacy of the blood supply. At the time of these accidents, the only therapeutic measure with a specific action directed at controlling one of the manifestations of radiation damage was the use of toluidine blue to counteract the increased amount of anti-coagulant in the blood (see Section VI). This was not used in the absence of clinical bleeding.

a. Consideration of Specific Drug and Hormone Therapy

A brief consideration of the drugs and other methods used to treat radiation sickness in patients as well as those tested in animals for their action on prolonging life and in the sparing of certain tissues illustrates how limited are the specific methods of treatment. Almost every conceivable drug, including sedation, sodium chloride, hydrochloric acid by mouth, antihistamine compounds, dramamine, liver extract and vitamins, particularly those of the B complex, have been used in the therapy of radiation sickness of patients receiving x-ray therapy. Each type of drug has advocates who believe in its effectiveness, but the true value of most of the methods, except possibly for the salt and antihistamine derivatives, is not clearly established (Painter and Brues, 1949).

Similarly, saline, vitamins, pentonucleotides, bone marrow and adrenal cortical extracts, nucleic acids, atropine, rutin and associated flavones and body refrigeration have been used in attempts to increase survival or prolong life in irradiated animals (Simmons, et al., 1946; Selle, 1949). Of these, only the administration of physiological saline and antibiotics seem to be unquestionably beneficial to the animals when given after exposure (Painter and Brues, 1949). Aureomycin, which combats infections due to certain of the gram negative intestinal bacteria, appears to be particularly effective in controlling the diarrhea and prolonging the life of irradiated animals (Bennett, et al., 1949-b; Howland, et al., 1949). Similarly, streptomycin and other antibiotics appear to be valuable in combating radiation injury in mice (C. P. Miller, quoted by A. Brues, personal communication). Some reports, however, claim that desoxycorticosterone and rutin increase the duration of life of irradiated animals (Ellinger, 1948; Field and Rekers, 1949), but the value of these drugs is questionable (Straube, et al., 1949; Cronkite, et al., 1948-a). Recently, atropine given one day before irradiation and daily thereafter has been shown to reduce the radiation mortality in mice (Larkin, 1949).

There are a number of substances which, if given before exposure, will protect against radiation effects. Of these, cysteine produces one of the most dramatic results (Patt, et al., 1949-b), but glutathione (Chapman, et al., 1949), as well as vitamin C, pentose nucleotides, and nicotinamide (Painter and Brues, 1949) also increase the life span and survival rate if given prior to irradiation. Estrogens (estradiol benzoate) given nine days before irradiation also decreases the mortality rate, presumably because of their stimulating effect on myelopoietic tissue (Treadwell, Gardner and Lawrence, 1943; Patt, et al., 1949-a). Similarly, a limited study suggests that pre-exposure injections of foreign protein offers some protection to mice exposed to doses of radiation in the lethal range (Graham and Graham, 1949). Recent experiments indicate a seemingly paradoxical finding in that previous exposure of animals to sublethal doses of radiation shortly before administration of a lethal dose also increases the survival rate (Cronkite, et al., 1948-b).

b. Physiological Considerations

It has been shown that protection of a portion of the hematopoietic tissues greatly enhances the ability of mice to survive radiation (Jacobson, et al., 1949-b). Shielding the spleen of anesthetized mice raises the 30-day median lethal dose from 600 roentgens in control animals to 1200 roentgens in the protected animals. The splenic tissue so protected loses its lymphoid elements, but promptly begins active myelopoiesis and erythropoiesis. Anemia does not occur in the shielded mice and there is prompt recovery from leucopenia.

c. Practical Considerations in the Treatment of Patients with the Acute Radiation Syndrome

Two recent review articles consider the best available methods of therapy of patients with the acute radiation syndrome (Painter and Brues, 1949; Cronkite and Chapman, 1949). They conclude that good nursing care and hygiene, maintenance of the fluid and acid-base balance and blood volume, and the liberal use of transfusions, antibiotics and the anti-hemorrhagic drugs are the only methods of treatment of proven value. It should be pointed out that the antibiotics of choice are those which affect gram-negative organisms of the coliform series, and that a mixture of antibiotics will probably be more effective in successfully combating the numerous strains of intestinal bacteria.

The difficulty in maintaining an adequate caloric intake and of administering intravenous medication to these patients is considerable. Fat, sugar and amino-acid absorption from the intestinal tract is delayed so that nutrition may not be good even though food intake is maintained (Martin and Rogers, 1924; Barron, Wolkowitz and Muntz, 1947; Altman, et al., 1949). If it is necessary to resort to transfusions or

other intravenous medication, one may encounter extreme difficulty particularly during the terminal stages of the illness because of circulatory collapse and an apparent increase in the fragility of the superficial veins. These factors must be taken into consideration in planning the treatment of a patient suffering from radiation injury.

12. Summary of the Systemic Reaction

The systemic reaction of the more severely injured patients followed the usual four phases of the acute radiation syndrome: (a) the initial reaction, (b) the period of relative well-being, (c) the toxemia associated with the gastro-intestinal reaction, cardio-vascular symptoms, fever, bleeding, and, (d) the eventual recovery or death. Prostration, nausea and vomiting, mild fever, transient hypotension and slight hemo-concentration were observed during the initial reaction. During the second phase of relative well-being, some degree of prostration and a mild fever continued in the severely injured patients. The acute toxic response began at the end of the first week in Cases 1, 3, and 4 with elevation of temperature and pulse rate. The gastro-intestinal reactions in Case 1 during the toxic period consisted of radiostomatitis, nausea, mild paralytic ileus followed by diarrhea and the development of therminal necrotic mouth lesions. Case 3 showed extreme gastric retention and moderate diarrhea. Case 4 suffered nausea and vomiting during the toxic period. The cardiovascular symptoms in Case 1 during the stage of toxemia consisted of paroxysmal auricular tachycardia followed by pericarditis. Case 3 showed a terminal circulatory collapse. Case 3 also showed terminal jaundice and skin hemorrhages. All of the more severely injured patients lost weight. During the fourth phase, two of the survivors experienced delayed fatigability and weakness of a severe degree.

The possible roles played by the immediate enhanced tissue degradation, adrenal dysfunction and infection in the symptomatology of the acute radiation syndrome are discussed. The periodicity of tissue reactions caused by ionizing radiation is also considered. The similarity between the systemic reactions in these patients and those in thermally burned patients is stressed. The treatment of the acute radiation syndrome is briefly discussed.

D. Addendum - Changes in the Eye of Case 4

The development of a cataract in the left eye of Case 4 three years after exposure may be considered to be a delayed manifestation of the acute radiation syndrome. It is similar in nature to those which have been described in the eyes of persons exposed to ionizing radiations of various types. Such lesions usually take the form of an initial subcapsular haze at the posterior pole which gradually develops into opacities, frequently disc-shaped and clearly demarcated from the rest of the cortex. The latent period between acute exposure of the eye to x-rays and the development of the opacities is usually between six and twenty-four months, but may be as long as eight to twelve years (Cogan, 1950). The latent period between exposure and diminution of vision in the case of the Japanese with radiation cataracts was less than one year in one instance, and from 2 1/4 to 2 1/2 years in five others (Cogan, Martin and Kimura, 1949). The cataracts in cyclotron workers which are presumably due to chronic neutron irradiation were detected about three years after exposure began (Abelson and Kruger, 1949). Radiation cataracts may be stationary but are usually progressive. They lend themselves to satisfactory surgical removal.

The minimal acute radiation dose which will cause a cataract when delivered locally to the eye has not been established with certainty, but is of the same order of magnitude as the erythema and epilation dose (Hunt, 1947). Thus, 875 roentgens of 130-kv x-rays and 1000 roentgens of gamma rays are reported to have caused cataracts. In the ten Japanese with visible changes in the lens, all showed epilation one to four weeks after exposure. In contrast to acute exposures, chronic or repetitive irradiation is believed to be more effective in producing permanent eye changes

than in causing a visible skin reaction (Cogan, 1950). Chronic exposure to neutrons has been shown to be highly cataractogenic in mice (Evans, T. C., 1948), and is believed to be equally effective in humans (Abelson and Kruger, 1948). The R. B. E. of neutron and gamma rays administered in this manner in man has been estimated to be 10 to 40.

The pathogenesis of radiation induced cataracts is not understood. It is conceivable that the extremely slow growth rate of the lens epithelium and therefore, the long life span of these cells may have something to do with the prolongation of the latent period between exposure and damage in the manner mentioned in Section IV-B. Just why the damage should be more extreme in the posterior pole is not clear since the fibers making up the bulk of the cortex are derived from the epithelium and extend from the equator to both the anterior and posterior poles.

482 35

VI. DISCUSSION OF HEMATOLOGICAL FINDINGS

It is important to consider the blood changes carefully, since this response is one that is readily produced by ionizing radiation. The reader is referred to the reports by Jacobson and Marks (1947), Lawrence, Dowdy and Valentine (1948), to the review by Dunlap (Warren, 1942), and to the article by Goodfellow (1936), for detailed references to observations on blood changes in irradiated humans and animals.

A. Methods

All pipettes and hemocytometer chambers were certified by the National Bureau of Standards. The total erythrocyte count was determined by counting two sides of a chamber filled with blood diluted and mixed in a single pipette. Four chambers of diluted blood from two pipettes were examined to obtain the total leucocyte count. The hemoglobin content of the blood was determined by the oxy-hemoglobin method of Evelyn (Evelyn and Malloy, 1948). The method of Dameshek (1932), was used to estimate the number of platelets and reticulocytes. Supravital preparations and dried smears of blood stained with Wright's stain were examined for cytologic changes. The usual supravital staining technicque was modified by increasing the strength of the dyes (Dickie and Hempelmann, 1947). The pre-accident differential counts were obtained by examination of two hundred fixed cells and one hundred leucocytes in supravital preparations. Two hundred leucocytes in supravital preparation were counted in order to determine the post-exposure hemograms. Blood smears stained with Wright's stain were not used in the quantitative studies of blood cells after the accidents. The number of refractive neutral red staining cytoplasmic bodies in fifty lymphocytes was determined in each examination. All blood samples prior to the accident were taken between 8:30 and 10 o'clock in the morning. The first blood samples after exposure were obtained within two hours of the accidents in all cases. Subsequent samples were taken twice daily (9 to 10 o'clock in the morning and 7 to 8 o'clock in the evening) for the first few days after the accident, and then once daily except for Case 1 on whom two blood counts per day were done throughout the entire illness.

The bone marrow specimens were obtained by aspirating tissue from the sternal cavity through a 15-gauge needle. Supravital preparations and Wright-stained smears of the clumps of aspirated marrow were studies for cytologic changes. The differential counts, however, were based only on examination of the supravital preparations.

The whole blood-clotting time was determined by the method of Lea and White (1913). This technique was repeated in whole-blood samples to which small quantities of an aqueous solution of toluidine blue were added in order to determine the amount of heparin-like anticoagulant in the circulating blood (Allen, et al., 1948). The prothrombin time was determined by the one-stage Quick procedure (Allen, Julian and Dragstedt, 1940).

B. Changes in the Blood Count

Graphs showing the changes in the formed elements of the blood during the period immediately after exposure accompany each case report in Section III, (Vol. I). However, only a limited amount of information can be gained from the study of these graphs because of their size and the way in which they are plotted. In order to present the hematological data in more detail, a statistical analysis of the changes in leucocyte and erythrocyte counts is provided in Appendix I (Vol. III), and the individual lymphocyte and neutrophil counts for the first two-week period after exposure are listed in Appendix II (Vol. III). The blood counts of Cases 1 and 3 were not analyzed statistically because the changes were obvious. Because the

blood count may fluctuate considerably under normal conditions, even in the same individual, the difficulty in interpreting the changes, even after the use of statistical analysis, is obvious. The interpretation of the hematologic response is complicated further by the fact that no pre-exposure counts were available for two of the patients. Nevertheless, an attempt is made to evaluate the changes by applying statistical methods to groups of blood counts in a single individual over arbitrary intervals of time. The time intervals used in the analysis of the rapidly changing leucocyte count were usually shorter than those used for the more slowly changing erythrocyte count. The deviations of these averaged counts from the pre-exposure mean (when known), and from the range of normal values (Wintrope, 1943), is studied. Wherever possible, the trend or pattern of the deviation is determined.

1. Lymphocyte Response

a. Development of Lymphopenia

Examination of the statistical data on the lymphocyte counts in Appendix II shows that lymphopenia occurred in all patients except Cases 2 and 10. The severest degree of lymphopenia in Cases 4, 5, and 6 occurred between the third and the tenth or fifteenth day. The lowest averaged lymphocyte count occurred in Case 7 during the thirtieth-to-the-seventieth-day period. The prompt and sustained reduction in the number of circulating lymphocytes noted in most of the present series of patients is understandable, since lymphoid tissue has long been known to be among the most radiosensitive in the body.

The speed with which the fall in the blood lymphocytes occurs (15 to 30 minutes in animals) has been explained by the short life span of the adult circulating cells (12 hours or less), by the limited reserve of this type of cell, and by the immediate destruction of the parent cells in the lymphoid tissue (30 minutes) Lawrence, Dowdy and Valentine, 1948).

It has been shown experimentally in humans and animals that reduction in the lymphocyte count occurs even after acute exposure of the individual to relatively small doses of total-body radiation. Because of more complete studies, lymphopenia of statistical significance has been demonstrated with smaller doses in the case of animals than in man. For example, 25 roentgens of x-rays causes a transient lymphopenia (Jacobson and Marks, 1947), while total-body irradiation of humans with divided doses of x-rays produces a fall in lymphocytes when the accumulated dose reaches 40 roentgens (Low-Beer and Stone, 1948).

A correlation between the early rate of fall in the lymphocyte count and the integral dose of radiation was first described in 1936 (Goodfellow). The relationship is best shown when the blood counts of groups of people are studied collectively, for it often cannot be demonstrated in a single individual (Ellis, 1942). Although the early rate of fall of the lymphocyte count is dependent upon the dose, the ultimate level to which the lymphocytes fall is not always proportional to the dose. The curve of the lymphocyte response with time has been worked out carefully for gamma radiation delivered therapeutically over a period of several days (Price, 1948). Following exposure, the curve can be broken down into two component parts: the first, an early rapid fall that continues from seven to ten days after the beginning of the exposure; and the second, a diminished rate of fall that persists thereafter. Similarly, if the lymphocyte count is plotted against the "volume" dose, a comparable pattern of the lymphocyte response can be shown. There is a rapid drop, per unit of radiation, during the initial application of the radiation; as more radiation is delivered, however, the reduction in lymphocytes is less for the same increment of radiation (Bush, 1943; Price, 1948). Thus, the administration of 20 megagram roentgens of gamma radiation to the pelvis in a series of women caused a fall in the mean lymphocyte count from 2400 to 550 cells per mm^3, or a decrease of 1850 cells per mm^3 An additional 20 megagram roentgens (total, 40 megagram roentgens) caused only a

fall to 200 cells or a decrease of 350 cells per mm^3. Each additional dose of the same magnitude produces a correspondingly smaller drop in the lymphocyte level.

b. Recovery of the Lymphocyte Count

It can be concluded from a study of the survivors of the present accidents that the return to the normal lymphocyte level in the blood after irradiation is usually an extremely slow process. The lymphocyte count of Cases 4 and 6 were still depressed below the pre-exposure level during the second year after exposure. That of Case 5, on the other hand, was low for only 60 days after exposure.

A similar slow recovery of the lymphocyte count was noted in patients given a therapeutic doses of 60 to 120 roentgens (Cantril, et al., 1947). In some of the cases that were followed for 150 days after exposure, the lymphocyte count still had not returned to the pre-exposure level. The lymphocyte count in many of the Japanese patients also was depressed at the conclusion of the study three months after exposure (Medical Report of Joint Commission, Section 6). In contrast to these observations in man, recovery of the lymphocyte count is much more rapid in animals. For example, in rabbits, the lymphocyte count returns to pre-exposure levels within 60 days even after exposure to 300 roentgens of x-rays (Jacobson, and Marks, 1947). It is of interest that, in both animals and man, the regeneration of parent lymphoid tissue is much more rapid than is the return of the lymphocyte count to normal level. This lack of correlation between the rate of recovery of the blood and tissue lymphocytes is discussed further in Section IX.

c. Cause of Radiation Lymphopenia

Lymphopenia is found in a number of conditions unrelated to radiation injury. Thus, severe infection, advanced Hodgkin's disease, and tuberculosis, chronic leukemias other than the lymphatic type, subleukemic lymphatic leukemia, and advanced agranulocytosis of any cause show varying degrees of reduction of the lymphocyte count (Sturgis, 1948). No significant change in the lymphocyte count occurs in persons with thermal burns even though generalized involution of the lymphoid tissue has been described in this as well as in other types of injury (Pack, 1926).

The lymphopenia following radiation has long assumed to be the result of the direct destructive action of radiation of the parent lymphoid tissue. Indirect factors originating outside the lymphoid tissue and transmitted through the blood stream have been searched for and not clearly demonstrated (Lawrence, Valentine and Dowdy, 1948; Barnes and Furth, 1943). Recently, the possibility has been suggested that an indirect effect of radiation on lymphoid tissue is mediated through the adrenal and pituitary glands (Dougherty and White, 1946).* This postulation, which is of particular interest because of the adrenal response to irradiation injury mentioned in Sections V and VII, is based on two experimental findings. The first is that, although large doses of x-rays (200 roentgens) cause the same degree of lymphopenia and involution of the lymphoid tissue in normal and in adrenalectomized mice, small doses (10 roentgens) have an effect on the lymphatic elements only in those animals with intact adrenal glands. The second observation supporting this theory is that lymphopenia and involution of lymphoid tissue can be demonstrated in mice, rats,

* For many years the adrenal gland has been suspected of influencing the circulating lymphocytes and lymphoid tissue since an increase in these cells has been observed in Addison's disease and in adrenalectomized cats (Drinker, Kent, and Yaffey, 1941). Attempts to increase lymphocyte production and to raise the lymphocyte count in animals by adrenalectomy, however, are not always successful (Valentine, Craddock and Lawrence, 1948).

rabbits, and dogs following the administration of adrenocorticotropic hormone and adrenal cortical extracts (Dougherty and White, 1944; Reinhard, Aron and Li, 1944; Farr, et al., 1948). It is not clear, however, that this happens in all animal species (Valentine, Craddock and Lawrence, 1948). In man, a single dose of pituitary adrenocorticotropic hormone causes a fall in blood lymphocytes and eosinophils and a marked rise in the number of circulating neutrophils (Hills, Farsham and Finch, 1948). Protracted administration of adrenocorticotropic hormone over a four-day period causes an initial drop in lymphocytes followed by a rise, during the course of the treatment, which exceeds the pretreatment level. It may be concluded from this short review that the role of the adrenal and pituitary in the production of radiation lymphopenia is not clearly understood, but hormonal imbalance due to dysfunction of these glands may contribute to this response.

2. Total Leucocyte and Neutrophil Count

a. Response of the Patients

The day-to-day variation in the total leucocyte count was due largely to changes in the number of neutrophils since the other leucocytes in the circulating blood remained at a relatively constant level after the first few post-exposure days. The following statements, therefore, with few exceptions, can be applied to both the neutrophil response and the total leucocyte count.

All cases showed an initial leucocytosis due to an increase in the number of mature cells of the polymorphonuclear series; in three and possibly four cases, this response was succeeded by leucopenia. The early increase in neutrophils except in Cases 1 and 3, both of whom showed a leucocytosis for several days, was not sustained, and soon gave way to a condition of marked instability of the leucocyte count which showed wide fluctuations ranging from the normal value to two or three times normal. Only Case 9 showed a single count below the lower limit of normal during the first four days after exposure. With certain exceptions (notably, Case 10), the height of the leucocyte count was roughly proportional to the dose of radiation. No constant correlation could be found in the times at which the peaks and troughs occurred in the curves of the leucocyte response of different cases. The initial period of normal-to-high neutrophil counts, which lasted four to five days, was succeeded by a neutropenia in Cases 1, 3, 4, and probably 5. It is significant that young neutrophils were not conspicuous until the leucopenic phase was established. The transient rise in neutrophils which occurred in Cases 1 and 4 between the ninth and fifteenth day is also noteworthy. During this period, there was no increase in the other cellular constituents of the blood, except for the platelets. In Case 4, the only patient with a clear-cut post-exposure neutropenia, the time required for recovery of the neutrophil count was about two months.

b. Initial Increase in Neutrophils

Initial leucocytosis following exposure to lethal or sublethal doses of ionizing radiation has been observed repeatedly in humans and animals. The leucocyte count of the patient with acute mesothorium poisoning remained at a level of about 20,000 cells per mm^3 for the first two days of the illness (Hamperl and Roemheld, 1936). However, such an early leucocytosis does not invariably occur in irradiated humans. Thus, in a series of patients given a single total-body exposure of 60 to 120 roentgens, no increase in neutrophils was observed (Cantril, et al., 1947). Divided doses, up to 500 roentgens, similarly did not cause leucocytosis in another group of patients (Low-Beer and Stone, 1948). Furthermore, two instances of leucopenia rather than leucocytosis were recorded in the few blood counts that were made on fatally injured Japanese during the first twenty-four hours after the atomic explosion (Medical Report of Joint Commission, Section 6).

In most animal species (dogs being a notable exception) leucocytosis occurs

immediately after irradiation, but it is less marked than that noted in these patients and is of shorter duration. Two peaks occur in the neutrophil response of rabbits, one at twelve, the other at twenty-four hours after exposure to either 500 or 800 roentgens of total-body x-rays (Jacobson and Marks, 1947). Rabbits show an "abortive" increase, not only in the number of granulocytes, but also in the number of lymphocytes and reticulocytes, between the fourth and the eleventh days (Jacobson and Marks, 1947).

The initial and the abortive granulocytosis is now interpreted as a reaction to tissue injury rather than the result of the alleged stimulating effect on the bone marrow mentioned in the earlier literature as an etiologic factor in radiation response (Bloom and Jacobson, 1948). Similar and equally prompt granulocytosis is observed in patients suffering from many other related and unrelated diseases. For example, within three hours after injury a total white cell count of over 20,000 cells per mm^3, largely due to an increase in the granulocytes, is not unusual in patients with severe thermal burns (Medical Research Council, 1944). The granulocyte level in such patients falls to normal or subnormal within two days. A secondary granulocytosis may occur at the end of the first week, presumably as a response to the infection of the burned surfaces. Early granulocytosis is also noted in patients with circulatory collapse due to intestinal obstruction (Moon, Kornblum and Morgan, 1941).

In interpreting the early granulocytosis in the present cases, one must remember that the patients were apprehensive during the period immediately following the accidents since all realized the potentialities of exposure to radiation such as they had received. Therefore, emotional factors cannot be excluded as factors that may have influenced the initial response of the formed elements of the blood.

c. Neutropenia

The neutropenia that occurs in patients severely injured by radiation is well known. It is a matter of considerable interest, therefore, that none of the patients exhibited unquestionable neutropenia except those most heavily exposed, i.e., Cases 1, 3, and 4. In Case 4, the clear-cut neutropenia was maximum during the twentieth- to fortieth-day post-exposure period and returned to the pre-exposure level during the fifty-fifth to seventieth-day interval. The neutrophil count in Case 5 fell to a level below the lower limit of normal (3000 cells per mm^3) during the twentieth to eightieth day post-exposure period. This probably represents neutropenia for this patient, but one cannot be absolutely certain of this since only two of the three averaged counts during this period fall outside one standard deviation of the mean of the pre-exposure average counts (see Appendix I). The neutrophil count of Case 6 did not fall so low as the pre-exposure level after the initial elevation, while that in Cases 7, 8, and 9 returned to low normal (3000 to 4000 cells per mm^3); only two of these three patients had blood counts before exposure, and, in each instance, the neutrophil count after the initial rise did not fall significantly below the pre-exposure mean.

The early blood counts made on the injured Japanese are so few that they contribute little to the information about the initial blood-cell response. Considerable data are available, however, about the duration of the leucopenia and the time of recovery. The lowest leucocyte count of the fatally injured patients was almost always less than 500 cells per mm^3, while that of the survivors was usually above this level. The maximum degree of leucopenia in the Japanese survivors occurred between the third and fourth week. The neutrophil count began to rise shortly after this time and was usually normal by the end of the eighth week. Thus, the time response of the neutrophils noted in Case 4 of the present series conforms well with that observed in the Japanese.

In interpreting the neutrophil response, as well as that of the other formed elements of the blood, it is well to keep in mind certain animal experiments that may be likened to the present conditions of exposure. It has been shown that

protection of part of the blood-forming tissues in mice minimized the fall in some of the formed elements of the blood (Jacobson, et al., 1949-b). In animals so protected, the degree of the neutropenia is not influenced, but recovery of the neutrophil count occurs more promptly than in unprotected animals. The limited range of the incident neutrons means that some of the deep-seated hematopoietic tissues must have received a relatively small dose of radiation, thereby making their exposure conditions similar to those produced in experimental animals. Similarly, in considering the precipitous drop in the neutrophil counts of Cases 1 and 3, one should recall the theory that correlates the rate at which the fall in neutrophils occurs with the life span of these cells in the circulating blood of animals (Lawrence, Dowdy and Valentine, 1948). In the cat, eighteen hours elapses between the time at which the neutrophil count begins to fall and the time at which the lowest granulocyte count is reached. This time is believed to be equal to the life span of circulating neutrophils. It is postulated that the reason for the delay of a day or so in the appearance of neutropenia is the good reserve of this type of cell in the vascular system.

Before attaching too much significance of a specific nature to the neutropenia in the terminal stage of the illness of fatally injured patients, it should be emphasized again that agranulocytosis may occur terminally in patients with other unrelated illnesses (thermal burns, intestinal obstruction, etc.).

3. Eosinophil Count

Study of the statistical data shows that there was a fall in the eosinophil count of the four most heavily exposed patients. In Case 4, the period of lowest eosinophil count (50 to 100 cells per mm^3) occurred between the twentieth and the seventieth day, while, in Case 5, it fell between the first and thirty-sixth post-exposure day. Of the six other patients, Cases 6 and 8 showed a definite eosinophilia, Cases 2, 7, and 9 had eosinophil counts over 250 cells per mm^3, while the eosinophil count of Case 10 showed no change over the pre-exposure level. The eosinophilia in Case 6 was striking, ranging between 550 and 750 cells per mm^3 in the period between two and twelve and one-half months after exposure. It is impossible to say that the eosinophil counts over the upper limit of normal (250 cells per mm^3) in Cases 2, 7, and 9 represented a true increase in eosinophils since, in two of these cases, there were no pre-exposure counts and, in the other one, the average pre-exposure count was high normal (200 to 300 cells per mm^3).

Eosinophilia is said to be an inconstant finding in persons chronically exposed to ionizing radiation (Goodfellow, 1936). Many Japanese showed a relative increase in the percent eosinophils (more than 10 percent of the total leucocyte count) particularly in the period from eight to twelve weeks after exposure (Medical Report of Joint Commission). The percentage of eosinophils was roughly proportional to the dose in the survivors of each city. However, the eosinophilia was much more striking in the patients receiving apparently comparable radiation doses in Nagasaki than in those in Hiroshima. In contrast to these observations, eosinophilia was not observed in recent human or animal studies of radiation effect (Jacobson and Marks, 1947; Cantril, et al., 1947; Low-Beer and Stone, 1948).

4. Monocyte Count

It can be seen from the curve of the monocyte response in Cases 1 and 3 that the former did not show a striking charge in the monocyte count even in the terminal stages, while the latter showed a definite monocytopenia before death. Consideration of the statistical data on the blood counts of the survivors indicates that the monocyte count of Case 4 fluctuated considerably after exposure, but showed no sustained deviation from the pre-exposure mean. The monocyte level in Cases 5, 6, and 10, on the other hand, showed a significant sustained increase over the pre-exposure averaged count, while, in Case 9, the monocyte count increased temporarily after exposure. In Case 5, the pre-exposure averaged monocyte count was quite low

(150 cells per mm^3) and only three of the eight groups of post-exposure counts were above the upper limit of normal (500 cells per mm^3). In Case 6, the maximum degree of monocytosis (900 to 1100 cells per mm^3) occurred between the third and thirtieth day. There appears to be a probable elevation of the monocyte count in Cases 7 and 8. In Case 7, which had no pre-exposure count, the mean of the monocyte count showed a steady downward trend from an initial post-exposure high of 980 cell per mm^3. The average monocyte count in Case 8 fluctuated, but four of the six counts were more than one standard deviation of the mean above the pre-exposure level. In contrast to these patients who showed a significant change after exposure, Case 2 showed a constant monocyte count which was considerably above the upper limit of normal; in the absence of pre-exposure counts, one cannot say that this represents an increase.

In all of the survivors, except Case 4, the averaged monocyte counts taken during the first three post-exposure days were high with extremes of 650 and 1100 cells per mm^3. The apparent increase in monocytes shown by many of these survivors is interesting in view of the delayed monocytosis described in a series of patients with rheumatoid arthritis treated by total-body irradiation (Low-Beer and Stone, 1948).

5. Basophil Count

It is impossible to draw definite conclusions about the basophil counts in the more severely injured patients because these cells may have been confused with toxic neutrophils. This is unfortunate, particularly in Case 3, in view of the large number of tissue mast cells found at autopsy in the irradiated tissues (see Section IX). In the other patients, the basophil count is also difficult to evaluate on a statistical basis because of the normally low incidence of this type of cell. However, basophils did not disappear in Cases 4, 5, and 6, although they may have been diminished in number. The question has been raised as to whether many of the so-called "toxic" granulocytes are not actually abnormal basophils, since many precursors of this type of cell have been seen in the marrow of patients subjected to total-body radiation (L. O. Jacobson, personal communication).

6. Erythrocyte Response

A gradual fall in the erythrocyte level associated with a comparable drop in hemoglobin occurred in Case 1 after the first week. Case 3 did not show anemia, perhaps because of the many transfusions. Cases 4 and 6 showed a decrease in red blood cells of probable statistical significance which was most pronounced forty to seventy days after exposure and which lasted six to eight months (Appendix I). The drop in hemoglobin level in these cases was relatively less than the decrease in red blood cells. The limited studies of the changes in cell volume and cell hemoglobin content indicated that the relative anemias in Cases 4 and 6 during the third week after exposure were of the macrocytic and hyperchromic type. The study of the red blood cell response of Case 5 is purposely omitted from this discussion because of the confusion introduced by the fact that the patient moved from a high to a low altitude shortly after the accident. In Cases 7 and 8, the only other patients whose erythrocyte counts were analyzed statistically, there appeared to be no significant change after exposure.

In addition to the observed fall in erythrocyte count, it should be mentioned that a slight rise in erythrocyte level occurred in Cases 1, 3, and 6 shortly after exposure. In Cases 1 and 3, this may be explained on the basis of dehydration; its cause, in Case 6, is obscure. Case 5 showed an increase in hemoglobin during the first week after exposure, but no variation in the erythrocyte count.

Anemias of a macrocytic and hyperchromic type are a classical finding in persons repeatedly exposed to ionizing radiation from an external source and in

patients in the body of whom radioactive materials have been deposited (Warren, 1942). The anemia that occurs in animals after acute exposure to x-rays is normochromic (Jacobson and Marks, 1947). Patients exposed to 150 or more roentgens in divided doses show a macrocytic anemia which reaches a maximum two hundred days after treatment (Low-Beer and Stone, 1948). The time of maximum anemia in Cases 4 and 6, as pointed out previously, occurred much more promptly. The relatively early anemia in these cases is in accord with the observations in the Japanese survivors in whom the lowest erythrocyte counts were obtained between the sixth and seventh weeks (Medical Report of Joint Commission, Section 6). The anemia of the Japanese patients was also of the macrocytic and hyperchromic type. It must be remembered, however, that the anemias in the Japanese, which sometimes were as marked as 1,000,000 cells per mm^3 may have been due, in part, to infection, thermal burns, malnutrition and other factors unrelated to ionizing radiation.

Normochromic macrocytic anemias, sometimes of a severe degree, frequently occur in patients with other injuries such as in those with severe thermal burns (Medical Research Council, 1944). The anemias of burned patients are believed to be due to the rapid destruction of the excessively fragile erythrocytes. Some experimental work suggests that this type of hemolysis is due to structural changes in those red blood cells that have been injured by being subjected to the rise in temperature at the time of the injury (Shen and Ham, 1943). Indirect evidence in animals indicates that there may also be a hemolytic factor in radiation-induced anemias (Schwartz, et al., 1946). This has been proposed on theoretical grounds in order to account for the rapid development of terminal anemia, since, in the absence of gross bleeding, the fall in red blood cell count in animals is too rapid to be explained solely on the basis of the inhibition of erythropoiesis. Studies of the urobilinogen excretion support, but do not prove, the occurrence of hemolysis. Since fragility of erythrocytes in dogs is not disturbed (Prosser, Painter and Swift, 1947-b), the mechanism underlying the development of possible hemolysis in the acute radiation syndrome is not clear.

7. Reticulocyte Count

The reticulocyte count of Case 1 showed no change even in the terminal stage of the illness; that of Case 3 showed a gradual fall. Cases 4 and 5 exhibited a gradual increase in reticulocyte count during the latter part of the first month after exposure. The failure of the reticulocyte count to fall in Case 1 is unexpected in view of the consistent decrease noted in irradiated animals (Jacobson and Marks, 1947; Lawrence, Dowdy and Valentine, 1948); it may possibly be explained, however, by the distribution of the radiation dose which resulted in the low exposure of some portions of the marrow, as has been emphasized previously.

8. Platelet Count

A fall in the platelet count was observed in Cases 1, 3, 4, 5, and possibly 6. Except in Case 3, the decrease did not occur before the third week. The platelet count of the survivors did not fall to the level usually associated with spontaneous bleeding.

9. Summary

All patients showed an increase in the neutrophil count immediately after exposure. In the two most severely injured patients, both of whom had extensive destruction of the hand tissues of a prompt nature, the marked neutrophilia was sustained for several days. All other patients showed a fluctuation of the neutrophil count from normal to moderately high levels during the first few post-exposure days. The initial neutrophilia was succeeded by neutropenia only in Cases 1, 3, 4, and probably 5. Lymphopenia of statistical significance appeared in seven of the patients. In two of the survivors, the lymphocyte count did not return to pre-exposure level until after the second post-exposure year. A fall in eosinophil count occurred in the

482 43

four most heavily irradiated patients; eosinophilia occurred in two and possibly five of the remaining six patients. A fall in monocyte counts occurred only in Case 3. In the survivors, all showed an initial monocytosis. In four of these eight patients, a definite increase in monocyte counts was noted and, in two others, there was an increase of probable significance.

Three patients showed an early rise in erythrocyte count and hemoglobin level. One fatally injured patient and two survivors showed a fall in red blood cell count. The relative anemias in the cases of the survivors were hyperchromic and macrocytic in nature. Except in Case 3, there was no decrease in the reticulocyte counts of these patients, but four and possibly five patients showed a fall in platelets.

C. Cytologic Changes in the Blood Cells

1. Lymphocytes

The occurrence of cells in varying degrees of disintegration was noted only in the peripheral blood of fatally injured patients. The lymphocytes of Cases 4 and 5 showed minor cytologic changes while those in the other survivors appeared entirely normal. All the degenerative changes seen in the circulating lymphocytes could also be observed in the histologic sections of tissue lymphocytes. These changes consisted of enlargement of the cell, clumping of the chromatin, vacuolization of both nucleus and cytoplasm, and some instances of nuclear pyknosis. It is interesting that a particular type of change could be confined to one part of the nucleus. Thus, half the nucleus of some cells was pyknotic or poorly stained, while the rest of the nucleus appeared relatively normal. Perhaps more difficult to explain than the disintegrating cells was the presence of the relatively normal lymphocytes in the blood of Cases 1 and 3 just before death. One wonders where in the body these cells could have been formed, especially in Case 3, in view of the histologic and cytologic picture of the lymphatic tissues after death.

The degenerative changes noted in the lymphocytes of these patients were not reported in the Japanese cases. However, an abnormal type of lymphocyte, similar to those seen in infectious mononucleosis, was seen in the blood smears of the Japanese (Medical Report of Joint Commission, Section 6). These cells, which were large with variable amounts of cytoplasm and with small eccentrically placed nuclei, the chromatin of which was coarse and bunched, sometimes constituted twenty-five percent of all circulating lymphocytes.

Although a few abnormal cells of this nature were seen in the blood smears of Case 1, this type of cellular change was not conspicuous in either the fatally injured patients or in the survivors.

The increase in the number of refractive neutral red staining bodies in the lymphocytes, noted in all but two patients, has been described previously in persons chronically or acutely exposed to ionizing radiation (Dickie and Hempelmann, 1947). This change has been observed recently in a series of four patients with beta-ray burns (Knowlton, et al., 1949). The increase in refractive bodies does not appear to be a specific effect of ionizing radiation, since it is also observed in the lymphocytes of persons exposed to certain toxic chemicals. The neutral red bodies are not seen in alcohol-fixed blood smears since they are alcohol-soluble. Recent evidence indicates that the refractive bodies are lipid in nature; they are birefringent, have a high refractive index, and are increased in numbers by the action of the neutral red component of the supravital dyes on the living cells. It has been suggested that these lipid bodies are associated with the Golgi apparatus (Knowlton and Hempelmann, unpublished data). If this is true, the change in these particulates seems to confirm previous reports that fragmentation and increase in size of the Golgi apparatus occurs in tumor cells after irradiation (Fogg and Warren, 1937). It should be pointed out that the increased number of refractive bodies persisted for more than two years in Case 4 and for over one year in Case 6.

2. Neutrophils

Degenerative changes consisting of swelling of the cells, vacuolization, pallor and swelling of the nuclei were observed in the more severely injured patients. It is interesting to note that two types of disintegration could occur simultaneously in the same cell. Thus, the nucleus of a single cell could show swelling of one lobe and pyknosis of another lobe.

Perhaps the most interesting change in the neutrophils was the marked degree of toxic granulation of the cytoplasm. This appeared promptly (one hour after exposure in Case 1) and lasted until the acute stage of the illness had passed. The granules were similar in appearance to those described in patients with severe infections. The granulation occurred in young and old cells alike, and the depth of staining of the granules was often accentuated by the pallor of the cell nuclei. Histochemical study indicated that the staining properties of the granules were not destroyed by incubating the blood smears with ribonuclease. As has been pointed out, it has been suggested that these cells might be basophils (Jacobson, personal communication). If these granules stained with the metachromatic dyes, this view would have been established as fact. Since the metachromatic reaction did not take place in destained blood smears of Case 1, this possibility is still open to question.

3. Monocytes

The "lacy" type of nuclear vacuolation described in the monocytes of Case 1 was not observed in the blood of the other patients in this series in the literature.

4. Platelets

The only morphologic change in the platelets was the increase in size observed in Case 1.

5. Summary

In summary, it may be stated that cytologic changes of the circulating leucocytes occurred only in the patients who had received the largest doses of radiation. The exception to this statement is the increase in neutral red staining lymphocytic bodies which was apparent in almost all patients. The changes in cell structure were of a degenerative nature and were consistent with those seen in fixed tissues.

D. Bone-Marrow Response as Revealed by Sternal Cavity Aspirations

It is impossible to get a satisfactory picture of the sequence of tissue destruction and repair from study of the limited number of bone-marrow aspirations done on this series of patients. Therefore, the pattern of bone-marrow response observed in the Japanese patients will be presented briefly (Medical Report of Joint Commission, Section 6) to be used as a basis for the interpretation of the observations on these patients.

The early bone-marrow response in the irradiated Japanese was not studied thoroughly, but enough observations were made to show that marrow destruction was prompt. Thus, in the single specimen obtained from a fatally injured patient on the second day after exposure, 5 percent of the leucocytes were lymphocytes, 29 percent histiocytes, 19 percent plasma cells and 44 percent were unidentifiable. In a few other marrow specimens obtained several days later, only a few plasma cells, degenerating metamyelocytes, and small lymphocytes were observed. All specimens obtained seven to ten days following exposure showed regeneration of the reticulum cells. Foci of mononuclear cells gradually developed in the hyperplastic reticulum. There was a gradual metamorphosis of the reticulum marrow into cells of the granulocytic, lymphoid, erythrocytic and megakarocyocytic series. Erythrocyte regeneration was delayed longer than was the recovery of the leucocytes. In most cases, the pattern of

myelopoiesis was normal seven or eight weeks after exposure, whereas the maximum intensity of enthropoiesis did not occur until the tenth to twelfth week. Bone-marrow samples of the survivors appeared completely normal between the twelfth and sixteenth week and thereafter.

In the present series of cases, the early destruction of the heavily irradiated sternal marrows of Cases 3, 4, and 5 is evident, with only a few indiscriminate cells surviving on the second day after exposure. In contrast, Cases 7 and 9 showed no evidence of marrow damage on the fourth day after the accident. Perhaps the most revealing studies are those done by Dr. Carl V. Moore on specimens obtained from Cases 4 and 6 twenty-two days after exposure. His observations indicate that there was marked hypoplasia of the marrow in both cases with only a few islands of normoblasts in Case 4, and a predominance of normoblasts in Case 6. The marrow of the latter patient also showed a striking increase in the number of phagocytic myelocytes and a moderate increase in tissue basophils. Hyperplasia of the reticulum was not observed. The marrow specimens of Cases 4 and 6, examined on the eleventh week or thereafter, showed a normal cellular pattern even though the erythrocyte count of the circulating blood was still depressed at this time. It is evident that these findings bear out the observations on the Japanese patients of rapid marrow destruction and regeneration within three months, but do not show the reticular hyperplasia noted in the Japanese.

Studies of the bone-marrow response of animals show that cell destruction occurs very shortly after exposure and that recovery is also relatively rapid (Bloom, 1948; Lawrence, Dowdy and Valentine, 1948). In rabbits given 800 roentgens of x-rays, for example, cell damage is evident thirty minutes after exposure and is very marked three hours later (Bloom, 1948). The erythroblasts disappear more rapidly than the myelocytes. The tissue destruction reaches its maximium five days after exposure at which time an "abortive" attempt at both myeloid and erythroid elements occurs. Sustained erythropoiesis begins on the tenth to fourteenth day, while myelopoiesis is somewhat delayed.

E. Disturbance of the Blood-Clotting Mechanism

Studies of blood coagulation in the patients involved in the second accident show that the blood-clotting time was definitely prolonged in Cases 3, 4, and 6, and slightly prolonged in Case 5. The prothrombin time remained normal in all patients except in Case 3 in whom there was terminal fall. The clotting time showed a progressive increase in Case 3 until at death the blood was uncoagulable. The delay in clotting time of Cases 5 and 6 was inversely proportional to the calculated dose. The maximum disturbance occurred on the sixth day after exposure in the survivors. The clotting time could be brought back to normal in all cases by adding small amounts of the dye, toluidine blue, to the sample of whole blood.

The increase in blood-clotting time of the blood of irradiated persons and animals is attributed, in part, to the presence of a material in the circulating blood which is believed to be heparin-like in nature, if not heparin itself (Allen, et al., 1948). The excess of this material can be neutralized by toluidine blue, or protamine, and the clotting time brought back to normal in vitro or in vivo, even in the presence of severe thrombocytopenia. This is evidence in favor of the heparin-like character of the anticoagulant since toluidine blue, a metachromatic dye, is known to combine specifically with the sulfuric acid ester type of linkage such as is possessed by heparin. The possible relation of this circulating anticoagulant to the tissue mast cells will be discussed in Section IX.

It should be emphasized that prolongation of the clotting time is only one cause of bleeding in the acute radiation syndrome, and that it is often not abnormal in irradiated animals (Cronkite, 1950). Decrease in platelets and increased capillary fragility, both of which have been observed in the injured Japanese and irradiated animals, obviously contribute to the development of the clinical bleeding that is usually

so conspicuous several weeks after exposure to lethal doses of radiation. These three phenomena are unrelated, as is indicated by the fact that the time of the appearance of the anticoagulant in irradiated dogs does not always coincide with the development of the thrombocytopenia (Allen, et al., 1948). In a report on the hemorrhagic phenomena in the animals exposed to the atomic explosion at Bikini, three types of bleeding were noted at different times after exposure. Capillary fragility with scattered petechiae occurred early, severe thrombocytopenia with purpura appeared later in the illness, and prolongation of the clotting time with severe bleeding was observed only during the terminal stages of the illness (Cronkite, 1947).

482 47

VII. CHEMISTRY OF THE BLOOD AND URINE

Prior to the last war the medical literature concerned with the changes in blood and urine chemistry induced by radiation exposure was limited to studies of a relatively few conventional metabolic systems. Much of the data is controversial as can be seen in the review of this subject by Dunlap (Warren, 1942). As will become evident, some of the inconsistencies may be explained by the fact that many of the chemical responses appear to be biphasic in nature; the reported data depend therefore, not only upon the dose but upon the time at which the studies were made. The more extensive chemical investigations of the blood and urines in irradiated humans and animals carried on during the last six years will soon be published in the Plutonium Project Report. A brief report of some of these studies appears in an article by Schwartz (1949).

A. Chemistry of the Blood

1. Quantitative Changes in Blood Chemistry

The methods used in studying blood chemistry are standard procedures which can be found for the most part in the book "Laboratory Methods of the U. S. Army" (Simmons and Gentzkow, 1946). Except for the definite changes noted in Case 3, the results of the rather limited studies do not signify a serious disturbance in the chemistry of the blood. Where deviations from the norm do occur, the changes are small and the significance dubious. When the data from all patients are collected in a single Chart (See Chart I), certain conclusions can be drawn concerning the blood chemistry in persons receiving large doses of radiation.

a. Sodium and Chloride Ions

A fall in serum sodium and whole blood chlorides occurred in Case 3. The slight but apparently significant drop in the sodium level was evident during the first few days of the illness before large quantities of fluid and electrolytes were lost as a result of gastric suction. The blood chlorides decreased strikingly during the last few days of the illness despite efforts to maintain the normal electrolyte levels by parenteral fluid administration. In the other cases, the studies of serum sodium and blood chloride were too limited to permit a good evaluation of the true state of the electrolyte balance. Nevertheless, there appeared to be no significant changes in these patients as is shown by the normal chloride level of Cases 4, 5, and 6, and the low normal values for serum sodium in the same patients during the first few days.

The observations of decreased serum sodium in Case 3 are unlike those obtained in recent studies of the sodium metabolism in irradiated rats where sodium retention in the blood and tissues was observed (Bennett, Bennett and Howland, 1949-a). Studies on patients who received radiation therapy, however, show decreased blood chlorides even though chloride retention in tissues occurs (Warren, Dunlap, 1942). Similarly, a progressive decrease in blood chloride and sodium often may be demonstrated during the first few days after injury in thermally burned patients (Harkins, 1942). Disturbances in body sodium of the most severely injured patients are not unexpected in view of the extensive adrenal damage which is known to occur. The rather slow fall in serum sodium in Case 3, however, is unlike the rapid drop in chlorides, and presumably sodium, noted in the general adaptation syndrome or alarm reaction which occurs as a result of many non-specific stimuli of which roentgen rays are allegedly one. (Selye, 1940, 1946).

b. Calcium

The calcium content of the blood was studied only during the terminal phase of the illness of Case 3. A decreased calcium value is usual in a patient with uremia

COMPOSITE CHART OF BLOOD CHEMISTRY

		Serum	Whole Blood					Serum				
Case	Days After Exposure	Sodium m Eq/l	Chlorides m Eq/l	Urea N mg %	Non-protein Nitrogen mg %	Uric Acid mg %	Calcium mg %	Icteric Index	Total Protein g %	Albumin g %	Globulin g %	A/G
1	14	-	-	-	38	2.7	-	-	-	-	-	-
3	1 (1 hr.)	140	-	-	-	-	-	-	-	-	-	-
	2	134	75.3	10	-	3.4	-	-	-	-	-	-
	3	132	-	-	-	-	-	-	-	-	-	-
	4	133	67.8	-	57.0	-	-	-	5.4	4.1	1.3	3.1
	8	-	68.3	24	-	5.6	8.8	-	7.9	5.8	2.1	2.8
	9	-	63.0	-	161.0	-	9.4	70	7.0	4.8	2.2	2.2
4	1 (1 hr.)	139	-	-	-	-	-	-	-	-	-	-
	2	142	-	-	-	-	-	-	-	-	-	-
	3	136	78.3	-	-	4.0	-	-	-	-	-	-
	4	-	-	15	-	-	-	-	6.6	5.2	1.4	3.7
	8	-	80.9	7	-	3.9	-	-	9.6	5.3	4.3	1.2
	17	-	-	-	-	-	-	-	7.0	4.2	2.8	1.5
	21	-	-	-	-	-	-	-	7.0	4.7	2.3	2.0
	28	-	-	-	-	-	-	-	5.7	4.1	1.6	2.5
5	1 (1 hr.)	146	-	-	-	-	-	-	-	-	-	-
	2	137	-	-	-	-	-	-	-	-	-	-
	3	-	78.8	12	-	4.1	-	-	-	-	-	-
	7	-	-	-	-	3.4	-	-	6.40	5.20	1.20	4.30
	8	-	78.7	14	-	-	-	-	9.10	5.30	3.80	1.40
	10	-	-	-	28.2	-	-	-	-	-	-	-
	15	-	-	-	-	-	-	-	5.30	3.20	2.10	1.40
	20	-	-	-	-	-	-	-	6.90	4.00	2.90	1.40
	22	-	-	-	-	-	-	-	7.50	5.34	2.16	2.47
	28	-	-	-	26.0	-	-	-	7.43	5.38	2.05	2.62
	30	-	-	-	28.0	-	-	-	7.34	-	-	-
	38	-	-	-	36.0	-	-	-	7.08	4.81	2.27	2.12
	42	-	-	-	34.0	-	-	-	7.70	5.52	2.18	2.53
	9 months	-	-	18	34.0	-	-	-	6.70	4.90	1.60	1.37

COMPOSITE CHART OF BLOOD CHEMISTRY (cont'd)

		Serum	Whole Blood					Serum				
Case	Days After Exposure	Sodium m Eq./l	Chlorides m Eq./l	Urea N mg %	Non-protein Nitrogen mg %	Uric Acid mg %	Calcium mg %	Icteric Index	Total Protein g %	Albumin g %	Globulin g %	A/G
6	1 (1 hr.)	141	-	-	-	-	-	-	-	-	-	-
	2	138	-	-	-	-	-	-	-	-	-	-
	3	-	71.9	10	-	3.8	-	-	-	-	-	-
	8	-	80.9	14	-	4.4	-	-	8.0	4.5	3.5	1.3
	14	-	-	-	-	-	-	-	6.5	3.9	2.6	1.5
	19	-	-	-	-	-	-	-	6.9	4.5	2.4	1.8
	23	-	-	-	-	-	-	-	7.8	4.8	3.0	1.6
	27	-	-	-	-	-	-	-	6.8	4.8	2.0	2.4
7	1 (1 hr.)	140	-	-	-	-	-	-	-	-	-	-
	2	-	70.1	9	-	3.6	-	-	-	-	-	-
8	1 (2 hr.)	140	-	-	-	-	-	-	-	-	-	-
	3	-	84.8	8	-	3.2	-	-	-	-	-	-
9	1 (2 hr.)	141	-	-	-	-	-	-	-	-	-	-
	3	-	73.3	14	-	4.0	-	-	-	-	-	-
10	1 (1-1/2 hr.)	138	-	-	-	-	-	-	-	-	-	-
	3	-	67.8	8	-	3.1	-	-	-	-	-	-
Normal Values*		136-145**	77.0-85.6	10-15	25-35	2-4	10-12	4-6	6.5-8.0	4.0-6.0	1.5-2.5	2.1-3.1

* Table 24, p. 242, Simmons and Gentzkow, 1946

** Serum sodium determinations done at Los Alamos Laboratory. For the normal range, see Mallory, Castleman and Parris, 1946

of this degree. It is undoubtedly the result of phosphorus retention which, although not measured, must have been a consequence of the impaired kidney function in this patient.

c. Non-Protein Nitrogen and Urea

Severe nitrogen retention was observed in Case 3. This was compatible with the observed extensive kidney damage.

In Case 1, however, the only blood urea determination gave a value at the upper limit of normal on the fourteenth day. One can only speculate as to whether azotemia occurred in the terminal week of life of this patient. In view of the good urine output and the absence of significant kidney lesions at autopsy, it seems unlikely that true or even extrarenal nitrogen retention of a serious degree occurred in Case 1. The blood urea and non-protein nitrogen determinations on the survivors of the second accident were done often enough to indicate that azotemia did not occur.

Increased excretion of non-protein nitrogen in the urine after abdominal irradiation of dogs was first demonstrated many years ago (Hall and Whipple, 1919). The animals which received lethal doses of x-rays usually showed no elevation of the blood non-protein nitrogen until shortly before death. Recent studies on dogs subjected to lethal doses of total-body irradiation show that there is an actual decline in blood non-protein nitrogen until 48 hours before death when a pronounced rise occurs (Prosser, Painter and Swift, 1947-b). In contrast to the delayed nitrogen retention observed in the terminal stages of injury by ionizing radiation, patients with severe thermal burns may show an elevated blood urea within a matter of hours after injury (Medical Research Council, 1944). As pointed out previously, the mild negative nitrogen balance shown by thermally burned patients (Section V) is not believed to be due to enhanced excretion of breakdown products of protein but rather to reduced caloric intake. In irradiated dogs, on the other hand, the negative nitrogen balance is presumed to be due to increased tissue breakdown. In Case 3, the disproportion between the non-protein nitrogen and urea on successive days is interesting since observations on irradiated dogs indicate that in the terminal azotemia, the ratio of these nitrogen fractions is unchanged (Prosser, Painter and Swift, 1947-b). In patients with thermal burns, however, a similar increase in the non-urea nitrogen fraction is said to be not uncommon (Taylor, et al., 1943-a). One of the non-urea fractions responsible for the azotemia is called residual nitrogen. This substance has not characterized completely, but an idea of its composition can be gained from the fact that it breaks down on hydrolysis to form amino-and amido-nitrogen.

d. Uric Acid

A terminal increase in the blood level of uric acid was noted only in Case 3. No attempt was made to follow the amounts of uric acid excreted in the urine; this might have given a much better indication of a disturbance in purine metabolism. An increase in amount of urinary uric acids has often, but not invariably, been reported in patients following radiation therapy (Pohle and Severinghaus, 1930). In dogs to which a lethal dose of x-rays has been given a terminal rise in urinary uric acid has been observed; in these same animals no significant change was noted in the excretion of allantoin (Schwartz, 1949).

e. Total and Fractionated Serum Proteins

The quantitative data for total and fractionated serum proteins are difficult to interpret. There is no obvious trend or gross change in pattern observed in the patients injured in the second accident. All of the patients of the second accident showed an increased value for serum proteins on the eighth day after exposure due to an elevation of the globulin fraction. This observation was not confirmed immediately, so it is not possible to be certain that this represents an actual increase in

serum proteins. Data gained from the study of irradiated dogs indicate that a slight decline occurs in plasma proteins during the first week after exposure (Prosser, Painter and Swift, 1947-b). In dogs receiving more than 300 roentgens of x-rays a rise up to 15 percent of the total circulating proteins occurred during the second and third weeks. Those animals which died showed a terminal decline in protein values following the initial rise. The infrequency of the protein determinations in these patients make it impossible to ascertain whether or not such a biphasic response occurred.

In patients with thermal burns, an increase in serum proteins may occur within the first few hours after injury (Cope, et al., 1948). Later, the protein level may fall below normal values, particularly in cases receiving inadequate therapy in the form of transfusions of whole blood or plasma.

2. Electrophoretic Pattern of the Plasma Proteins

The electrophoretic pattern of five heparinized blood samples from these patients was determined in a Tiselius apparatus (Tiselius, 1937), using the Schlieren scanning technique (Longsworth, 1939). The resultant ascending patterns of the patient's plasma proteins as well as those of six control samples were analyzed by measuring the area under each peak in the curve with a planimeter. The proportion of protein in the albumin fraction and in each of three globulin fractions are shown in the accompanying table. Significant changes in the plasma proteins are found only in Case 3. Here the plasma albumin level diminished while an increase in alpha globulin fraction occurred. This change in the electrophoretic pattern is shown in Fig. 61. Minor changes of questionable significance are also found in the electrophoretic patterns of Cases 5 and 6.

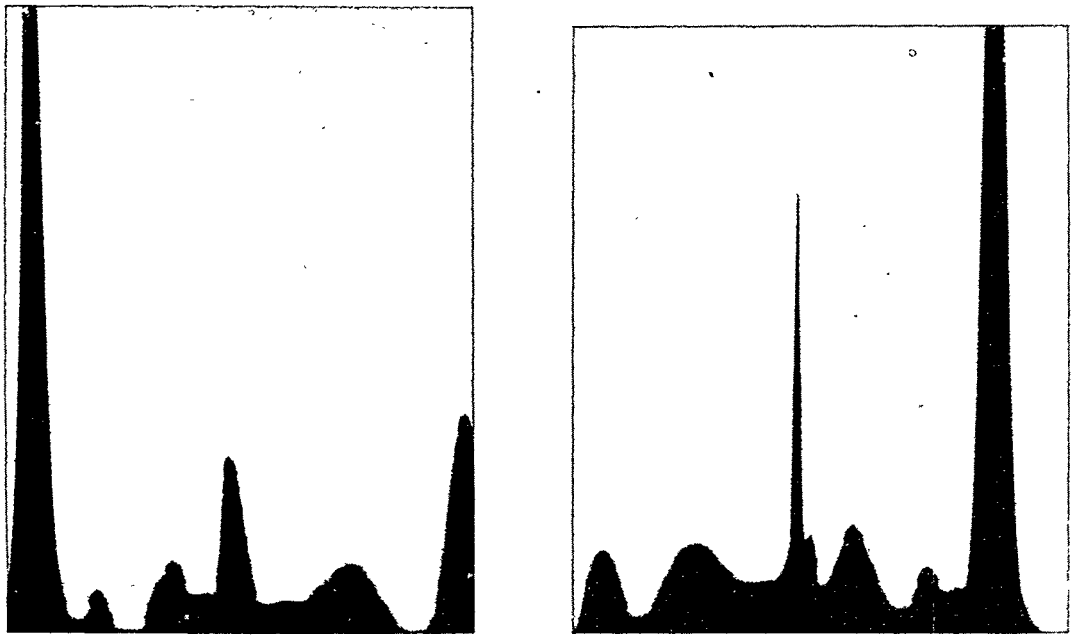
Changes in the electrophoretic pattern of plasma proteins of dogs given large doses of x-rays have been described (Muntz, Barron, and Prosser, 1949). These changes consist of an increase in the alpha globulin fractions of the plasma and a diminution of the albumin fraction. The increase in globulin coincides in time with the development of fever in severely injured animals. A similar change in the electrophoretic pattern has been observed in dogs poisoned with sulfur mustard (β-β-dichlordiethyl sulfide) and injured in other ways including thermal burning (Chanutin and Gjessing, 1946). A marked increase in the alpha globulin fraction has also been observed in humans with pneumonia and other febrile diseases (Blix, 1939).

It may be concluded from these studies that the changes in plasma protein observed in Case 3 are not specific for radiation damage. It has been suggested that the close resemblance between the protein changes in severe bodily injury and in acute febrile infections may indicate that the reaction of the plasma proteins to injury is related to adrenal stimulation and the protein catabolic response (Luetscher, 1947).

It should be pointed out that the electrophoretic method of analyzing proteins is more accurate and reproducible than the "salting out" method usually used in fractionating proteins (Leutscher, 1947). The results determined by this method are undoubtedly more reliable than the protein fractions and albumin/globulin ratios reported in the table.

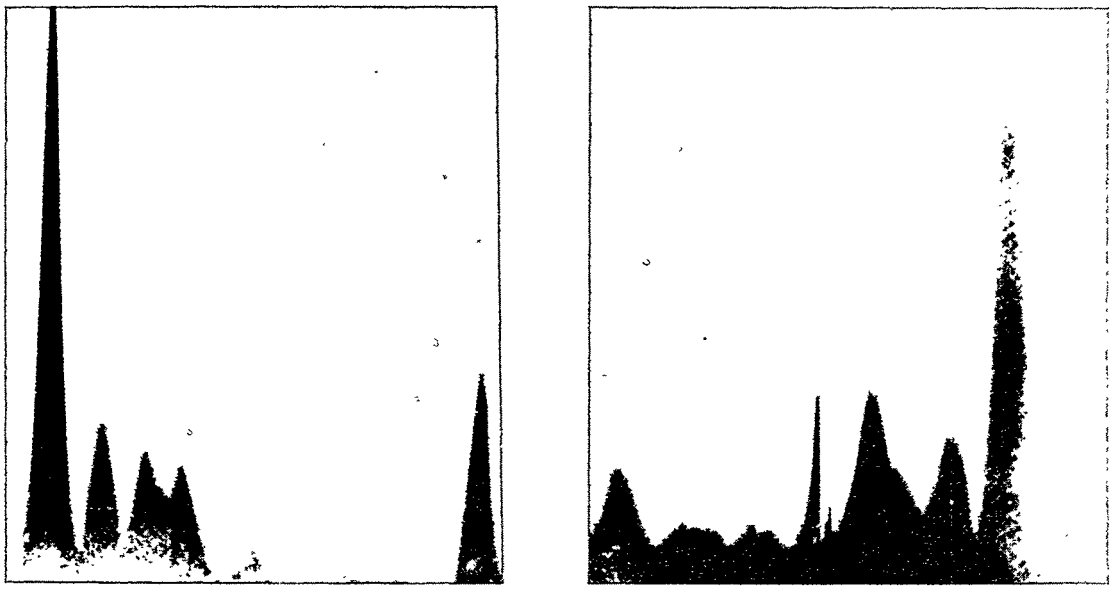
3. Summary

Omitting the first two cases from the discussion because of inadequate study, the changes in blood chemistry may be summarized by saying that significant abnormalities were demonstrated only in Case 3, and were not of a sufficient degree in themselves, to be the immediate cause of death. These changes consisted of a gradual fall in serum sodium and whole-blood chlorides, a slight elevation of uric acid, a terminal rise in nitrogen associated with a depressed calcium level, and a modification of the plasma protein (decreased albumin and increased alpha globulin fraction). Such changes are consistent with those noted in animals given lethal doses of ionizing



Ascending Descending

Fig. 60 Normal human plasma protein pattern



Ascending Descending

Fig. 61 Plasma protein pattern of Case 3 seven days after exposure

482 53

radiation. The limitations of this study are such that minor variations in the blood chemistry in the survivors would probably not have been detected.

B. Chemistry of the Urine

Except for small aliquots used for clinical microscopy and radioactivity studies, all of the urine collected from the patients involved in the second accident was sent to the Argonne National Laboratory in Chicago for analysis. For the most part, twenty-four-hour pooled samples under toluene were sent via air express after careful packaging and refrigeration with dry ice. In all instances, however, the dry ice had evaporated by the time the samples reached Chicago so that the urines were unchilled at least part of the time in transit. Upon being received at the Argonne Laboratory, quantitative analyses of the corticoid-like substance, urobilinogen and porphyrins were undertaken promptly. The remainder of the urine samples were stored in an icebox at 40° F. Analysis of urinary amino acids was started one year later and determinations of potassium content of the samples were done three years later.

1. Corticoid-like Fraction*

Studies of the urinary corticoids are of interest because abnormalities in quantity may indicate dysfunction of the adrenal cortex, such as is known to occur after trauma or other forms of stress (Selye, 1940, 1946); thus, corticoids have been shown to be present in abnormally large quantities in patients who have been operated on or who have suffered severe thermal burns (Shipley, et al., 1946). In one thermally burned patient of their series the urinary corticoid content was elevated for more than 12 days. Chemical studies of corticoid-like urinary extracts also have shown increased excretion levels in patients with thermal burns (Talbot, et al., 1945). Investigation of the excretion of neutral 17-ketosteroids, which although a poor measure of adrenal activity are nevertheless influenced by adrenal cortical function, also bring to light some interesting abnormalities in patients with thermal burns (Aub, et al., 1943). Thus, an increased excretion of 17-ketosteroids occurs from three to seven days after injury; following this the urinary content of this type of steroid falls precipitously to low normal or subnormal levels. The duration of the depressed 17-ketosteroid level is said to be correlated closely with the severity of the injury and the period of convalescence (Aub, et al., 1943). A recent report indicates that irradiated dogs also show an initial increase in urinary 17-ketosteroids followed by a fall to subnormal values 5 to 12 days after exposure.

The investigation of corticoid-like materials in the urine of these patients was patterned after the colorimetric method of assaying urinary extracts (Talbot, et al., 1945). This method employs the measurement of the reducing properties of a ketonic water soluble lipid component of urine which is believed to be composed largely of

*Since the terminology on the field of adrenal cortical hormones has not been agreed upon, it seems advisable to define the terms as they are used in this article. The following definitions have been suggested by Dr. L. L. Engel (personal communication). The term cortin refers to adrenal cortical extracts which have the total biological activities of the adrenal cortical hormone. Corticoid, on the other hand, is applied to urinary extracts which have some of the biological properties of cortin. The tautological term corticoid-like is used to describe those urinary substances which are presumed to be corticoids because of similar chemical and physical properties, but which have not been identified by biologic assay methods. Corticosteroid refers to the cortical hormones, the chemical structure of which is known, e.g., corticosterone, compound E, etc. (See Fieser and Fieser, 1949, for a discussion of the chemistry and physiological properties of the adrenal cortical hormones).

COMPOSITE CHART OF URINE CHEMISTRY

Case	Days After Exposure	Volume (cc)	Specific Gravity	Corticoid Fraction			Coproporphyrin		Urobilinogen*	
				(a) 220/240 mμ	(b) 240/260 mμ	a/b	μg %	μg/day	mg %	mg/day
1	24			1.45	3.6	0.40	21	252		
3	1 (12 hrs)	310	1.022	1.28	1.36	0.94	1.8	-	trace	-
	2	500	1.021	1.47	1.17	1.25	2.4	-	0.02	0.1
	5	990	1.027	0.93	1.62	0.57	18.8	186	0.06	0.6
	6	1770	1.017	1.09	1.40	0.78	25.5	451	0.16	2.8
	7	466	1.016	1.70	1.18	1.44	25.1	117	1.33	6.2
4	1 (2 hrs)	175	1.020				11.1)	)	)	
	1 (4 hrs)	160	1.012				2.0)	52)	trace)	trace
	1 (15 hrs)	660	1.020				4.5)	)	)	
	2	1825	1.022	1.40	1.32	1.06	19.8	362	0.01	0.2
	5	1520	1.020	1.40	1.75	0.80	11.2	170	0.08	1.0
	6	1390	1.020	1.07	1.95	0.55	10.1	140	0.48	6.7
	7	1720	1.017				3.8	65	0.01	0.2
	8	1710	1.016	1.43	1.58	0.90	2.5	43	0.01	0.1
	9	3045	1.015	1.91	1.52	1.25	5.0	152	0.04	1.1
	10	1795	-	-	-	-	-	-	-	-
	11	3050	1.016	-	-	-	4.1	125	0.09	2.6
	18	2200	1.013	1.62	1.06	1.53	1.6	35	0.01	0.2
	21	3650	1.018	1.49	1.15	1.30	5.7	206	trace	trace
	24	2780	1.020	-	-	-	2.4	67	trace	trace
	26	2565	1.020	-	-	-	6.7	172	trace	trace

* Urobilinogen values probably too low because of lack of complete refrigeration during trip to Chicago

COMPOSITE CHART OF URINE CHEMISTRY (cont'd)

Case	Days After Exposure	Volume (cc)	Specific Gravity	Corticoid Fraction			Coproporphyrin		Urobilinogen*	
				(a) 220/240 mμ	(b) 240/260 mμ	a/b	μg %	μg/day	mg %	mg/day
5	1 (3 hrs)	100	-	1.40	1.53	0.91	-	-	-	-
	1	295 (?)	1.030	0.78	2.96	0.27	4.5	13 (?)	trace	
	2	2300	1.015				5.4	124	trace	
	5	2660	1.013				2.7	72	trace	
	6	2370	1.015	1.31	1.72	0.76	2.7	64	0.3	0.8
	7	2725	1.013	1.31	1.72	0.76	1.4	38	trace	
	8	1730	1.016	1.29	1.73	0.75	1.7	29	trace	
	9	1910	1.013	-	-	-	2.0	38		
	10	4535	1.013	-	-	-	1.9	86	0.04	1.8
	12	2730	1.015	-	-	-	1.4	38	trace	0.3
	30	-	1.022	1.40	1.50	0.93	5.6	-	trace	
6	1 (4 hrs)	134	1.020	-	-	-	2.5	-	trace	-
	1	630	1.020	-	-	-	4.3	-	trace	-
	2	435	1.030	-	-	-	5.7	25	trace	-
	5	2660	1.015	-	-	-	1.6	42	trace	-
	6	3050	1.015	0.92	2.63	0.35	4.2	128	trace	0.2
	7	2815	1.010	-	-	-	1.8	51	trace	-
	8	1760	1.012	-	-	-	5.5	97	0.04	0.7
	9	2355	1.013	-	-	-	4.1	97	0.01	0.3
	10	3300	1.010	-	-	-	1.9	63	0.02	0.7
	11	3655	1.007	-	-	-	1.7	62	0.03	1.2
	12	2585	1.015	-	-	-	2.6	67	0.01	0.36
	19	2920	1.010	1.43	1.59	0.90	2.9	85	trace	0.2
	21	1880	1.020	1.32	1.87	0.71	1.3	24	trace	-
	26	2730		-	-	-			0.03	0.9
	29	1820		-	-	-			0.02	0.3

* Urobilinogen values probably too low because of lack of complete refrigeration during trip to Chicago

COMPOSITE CHART OF URINE CHEMISTRY (cont'd)

Case	Days After Exposure	Volume (cc)	Specific Gravity	Corticoid Fraction			Coproporphyrin		Urobilinogen*	
				(a) 220/240 mμ	(b) 240/260 mμ	a/b	μg %	μg/day	mg %	mg/day
7	1 (12 hrs) 1	1270 310	1.013 1.015	1.88	0.85	2.21	4.8) 3.1)	72	0.03) 0.01)	0.4
8	1 (12 hrs) 1	115 1250	- 1.029	1.72 -	0.98 -	1.76 -	- 10.1	- 126 +	- 0.02	- 0.3 +
9	1 (4 hrs) 1	171 870	1.016 1.017	1.97	1.07	1.84	7.5 2.3	- -	trace trace	- -
10	1 (4 hrs) 1	128 1285	1.015 1.011	- 1.88	- 1.08	- 1.74	3.2 1.9	- 24	trace trace	- -
?	** ***			1.15 1.38	1.77 1.33	0.65 1.04				
Controls Usual Range			1.015 1.030	1.5-1.7	0.9-1.1	1.4-1.8	4-9	30-100	trace to 0.2	trace to 2

* Urobilinogen values probably too low because of lack of complete refrigeration during trip to Chicago

** Three days before death

*** Day of death

the corticosteroids. Circumstances did not permit complete extraction and purification of the corticoid fraction of the urine samples in the present series of cases. Also, the procedure was modified somewhat from that originally reported, in that spectrophotometric analysis rather that colorimetric assay was used for quantitative determination of excretion products. The method of separation of the corticoid-like fraction consisted, in brief, of extracting urine aliquots several times with chloroform and benzene. The crude extract was not separated further into ketonic and non-ketonic fractions. Its absorption pattern in methanol was determined by use of a Beckman spectrophotometer. The ultraviolet absorption pattern from 220 to 340 mμ (millimicrons) of the crude extract of several selected urine samples is shown in Graph I. It is evident from this graph that there is an increase in specific absorption in the region of 230 mμ in the urine extract of Cases 1 and 3 and of the thermally burned patient. In other samples from severely burned patients not shown on this graph there was a plateau or actual peak at about 230 mμ, although not as extreme as that in Case 1. The adrenal cortical hormones, e.g., corticosterone, show specific absorption in this region of the ultraviolet spectrum due to the presence of alpha, beta unsaturated ketones in their chemical structure (Fieser and Fieser, 1949). Because of the method of extraction it seems possible that the absorbing materials measured in this study are composed, at least in part, of the corticoid-like water soluble reducing substances measured by the colorimetric method of Talbot.*

In an effort to show disturbance in the absorption pattern in the 230 mμ region, figures are given on the composite chart and in individual charts to indicate the slope of the curve from 220 to 240 mμ (a) and from 240 to 260 (b). The greater the absorption at 230, the steeper is the slope from 240 to 260 mμ, or the larger is the ratio of 240/260 (b). Similarly, abnormalities in the curve will be reflected in the ratio a/b. No effort is made to express the absolute amounts of absorbing material.

Examination of the composite chart indicates an increase in absorption at 230 mμ for Cases 1, 3, 4, 5, and 6. This increase was most marked in the single urine specimen of Case 1 obtained shortly before death. In Case 3 it was less marked and inconstant, falling almost to normal on the second and again on the eighth post-exposure day. In this case, however, as well as in Cases 4 and 6, the greatest increase occurred between the fifth and seventh days after exposure. In Case 5, on the other hand, maximum absorption occurred on the second day. Disturbance of absorption in the 230 mμ region was still present in Case 5 four weeks after exposure, and in Case 6, three weeks after the accident. In contrast to the observations on these patients, the absorption pattern in the urine of the less heavily exposed patients (Cases 7 to 10) was not abnormal. Studies in the latter cases, however, were limited to one determination in the urine sample passed immediately after the accident. It is possible that observations on later specimens would have shown abnormal absorption spectra.

It must be emphasized again that the ultraviolet absorbing substance measured in these studies may not be a single compound and that there is only circumstantial evidence indicating that it is similar to the reducing substance measured by colorimetric assay. It seems quite possible, however, that the two substances are similar in nature or at least are composed of some of the same steroids. The reducing substance measured colorimetrically by Talbot has been found to be elevated in patients with adrenal hyperfunction and is believed, therefore, to be composed of corticoids. It might be expected that the excretion of increased amounts of the material with ultraviolet absorption at 230 mμ in these cases also was the result of hyperactivity of the adrenal cortex. This would fit in with the Selye concept of the

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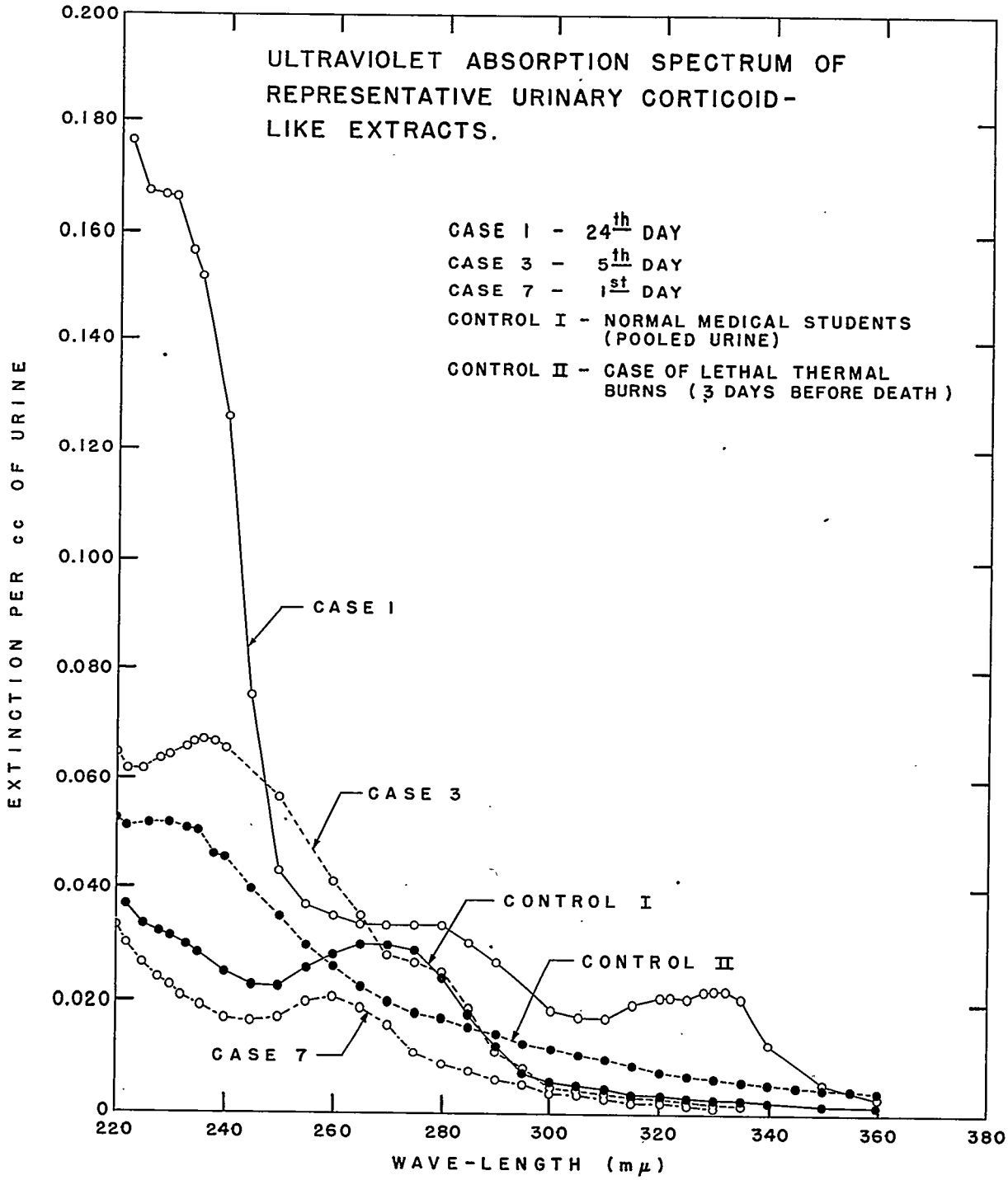
*In a single patient receiving ACTH, strong absorption in the 230 mμ region has been demonstrated in the chloroform soluble fraction of the urine (L. L. Engel, personal communication).


ULTRAVIOLET ABSORPTION SPECTRUM OF REPRESENTATIVE URINARY CORTICOID-LIKE EXTRACTS.
CASE 1 - 24th DAY
CASE 3 - 5th DAY
CASE 7 - 1st DAY
CONTROL I - NORMAL MEDICAL STUDENTS (POOLED URINE)
CONTROL II - CASE OF LETHAL THERMAL BURNS (3 DAYS BEFORE DEATH)
EXTINCTION PER cc OF URINE
0.200
0.180
0.160
0.140
0.120
0.100
0.080
0.060
0.040
0.020
0
CASE 1
CASE 3
CONTROL I
CONTROL II
CASE 7
220
240
260
280
300
320
340
360
380
WAVE-LENGTH (mμ)

adaptive reaction of the body to injury.*

The terminal decrease in Case 3 in the excretion of this substance, if it is a corticoid, would be expected in view of the clinical circulatory collapse of this patient. There was no evidence in these studies to indicate late adrenal hypofunction which some of the clinical symptoms of Cases 4 and 5 (asthenia and salt hunger) might suggest.

2. Urobilinogen

Although admittedly not a specific test for liver damage, the quantitative studies on urobilinogen excretion were used in this study as an index of liver function (Watson, 1942). Increased urobilinogen excretion is also found in conditions other than liver injury; for example, abnormal amounts of fecal urobilinogen (as well as a less marked elevation of urine urobilinogen) are found in patients with enhanced hemolysis (Steigman and Dyniewicz, 1943).

The method employed for determination of urobilinogen in urine was that devised by Schwartz, Sborov, and Watson (1944). Before considering the results of these determinations, it should be pointed out that all values in the present study may be low because of improper preservation of the specimens. Taking the figures at their face value, however, it is evident that abnormally high urobilinogen excretion was

*The clinical picture of the General Adaptation of Alarm Reaction caused by injurious agents of various types has been clearly described, and its association with hyperplasia and hyperfunction of the adrenal cortex has been stressed (Selye, 1940, 1946). More recently, the clinical findings have been correlated with the release of adrenocortical hormones (Albright, 1943; Albright and Reifenstein, 1948). As this hypothesis helps to explain the urinary corticoid-like findings in this part of the chemistry discussion, it is reported briefly here: The adrenal cortex normally secretes, among others, two types of steroid hormones: (1) the Nitrogen of "N" hormones, and (2) the Sugar or "S" hormones. The N hormones which are testosterone-like in their properties, and the metabolic products of which are the urinary 17-ketosteroids, stimulate the synthesis of protoplasm (anabolism). The S hormones, which have a profound influence on fat and protein, as well as on carbohydrate metabolism, supposedly inhibit anabolism. The action of these two hormones, then, is, in part, antagonistic. The S hormones are composed of 11-oxy-corticosteroids, the metabolic products of which are presumed to have been measured in the urine of the patients of the present series as well as in those mentioned in the literature (Shipley, et al., 1946; Talbot, et al., 1945).

The Albright hypothesis of the mechanism of the adrenocortical response to injury suggests that following body trauma of any kind, release of both the N and S hormones takes place. The increase in production of the N hormones is of a temporary nature and is soon followed by a decrease which lasts until convalescence is nearly completed. The resultant excretion of 17-ketosteroids, then, is first increased and later diminished to subnormal levels. The enhanced production of S hormones persists until convalescence, thereby accounting for the increased urinary excretion of 11-oxy-corticosteroids which Albright believes the corticoids to be. This imbalance of adrenal cortical function with excessive secretion of S hormone may account for the following symptoms noted after injury: leucocytosis, atrophy of lymphoid tissue, insulin resistance, muscular weakness, retarded growth and decreased synthesis of protoplasm in general. The adrenal response, as well as it can be reconstructed from the data in these cases and in irradiated animals, does conform to the general over-all pattern described by Selye and Albright. The reader is cautioned, however, against placing too much emphasis on any single type of response of this nature; rather, one must keep in mind the numerous direct and indirect factors mentioned in Section IV in order to understand the complete clinical picture of the Acute Radiation Syndrome.

detected only in Cases 3 and 4. The elevation in Case 3 presumably occurred as a result of terminal liver failure. The urobilinogen level in Case 4 was high only on the sixth post-exposure day. Although no quantitative determinations were done in Case 1, dilution tests and the incidental observation of strong absorption in the ultraviolet spectrum at 490 mμ noted in the study of the urorosein content of a urine sample two days before death suggest that the urobilinogen excretion was high.

It should be emphasized again that, in evaluating the urobilinogen determinations, one must keep in mind the probable destruction of this compound in urine specimens due to lack of refrigeration. Therefore, minimal changes in liver function probably would not have been detected in these studies. Nevertheless, terminal liver failure and the associated increase in urinary urobilinogen excretion observed in these fatally injured patients is consistent with the data obtained from dogs given lethal doses of x-rays (Schwartz, 1949), and with the observations in Japanese patients of increased urinary urobilinogen (as shown by qualitative tests) and other evidence of liver dysfunction (Medical Report of Joint Commission, Section 2). The temporary liver dysfunction in Case 4 is also compatible with the observations in dogs treated with moderately large doses of x-rays (Schwartz, 1949). In thermally burned patients, increased urobilinogen excretion may also occur during the first few days after injury (Medical Research Council, 1944).

3. Porphyrins

Disturbance in porphyrin metabolism is found in a number of apparently unrelated diseases; thus, increased amounts of urinary porphyrin may be found in patients suffering from liver disease, certain types of anemia, metal poisoning, febrile diseases of various types, and congenital porphyrinuria (Watson and Larson, 1947). Of the two naturally occurring isomers of coproporphyrin, Type I predominates in normal urines, while either isomer (Type I or Type III) may appear in abnormally large quantities in the above mentioned diseases. Usually, the Type II isomer predominates in the urine of patients with metal poisoning and with chronic liver disease.

A quantitative fluorimetic method of determination was used to study the urinary porphyrins (Schwartz, et al., 1947-a). It is apparent from the examination of the composite chart that the upper range of normal coproporphyrin (4 to 9 micrograms percent or 30 to 100 micrograms per day) was exceeded in the single specimen from Case 1. Increased porphyrin excretion was also found in the urines passed by Case 3 after the fifth post-exposure day, and in the urine samples passed on the second and fifth days by Case 4. In practically all urine samples from the other patients involved in the second accident, the coproporphyrin levels were within the limits of normal. The only instances of depressed urinary coproporphyrin are found in the first two specimens of Case 3 passed within twelve hours after exposure. Only two urine samples were fractionated into porphyrin isomers. In each case, Type III coproporphyrin predominated. Thus, in the first day urine sample of Case 3, 77 percent of the total porphyrin was Type III, while in pooled samples from the urine passed by Case 4 during the first twelve days, 95 percent of the pigment was Type III.

The terminal elevation in the urinary coproporphyrin level of the fatally injured patients is consistent with the observations in dogs given lethal doses of x-rays (Schwartz, 1949). It has been postulated that liver failure is responsible for the late increase in coproporphyrin excretion. In heavily irradiated dogs, the terminal increase is preceded by a depression of urinary porphyrin. This response, which could be due to the inhibition of erythropoiesis noted histologically, also occurred in Case 3.

4. Urorosein and Other Pigments

Urorosein is a red pigment formed in strong acid solution by the oxidation of indolacetic acid, a tryptophane derivative excreted in the urine. Studies of the urorosein content of these urine specimens were done. The exact details of the procedure

482 61

COMPOSITE CHART OF AMINO ACID EXCRETION

amounts expressed in mg/ml (approximate concentration)

Case	Day of Illness	Cysteic Acid	Aspartic Acid	Glutamic Acid	Glycine	Taurine	Alanine	Valine	Leucine	Unknown Below Alanine	Unknown Below Valine	Other
1	23	0.5	0.5	1.0	1.5	3.0	trace	0	0	0	0	hydroxy-proline 1.5
3	1	0.1	0.1	0.1	1.0	0.75	0.5	0.25	0	0.25	0.1	phenyl-alanine 0.1
	2*	0	0.1	0.1	0.75	1.0	0.5	0.5	0	0.25	0	phenyl-alanine 0.1
	6	0	0.5	1.0	1.0	1.75	0.5	0.25	0.25	0.25	0	serine-tr. unknown - 0.25 arginine - 0.1
	7	0	0.5	1.0	1.0	1.0	0.25	0.5	0	0	0	---
4	6 hr. 1	0.25	0.25	0.5	1.0	0.75	0.25	trace	0	0	0	arginine - 0.1 unknown - 0.1
	1	trace	0.25	0	2.5	0.75	trace	0.1	0.1	trace	0	unknown - 0.25
	2	0	0.5	0.5	2.0	0.75	0.25	0.1	0.1	0.25	0	---
	4	0.1	0.25	0.5	trace	1.0	0	0.1	0	0	0	---
	6	0.25	0.5	0.75	1.5	1.0	0.25	0.1	0.1	0.1	0	arginine-tr.
	9	trace	0.25	0.25	0.5	0.5	0.25	0.1	0	0.1	0	---
	10	0.25	trace	0.1	0	0.75	0	trace	trace	0	0	---
	11	0.1	0.25	0.25	0.5	0.75	trace	0.1	0	trace	trace	---
	12	trace	0	0.25	1.0	0	trace	0	0	0	0	---
	20**	0.1	0.1	0.1	0	1.0	0	trace	0	0	0	---
	22	0	0	trace	0.25	0	0	0	0	0	0	---
	23	trace	0	trace	trace	0	0	0	0	0	0	---
	27	trace	0	trace	0	0	0	0	0	0	0	---
	30	0	0	trace	0	0.1	0	0.1	0	0	0	---

* 12-hour sample
** Partial 24-hour sample

482 62

COMPOSITE CHART OF AMINO ACID EXCRETION (cont'd)

amounts expressed in mg/ml (approximate concentration)

Case	Day of Illness	Cysteic Acid	Aspartic Acid	Glutamic Acid	Glycine	Taurine	Alanine	Valine	Leucine	Unknown Below Alanine	Unknown Below Valine	Other
5	1	0	0.25	0	0	0	0.1	0	0	0	0	---
	1 1/2	trace	0.25	0	1.0	0.5	0.1	trace	0	trace	0	arginine - 0.1
	2	0	0	0	0	0	0	0	0	0	0	---
	6	0	0	0	0	0.25	0	trace	0	0	0	---
	8	0	0	0	0	0	0	0	0	0	0	---
	10	0	0	0	0	trace	0	0	0	0	0	---
	12	0	0	0	0	0	0	0	0	0	0	---
	13 1/2**	0	trace	0.1	0.1	trace	trace	0	0	0	0	---
	14	0	0	0	0	trace	0	0	trace	0	0	---
6	6 hr.	0	0	0	0	0	0	0	0	0	0	---
	1	0	trace	0	0	trace	0	trace	0	0	0	arginine - 0.1
	6	0	0	0	0	0.5	0	0.1	0	0	0	---
	7	0	0	0	0	0	0	0	0	0	0	---
	8	0	0	0	0	0	0	0	0	0	0	---
	9	0	0	0	0	0	0	0	0	0	0	---
	11	0	0	0	trace	trace	0	0	0	0	0	---
	12	0	0	0	0	0	0	0	0	0	0	---
	20	0	0	0	trace	0	0	0	0	0	0	---
	23	0	0.1	0	0	trace	0	trace	0	0	0	---
	24	0	0	0	0	0	0	0	0	0	0	---
	30	0	0	0	trace	0	0	0	0	0	0	---

** Partial 24-hour sample

COMPOSITE CHART OF AMINO ACID EXCRETION (cont'd)

amounts expressed in mg/ml (approximate concentration)

Case	Day of Illness	Cysteic Acid	Aspartic Acid	Glutamic Acid	Glycine	Taurine	Alanine	Valine	Leucine	Unknown Below Alanine	Unknown Below Valine	Other
7	6 hr.	0	0	0	0	0.1	0	0	0	0	0	---
	1 1/2	trace	trace	trace	0	0.5	0	0	0	0	0	---
8	6 hr.	0	0.1	0.25	0.25	trace	0.1	trace	trace	0	0	---
9	6 hr.	0	0	0	0	0.25	0	0	0	0	0	---
	1	0	0	trace	0.5	0.25	trace	0.1	0	0	0	---
10	6 hr.	0	0	0	0	0	0	0	0	0	0	---
	1	trace	0	0	0	0.25	0	trace	0	0	0	---
Controls 3 Cases 8-24 hrs.		0	0	0	0.1-0.25	trace	trace 0.1	trace	trace	0	0	serine-tr. -alanine-tr. arginine-tr.-0.1

THE ELECTROPHORETIC PATTERN OF PLASMA PROTEIN

Sample	Days After Accident	Non-protein Nitrogen mg	Total Protein g	Albumin g	Globulins g %			A/G
					α	β	γ	
Case 3	9	163.0	7.03	2.78	2.64	.75	.85	.65
Case 5	10	28.2	6.15	3.34	1.07	.57	1.19	1.19
	24	29.6	5.73	3.32	.88	.63	.91	1.38
	28	21.4	6.16	3.39	1.17	.71	.90	1.19
Case 6	10	33.8	5.99	2.99	1.15	.65	1.22	1.00
Controls Avg. - 6 males			6.45	3.57	.96	.98	1.06	1.24

are given elsewhere (Schwartz, 1949). Relatively slight changes in the urorosein and other pigments were found only in the two fatally injured patients. Since the urinary urorosein content seems to be related to the nutritional state, it is not surprising to find only mild changes in urorosein excretion in these acutely ill patients who were previously in good health. In contrast, a marked increase in urinary pigments has been observed in chronically ill hospital patients who were undernourished at the time of radiation therapy (Schwartz, 1949).

5. Amino-Aciduria

A quantitative determination of the individual amino-acid content of the urine samples was made by paper partition chromatography according to the scheme used by Dent (1946-b). The individual amino acids were identified by comparing their positions on filter paper with those of a synthetic mixture of pure amino acids studied simultaneously with the same solvents. A rough quantitative estimate was made by matching the intensity of the color of the ninhydrin reaction used to demonstrate the presence of the urinary amino acid with that of known amounts of pure amino acids. Although these studies were carried out on urine samples which had been kept for more than a year at 4°C, the consistent results of determinations made on the same specimens over a period of several months suggests that no marked loss of amino acids had occurred.

Studies of the amino acids in the urine of normal persons by partition chromatography show measurable quantities of glycine and alanine, as well as traces of leucine and valine (Dent, 1946-a). The analysis of the urine of healthy people used as controls in this study largely confirms this finding. However, in addition to the four amino acids already mentioned, trace quantities of taurine were noted consistently and, occasionally, small amounts of other amino acids were detected in highly concentrated urine samples. In contrast to these findings in the normal, many amino acids were demonstrated in considerable quantities in the urines of Cases 3 and 4, and in a single specimen from Case 1. In these cases the following amino acids were identified: cystine (as cysteic acid), aspartic acid, serine, glycine, taurine, alanine, valine, leucine, hydroxyproline, arginine, and phenylalanine. Two amino acids were not completely identified but may have been alpha-amino-n-butyric acid and tryptophane.

The quantities of individual amino acids in the post-exposure urine specimens of the heavily irradiated patients varied from trace amounts to 3 milligrams of taurine per milliliter of urine in the sample passed by Case 1 the day before his death. In Cases 3 and 4, whose urinary output of amino acids was closely followed, the pattern of excretion was similar. In each case, abnormal amounts of amino acids were observed in the urine sample collected during the first post-exposure day. The degree of amino-aciduria increased, reaching a peak on the sixth post-exposure day in both cases. The amount of urinary amino acid was diminished in the ninth day specimen of Case 4. After the twelfth post-exposure day, the amino acid content of the urine of Case 4 decreased rapidly although traces of abnormally occurring amino acids were still evident during the fourth week after exposure. Besides the sustained amino-aciduria in these two patients, an inconsistent increase of individual amino acids was noted in the other survivors of the second accident shortly after exposure. Thus, Case 5 showed 0.25 milligrams of aspartic acid per milliliter of urine passed during the first 36-hour post-exposure period. Similarly, a measurable quantity of aspartic acid was demonstrated in the six-hour post-exposure sample of Case 8.

The amount of amino acids excreted in the urine depends largely upon the diet in normal persons, and on other factors in patients suffering from certain diseases. In healthy persons, it has been shown by micro-biologic assay that of the eight amino acids studied, two to six percent of the total quantity ingested as protein was excreted

482

65

in the urine (Sheffner, Kersner and Palmer, 1948). A marked increase in the quantity of urinary amino acids has been demonstrated in a patient with liver damage (Uzman and Denny-Brown, 1948) and in patients with Fanconi Syndrome (Dent, 1947). In the former case, the inability of the damaged liver to de-aminate plasma amino acids is assumed to be the most likely cause of the amino-aciduria; in the Fanconi Syndrome, on the other hand, a lowered kidney threshold for amino acids and other plasma constituents has been demonstrated as the probable etiologic factor responsible for the amino-aciduria. In neither instance was the degree of amino-aciduria as marked as that in the present cases. In contrast to that observed in these pathological states, amino-aciduria has not been described in patients with thermal burns (Taylor, et al., 1943-b).

The significance of the amino-aciduria in patients with the acute radiation syndrome is not entirely clear; it may have been the result of liver damage, of enhanced protein degradation, or of both. Conclusive evidence of liver damage was present in Case 3 and it seems likely that similar, though less marked, damage occurred in Cases 1 and 4. Although protein metabolism was not studied in these patients, data obtained from animal experimentation, i.e., nitrogen balance studies in dogs (Prosser, Painter and Swift, 1947-b), and investigation of protein turn-over using radioactive alanine (Hempelmann, et al., in press) suggest that irradiation accelerates protein degradation. Thus, it seems likely that the amino-aciduria in the present cases may be explained by enhanced protein breakdown occurring in persons with liver damage.

6. Potassium

Quantitative studies of urinary potassium were carried out in the urine samples of Cases 3 and 4 three years after the accident. Since the potassium intake of these patients is not known, it is impossible to draw definite conclusions about their potassium balance. However, the potassium excretion in Case 4 was higher than that usually observed in healthy persons on a normal diet. The urinary potassium in Case 3, however, was elevated only in the sixth day post-exposure specimen. Although a high potassium intake might account for the elevated urinary level in Case 4, the weight loss in this patient suggests that the increased excretion was related at least, in part, to cellular breakdown (see Section V). His weight loss, about one pound of weight a day, if due solely to tissue breakdown, would be responsible for a daily output of 50 milliequivalents of urinary potassium. Since the caloric intake of this patient was not reduced except during the first day and again from the fifth to the tenth day, it seems probable that at least some of the urinary potassium can be accounted for by degradation of protoplasm.

In thermally burned patients, however, the potassium balance is maintained except for a transient negative phase shortly after injury (Moore, et al., in press). There is no correlation in cases of thermal burns between the potassium and nitrogen balance. The latter is often negative for several weeks after injury.

7. Summary

The more heavily irradiated patients showed an increased excretion of urinary corticoid-like substance. This presumably indicated a change in function of the adrenal cortex which persisted for four weeks in Case 5. Increased amounts of coproporphyrin (mainly Type III), and urobilinogen were found in the urine of Case 3, denoting liver damage and other organ dysfunction. Mild transient changes in the porphyrin and urobilinogen excretion of questionable significance were found in Case 4. Minor changes in urinary urorosein were observed in Cases 1 and 3. Amino aciduria, presumably due to liver damage and increased protein breakdown, was pronounced in Cases 1, 3, and 4. The potassium excretion of Case 4 was higher than that usually observed in healthy people on a normal diet.

VIII. INDUCED RADIOACTIVITY

One of the most interesting features of this series of cases is the neutron induced radioactivity of the tissues of these patients. Besides being a unique observation in itself, the study of the induced activity proved to be of great value in calculating the radiation dosages. Because of its importance in this respect, and because the neutron doses accounted for most of the energy dissipated in the bodies of the present patients, the problem of interaction of neutrons with tissue will be briefly considered. A more thorough discussion of the problem can be found in an article by Zirkle (1947), as well as in any of the standard textbooks on nuclear physics (Lapp and Andrews, 1948).

A. Interaction of Fast Neutrons and Tissue

When neutrons interact with tissue, several kinds of nuclear reactions take place. Before such reactions occur, however, it is usually necessary for the fast neutrons to be slowed down by collisions with atomic nuclei, chiefly with those of hydrogen atoms. The nuclei involved in such a collision are knocked out of the molecule of which they form a part and are stripped of their orbital electrons. These electrically charged hydrogen nuclei (protons), which recoil at high speeds, produce short but dense ionization tracks in the tissue. The ionization leads to chemical changes in the protoplasm. This mechanism of energy transfer by means of the recoil process accounts for almost half of the total energy dissipated in the bodies of most of these patients.

B. Interaction of Slow Neutrons and Tissue

After a number of collisions, the speed of the neutrons is greatly reduced and they are then easily captured by the nuclei of tissue atoms. Atoms differ greatly in their ability to capture slow neutrons and in the nuclear transformation which results from such captures. Three types of slow nuclear reactions predominate in a tissue subjected to neutron bombardment.

1. The (n, p) Reaction

Some atomic nuclei (mainly nitrogen and carbon in the case of aqueous tissue) give off energetic protons after capturing a neutron. The protons produce tissue damage in the same way as do the recoil protons which result from the collisions between fast neutrons and hydrogen atoms. Ionization from this type of nuclear reaction (called the (n, p) reaction, or neutron-in, proton-out reaction) contributes only a small fraction to the total ionization which occurs when tissue is bombarded with fast neutrons.

2. The (n,γ) Reaction

Other nuclei emit energetic gamma rays upon capture of a neutron. The (n, γ) (neutron in, gamma ray out) reaction occurs mainly in tissue hydrogen.. Capture by hydrogen is the ultimate fate of the vast majority of neutrons which enter the body. The gamma ray emitted, as a consequence of the reaction, is very hard (2.2 million electron volts). After emission of the gamma ray, the hydrogen nuclei which have captured neutrons are transformed into deuterium, the heavy stable isotope of hydrogen. A gamma radiation from neutron capture in hydrogen contributes heavily to the total radiation dose of a body irradiated by fast neutrons. In most of the present cases, it accounted for about one-quarter of the total dose.

3. The Neutron Capture Reaction

A third class of atomic nuclei become radioactive as a result of the nuclear reaction. Chlorine, sodium, phosphorus, potassium, and sulfur are the outstanding elements in tissue which undergo such a reaction with the formation of radioactive

482-67

isotopes. The intensity of activation decreases in the order of the elements named. The way in which such radioisotopes can be used in computing the radiation dose is discussed below.

All of the above nuclear reactions occur during neutron bombardment and the chemical changes that result are responsible for the ultimate tissue damage.

C. Use of Neutron-Induced Activity in Calculating Radiation Dosages

The production of radioactive isotopes in the body is of great practical importance in this study, since measurable radioactivity of the tissues persists for a considerable period of time after exposure of the body to neutrons. Since the quantitative aspects of nuclear reactions are reasonably well known, the determination of the induced radioactivity provides data from which the number of neutrons that entered the body can be calculated, provided that the average energy of the incident neutrons is known. In these studies, only the radioactivity of the sodium and phosphorus was determined and the former was used in the calculation of the neutron dosage. Radiochlorine, which decays rapidly (37-minute half-life) was not studied for practical reasons. The specific activity of sodium and phosphorus, i.e., the ratio of the number of radioactive to stable atoms, in blood serum and in the urine of these patients are given in Tables I, II, and III. It should be pointed out that these values are not the only ones determined, but were selected as being the most valid and representative. The decay curves of blood serum and urine samples are given in the following charts. In these graphs, the radioactivity measurements were determined experimentally and the theoretical decay curve of sodium and phosphorus, indicated by the straight lines, have been fitted to them.

The assessment of the whole-body dosage due to a fast-neutron dose can be made by either of two methods. First, the induced radioactivity can be measured in samples of serum and urine. Second, the induced radioactivity can be estimated by measurement of the gamma radiation emitted from the body as a whole at selected distances. In both methods, the induced radioactivity is a measure of how many slow neutrons existed in the tissue. From this number of slow neutrons, the total-radiation dose can be estimated. The first method is more accurate, but requires a more elaborate measuring system. It has been suggested earlier that the specific activity of serum sodium, so determined, be used as an empirical unit of neutron dose in radiation exposures of this type. The second method employs only a portable gamma-roentgen survey meter and can be carried out quickly.

D. Contribution of Radiation From Induced Tissue Activity to Total-Body Dose

The radiation dose received by these patients from the induced radioactivity of their tissues has been mentioned briefly. It has been calculated that radiation from the radioactive elements themselves could not have accounted for more than one percent of the total dose received by the body (Hoffman, 1947). It is probably true that the irradiation of tissues adjacent to structures containing large amounts of phosphorus (bones and teeth) may have been greater than that from the radioactivity in the soft tissues but it is improbable that the dosage was significantly raised even in these regions. It is certainly true, however, that there was a high radiation level around the gold inlays in the teeth of these patients. This is to be expected since gold atoms are known to have a much larger capture cross section for slow neutrons than do most elements in the body. It has been pointed out that a small ulcer was produced in the mouth of Case 3, presumably by the radiation given off by a gold filling.

E. Unusual Observations Related to Induced Radioactivity

Examination of the data in the tables on induced radioactivity discloses four interesting observations, three of which cannot be explained with certainty in the light of our present knowledge. A discussion of these interesting phenomena follows.

TABLE I

Na^{24} and P^{32} Activities in Serum

Case	Sample	Time taken (hours after exposure)	mg Na/cc	cc Plated	mg Na Plated	Na^{24} cpm@ t = 0	Na^{24} Corr.* dps	Na^{24} Corr.* dps/mg Na	P^{32} cpm/mg P @t = 0	P^{32} Corr.* dps/mg P
1	Serum I	17.4	-	5	16.8**	900	300	18.0	-	-
2	Serum I	17.4	-	5	16.8**	56	18	1.1	-	-
3	Serum I	1.2	3.22	3	9.66	6400	711	73.6	103**	12.8
	II	46.9	3.09	5	15.45	10,240	1137	73.6	bkg.	bkg.
	III	66.1	3.03	5	15.15	10,700	1177	77.8	-	-
4	Serum I	1.1	3.20	3	9.60	1150	128	13.3	bkg.	bkg.
	II	46.8	3.27	5	16.35	2100	222	13.6	-	-
	III	65.8	3.13	5	15.65	1800	200	12.8	-	-
5	Serum I	1.4	3.36	3	10.08	900	100	10.0	bkg.	bkg.
	II	46.6	3.22	5	16.10	700	77.8	4.83**	-	-
6	Serum I	1.9	3.25	3	9.75	620	69	7.1	bkg.	bkg.
	II	46.7	3.16	5	15.80	540	60	3.8***	-	-
7	Serum I	1.8	3.22	3	9.66	260	29	3.0	-	-
8	Serum I	1.5	3.28	3	9.84	180	20	2.03	-	-
9	Serum I	2.0	3.24	3	9.72	135	15	1.52	-	-
10	Serum I	1.4	3.18	3	9.54	110	12.2	1.22	-	-

* Corrected for counter efficiency of 0.15

** Based on a 3 cc sample of serum which had 0.39 mg phosphorus and 40 cpm at t = 0

*** Counting rate less than 100% of background

482 69

TABLE II

P^{32} Activities in Urine

Case	Sample	Time taken (hours after exposure)	mg P/cc	cc Plated	mg P Plated	P cpm @ t = 0	P Corr.* dps @ t = 0	P Corr.* dps/mg P
1	Urine I	17.4	-	25	5.3	1800	600	110
2	Urine I	17.4	-	25	5.3	70	25	4.7
3	Urine I	1.2	-	-	-	11,600	1290	-
	(pooled) II**	3.8	-	-	-	3200	356	-
	III	5.3	0.256	5	1.28	4120	457	357
4	Urine I	1.2	1.22	5	6.10	2630	290	47.5
	II	3.1	0.101	5	0.505	310	34.4	68.1
5	Urine I	1.9	1.37	5	6.85	360	40	5.84
6	Urine I	2.2	0.735	5	3.67	450	50	13.6
7	Urine I	1.2	1.27	5	6.35	170	18.9	2.97
8	Urine I	2.2	1.32	5	6.60	150	16.7	2.53
9	Urine I	1.2	1.12	5	5.60	70	7.8	1.4
10	Urine I	2.2	1.12	5	5.60	70	7.8	1.4

* Corrected for counter efficiency of 0.15
** Specimens passed at 1.2 and 3.8 hours were pooled

TABLE III

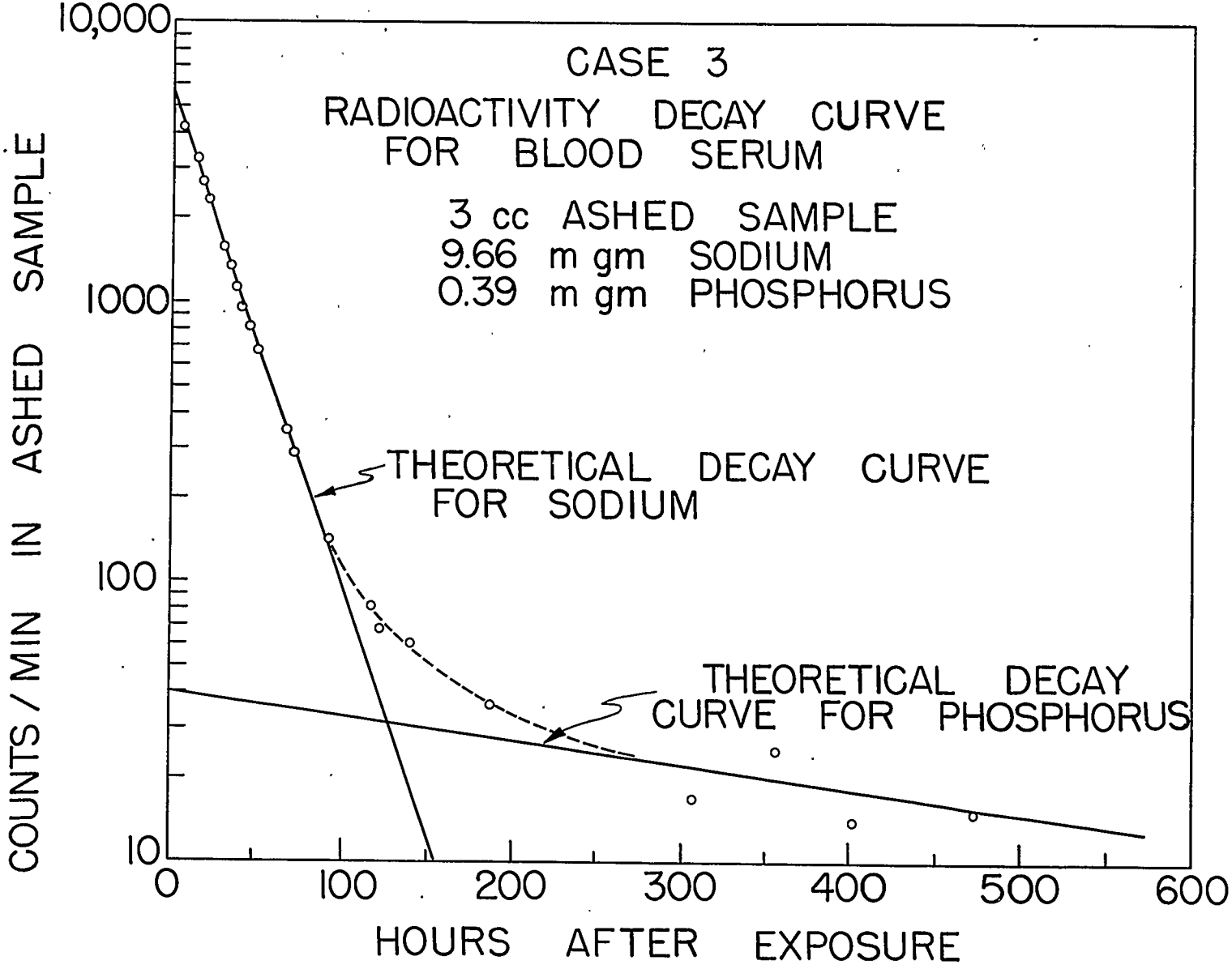
Na^{24} Activities in Urine

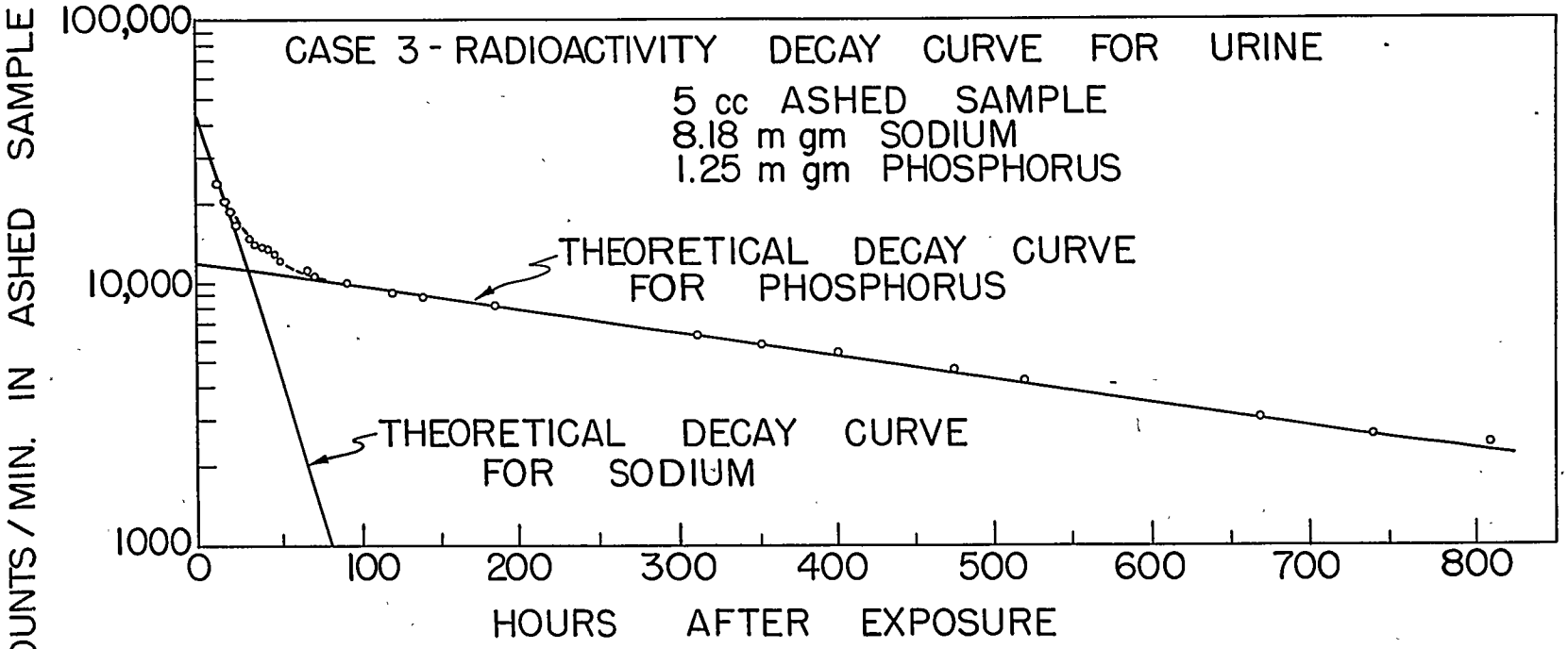
Case	Sample	Time taken (hours after exposure)	mg Na/cc	cc Plated	mg Na Plated	Na^{24} cpm @ t = 0	Na^{24} Corr.* dps @ t = 0	Na^{24} Corr.* dps/mg Na
1	Urine I	17.4	3.00**	25	75	5100	1700	22.5
2	Urine I	17.4	3.00**	25	75	320	102	1.38
3	Urine I	not run						
	II	3.8	1.65	5	8.18	18,300	2033	248
4	Urine I	1.2	2.94	5	14.7	2000	222	15.1
5	Urine I	1.9	4.87	5	24.3	3000	333	13.7
6	Urine I	2.2	6.01	5	30.1	2100	233	7.75
7	Urine I	1.2	1.62	5	8.1	490	43.3	5.35
8	Urine I	2.2	3.32	5	16.6	500	55.6	3.35
9	Urine I	1.2	2.79	5	13.9	240	26.7	1.92
10	Urine I	2.2	2.98	5	14.9	170	18.9	1.27

* Corrected for counter efficiency of 0.15

** Inert sodium not determined chemically but assumed from tables of normal values

482 71


CASE 3
RADIOACTIVITY DECAY CURVE
FOR BLOOD SERUM
3 cc ASHED SAMPLE
9.66 m gm SODIUM
0.39 m gm PHOSPHORUS
THEORETICAL DECAY CURVE
FOR SODIUM
THEORETICAL DECAY
CURVE FOR PHOSPHORUS
COUNTS / MIN IN ASHED SAMPLE
10,000
1000
100
10
0
100
200
300
400
500
600
HOURS AFTER EXPOSURE


CASE 3 - RADIOACTIVITY DECAY CURVE FOR URINE
5 cc ASHED SAMPLE
8.18 m gm SODIUM
1.25 m gm PHOSPHORUS
THEORETICAL DECAY CURVE FOR PHOSPHORUS
THEORETICAL DECAY CURVE FOR SODIUM
COUNTS / MIN. IN ASHED SAMPLE
100,000
10,000
1000
0
100
200
300
400
500
600
700
800
HOURS AFTER EXPOSURE

1. The Constancy of the Specific Activity of Serum Sodium

The specific activity of the sodium in the blood serum of Cases 3 and 4, if corrected for decay, shows relatively little change in the period lasting from 1.2 to 72 hours after the accident. The fall noted in Cases 5 and 6 is probably not as reliable, since the latter determinations were made on samples which had a counting rate of less than 100 percent of background. Thus equilibrium was established quickly between the inert sodium and the radioactive isotope. The lack of change in the specific activity of the serum sodium indicates that this radioisotope could not have been selectively excreted by the kidney; therefore it presumably existed in the usual chemical forms. The observed rapid equilibration of radiosodium is consistent with the animal studies of the fate of intravenously injected radiosodium chloride (Manery and Bale, 1941). These investigations show that radiosodium diffuses quickly into most tissues, equilibrium with the normal isotope being established within 20 minutes after injection. In certain other tissues (bone, brain, and testes) the penetration of the sodium is slower and not complete for twelve hours. Since radiosodium was produced simultaneously in all sodium compartments of the present series of patients, it is easy to understand how a constant specific activity of the serum sodium was established so promptly. This constancy of specific activity in serum sodium makes it the logical measurement to use as a basis for neutron dose computations. In contrast to the rapidly mixing radiosodium, radiopotassium does not reach equilibrium with the body potassium until 36 hours after subcutaneous injection (Moore, 1946).

2. The Inequality of the Specific Activity of Serum and Urine Phosphorus

The specific activity of phosphorus is much higher in the urine than in the blood serum. In Case 3, the specific activity of the urinary phosphorus was 17.9 times higher than that in the serum. In all other cases, the radioactivity of urinary phosphorus was easily measurable, while that in the serum was at background level. A similar disproportion in the specific activity of urinary and serum phosphorus has also been observed in neutron irradiated animals (Perley and Langham, unpublished data). The inequality of the specific activity of phosphorus in these two-body fluids suggests that the radiophosphorus in the blood existed in a chemical form that was more efficiently excreted by the kidneys than was the major part of the inactive serum phosphorus. The chemical form in which radiophosphorus exists after nuclear bombardment of tissue is not known, but the reaction discussed below suggests a mechanism by means of which it might be expected in an unusual chemical state.

Atoms which undergo nuclear reactions become dissociated from the molecules of which they formed a part (Szilard-Chalmers reaction) (Lapp and Andrews, 1948). When certain organic molecules are irradiated in the dry state with neutrons, a small fraction of the dissociated radioactive atoms are re-incorporated into the molecules in the original positions; thus, neutron bombardment of cystine results in the production of a small quantity of this compound containing radiosulphur in the usual position (Ball, Solomon and Cooper, 1949). In the case of water solutions of phosphorus compounds, an appreciable amount of restitution is not expected. The atoms of elemental radiophosphorus produced in the irradiated tissues as a result of the Szilard-Chalmers reaction can be expected to undergo chemical oxidative reactions with the formation of unknown phosphorus compounds. Some of these radiophosphorus-containing compounds may well be selectively excreted by the kidney.

The rapid selective excretion of radiophosphorus accounts for the low radioactivity of the tissues of Cases 1 and 3 at autopsy. Radiosodium would have decayed to an insignificant fraction of its original activity by the time of death of both patients, but physical decay was not responsible for the rapid disappearance of radiophosphorus, particularly in Case 3 where death occurred within one half-life of the radioisotope. The marked variation of phosphorous specific activity in two-body fluids would seem to make this isotope undesirable for use in neutron dose computations.

3. The Unexpectedly High Specific Activity of Urine Sodium in Case 3

The specific activity of the sodium is higher in the urine of Case 3, voided immediately after exposure, than that measured in the serum. This was not the case, however, in the other patients whose urine was similarly studied. This phenomenon cannot be explained in the same way as was the increased specific activity of urinary phosphorus. It seems likely that the high sodium specific activity in the early urine sample was the result of neutron irradiation of urine present in the bladder at the time of the accident. The severe reaction of the lower abdomen of Case 3 indicates intense irradiation of this part of the body, so that it is highly probable that the bladder urine was heavily bombarded by neutrons. The explanation fits the facts of the case, although evidence in favor of it is admittedly circumstantial, since the radioactivity of urine samples of Case 1 passed after the first few hours was not studied.

It should be pointed out that the use of the specific activity of urinary sodium rather than serum sodium in neutron dose computations would not have changed the doses much except in Case 3 where the dose would have been raised by a factor of 3.4.

4. Relative Specific Activities of Serum Sodium and Phosphorus Compared with the Known Neutron Capture Cross Section of These Elements

The relative specific activity of serum sodium and phosphorus observed in Case 3 is not consistent with the physical data concerning their ability to capture slow neutrons. Thus, the capture cross section of sodium for slow neutrons is known to be 90 times that of phosphorus. This means that when sodium and phosphorus are exposed to the same neutron flux in a physical experiment, the activity per milligram of sodium should be 90 times that of phosphorus. Instead of following the physical laws, the specific activity of serum sodium in Case 3 is only 5.6 times as great as that of the serum phosphorus. This single clinical observation is supported by a number of similar findings in animals exposed to neutrons (Perley, Langham and Hoffman, unpublished data). The biological factors responsible for this finding are not evident unless one supposes a very rapid transfer of P^{32} from tissues to the blood stream comparable to that from the blood stream to the urine which must have occurred in the kidney.

F. Summary

Fast neutrons interact with tissue by losing energy through collisions with light nuclei, chiefly hydrogen. The charged recoil proton, which results from the collision, causes dense ionization and hence tissue damage. When the fast neutrons have been slowed down they are captured by atomic nuclei and the resultant nuclear reaction may be one of three types; (1) a proton may be given off, (2) a gamma ray may be emitted at the time of the reaction, and (3) a radioactive isotope may be formed which decays according to a well understood physical law. The second reaction appears to be the most important way in which the slow neutrons cause tissue damage. The third reaction is important in these cases because it provides a measure of the number of slow neutrons which were present in the body. Four interesting phenomena are discussed in this section; (1) the failure of the specific activity of the serum sodium to decrease during the first few days after exposure, (2) the selective excretion of radiophosphorus by the kidney, (3) the high specific activity of the sodium in the urine in Case 3 but not in the other cases, and (4) the discrepancy between the measured specific activity and neutron capture cross section of sodium and phosphorus.

IX. DISCUSSION OF PATHOLOGICAL FINDINGS

The histopathological observations have been recorded in considerable detail in Section III (Vol. I). It can be seen by referring to the review article by Warren (I-XV, 1942-3), the book by Bloom (1948). and the pathological study of the Japanese patients (Liebow, Warren, and DeCoursey, 1949), that the present observations, with few exceptions, conform to or complement the information in the medical literature concerning the effect of radiation on normal tissues. The present discussion correlates the pathological findings with the clinical observations in these cases and emphasizes the unusual features of the histologic observations.

A. Methods

The postmortem examinations in Cases 1 and 3 were performed about one hour after death. The examinations were complete with the exception of the cranial cavities and the organs of the neck in both cases, and the hand tissues in Case 3. The tissues were fixed in Helly's fluid, absolute alcohol, and 10 percent neutral formalin, and then embedded in celloidin. The sections were stained according to Maximow's hematoxylin-eosin-azure II method.

B. General Features of the Tissue Reaction

As a result of a study of the histologic changes observed in Cases 1 and 3, certain general statements can be made about the effect of radiation on tissues. These observations are not new, but are presented briefly in order to tie together aspects of these cases that have not been discussed up to this time.

1. Suppression of Mitoses

The prompt effect of ionizing radiation in suppressing mitosis is well known, and the varying rate of recovery of different tissues from this effect also has been established. In general, it appears that the cell types with high mitotic activity (and presumably short life span) show more rapid recovery from this action of radiation than do the more slowly proliferating, longer-lived cells (Knowlton and Hempelmann, 1949). It is not surprising, therefore, to find that considerable recovery of mitotic activity had taken place in the 10 to 24 days between exposure and the time of death in Cases 1 and 3. The incidence of cell division in most tissues of these cases, although not measured exactly, did not appear decreased, except in the skin of Case 3, and in the few remaining marrow cells of the same case. Apparently, the mitotic activity of the lymphoid tissue of Case 3 was even greater than normal. Such delayed "overcompensation" of mitotic activity is well known (Canti and Spear, 1929).

Before placing too much emphasis on the quantitative aspects of tissue proliferation, one must consider certain features of the mitotic activity in these damaged tissues and in autopsy material in general. In the present cases, a great many dividing cells, particularly those in the lymphoid tissues, were in a dying or disintegrating phase which may be presumed to be more prolonged than the usual brief period of mitosis (15 to 90 minutes). Doubling or tripling the mitotic time would increase the number of dividing cells visible at any given time, even though the mitotic activity of the tissue was unchanged. Thus, the incidence of mitoses in these damaged tissues may give a falsely high impression of the proliferation rate. It should also be stressed that the difference in time elapsing between exposure and death of these patients prevents one from comparing the degree of initial suppression, since more recovery from this suppression of mitotic activity had taken place in Case 1 than in Case 3. Finally, it must be remembered, that, in considering mitotic activity in autopsy material, some of the dividing cells are able to complete mitosis in the time elapsing between death of the patient and fixation of the tissues. At least, this has been observed in the human epidermis (Thuringer, 1928). Thus, the mitotic activity in both cases, as demonstrated in sections taken at autopsy may be lower

than it actually was during life. The conflicting factors just reviewed show the difficulties one encounters when trying to interpret quantitatively the influence of radiation on the cell proliferation in these cases.

2. Cell Damage and Death

Cell damage and death is obviously the fundamental lesion responsible for the tissue and constitutional responses observed in irradiated individuals. Tissue regeneration and even the alleged instances of stimulation of cellular activity are secondary to this cell-damaging action of radiation. In many cases, cell death is of the kind which is observed in normal tissues and is unusual in irradiated tissues only in its high numerical incidence. In contrast to this apparent acceleration of the normal process of cell death, an unusual type of cytolysis occurs which, while not unique in radiation injury, is not caused by the commonplace toxic agents. The nuclear and cytoplasmic aspects of both these types of death are now described.

a. Nuclear Changes

(1.) Usual type of nuclear disintegration: The changes in the nuclei of dying cells consist of swelling, pyknosis, hyperchromatosis, karyorrhexis, and karyolysis. As pointed out above, they are indistinguishable, in the usual histologic sections, from occasional nuclear changes seen in normal tissue except for their abnormality in numbers. Such disintegration is most prominent in the radiosensitive tissues and presumably represents death of the cell during its resting phase.

(2.) Unusual or chromosomal type of nuclear damage: As discussed in Section IV, cytolysis due to chromosomal change is a common form of death in radiation-injured cells. As is expected, these changes are found primarily in the tissues which normally are actively proliferating. The damaged nuclei, sometimes called "owl's eye" nuclei, are greatly swollen and often appear to be in the process of "exploding". Fragmentation of chromosomes, bridging between chromosome fragments, and multipolar mitotic figures are conspicuous, as is the swelling of the nuclear material and chromosomal remnants in dying or dead cells. As will be brought out later, many of the daughter cells of these abnormal mitoses appear to be nonviable. Sometimes nuclear division proceeds without cytoplasmic separation and double or multilobed nuclei can be observed in a single cell. The mitotic aberrations which can be seen in the photomicrographs of Cases 1 and 3 are unusually well illustrated in the monograph by Politzer (1934) and are also described by Miescher (1925) and Harperl (1936). Although this type of nuclear change is the most unusual aspect to radiation-induced cellular injury, it must be emphasized that the reaction does not represent a specific effect of radiation. Identical changes can be found in the proliferating tissues of patients or animals treated with nitrogen mustards or aminopterin.

b. Cytoplasmic Changes

(1.) Evidence of cytoplasmic dysfunction: The changes seen in the cytoplasm of damaged cells which show either orderly nuclear disintegration or chromosomal abnormalities, are in no way characteristic of radiation injury. Thus, vacuolization, increased basophilia, and other specific indications of cytoplasmic modification, e.g., waxy degeneration of muscle fibers, can be found as a result of many types of tissue injury. It was once believed that the increase in size of these injured cells was related solely to the increased intracellular osmotic pressure, but it has since been shown that the swelling cannot be explained by water imbibition alone (Failla, 1940). That synthesis of protoplasm sometimes continues in damaged cells is shown by the fact that, under certain conditions, irradiated yeast cells may achieve as much as ten times their normal volume without apparent dilution of their protoplasm (K. Brace, personal communication from R. E. Zirkle). The increased basophilia, which is particularly conspicuous in tissue lymphocytes, epidermal cells, and

proliferating connective tissue cells may well be the result of the accumulation of ribonucleic acid in the cytoplasm (Mitchell, 1942). Sometimes the basophilia is more intense than that seen in actively proliferating, relatively undifferentiated cells, but often it is indistinguishable from that seen in these normal stem cells.

(2.) Cellular differentiation: Recently, in the British literature, emphasis has been placed upon the action of radiation in causing differentiation of of anaplastic tumor cells (Glucksmann, 1946). Such an effect, while it may be striking in neoplastic tissue, would, of course, be difficult to detect in most adult tissues. Thus, although one may consider the metaplasia of the duct epithelium of the sweat glands to be a similar phenomenon, such a deviation from normal should not be interpreted as true cellular differentiation.

3. Interstitial Tissue Response

Most of the connective tissue elements of Case 3 showed a striking degree of edema which was associated with dilatation of blood vessels and lymphatics. In contrast, the interstitial tissues of Case 1 showed little edema. The edema in Case 3 may have been related, in part, to the circulatory collapse and therefore, not be solely the direct response of irradiation. Although discrete points of gross and microscopic hemorrhage were evident in Case 3, there was no generalized extravasation of erythrocytes into the interstitial tissues in either case. There was some early hyalinization of collagen fibers, particularly in the regions around the sweat glands and hair follicles in the dermis of the more heavily irradiated areas of the abdomen of Case 3. There was no apparent increase in the amount of interstitial metachromatic staining ground substance as had been previously reported in animals (Sylven, 1940). In contrast to the relatively minor changes in the interstitial tissues of the body, there was widespread necrosis of the connective tissues of the right hand of Case 1.

4. Blood Vessel Changes

Except in the heavily irradiated hand tissues of Case 1, the vascular changes, aside from prominent dilatation, were limited to swelling and occasional death of the endothelial cells in the smaller vessels. There was no endothelial proliferation, or thickening and hyalinization of the walls of blood vessels such as has been described as a late effect of chronic irradiation (Wolbach, 1909). Similarly, the adventitial connective tissue changes and loss of collagen in the media described in animals as a consequence of the exposure to radiation in the thirty day median lethal dose range (Bloom, 1948) were not observed.

In contrast to this limited effect on the blood vessels of the body tissues, vascular changes were pronounced in the hands of Case 1. These changes, which were quite similar in appearance to those found in frostbite and immersion foot, consisted of necrosis of the vessel walls and thrombosis in small and large vessels. It is apparent that most, if not all of the late gangrene of the hand tissues of Case 1 was due to interruption of the blood supply to these tissues.

5. Cellular Reaction

a. Inflammatory Cells

There was no generalized infiltration of the tissues with leucocytes except for occasional plasma cells and other unidentified mononuclear cells, presumably of the lymphoid series. In the acutely inflamed tissues of Case 1, as in the exposed dermis, the cellular reaction was relatively scant and was composed predominantly of mononuclear cells. Polymorphonuclear leucocytes were rare in these regions as well as in the pneumonic patches and denuded submucosa of the intestines of Case 3. This is consistent with the terminal agranulocytosis which occurred in both instances.

b. Mast Cells and Their Probable Relationship to the Anticoagulant in the Circulating Blood

The occurrence of large numbers of mast cells in almost all of the tissues of Case 3 and of a smaller, but nevertheless definite, increase in number in the tissues of Case 1 is a striking finding which deserves further comment, particularly in so far as its relationship to the heparin-like material in the circulating blood is concerned. This observation of mast cell increase is not unexpected, however, since a similar finding was reported in the study of the Japanese killed by radiation (Liebow, Warren and DeCoursey, 1949).

Tissue mast cells are usually not conspicuous in human autopsy material stained by the usual methods, but they can be easily demonstrated in histological sections of connective tissues stained with metachromatic dyes (Janes and McDonald, 1948). The number of mast cells is increased in tissues which show a chronic inflammatory reaction; they are not increased, however, in acutely inflamed tissues (Janes and McDonald, 1948). Other examples of pathological states in which mast cells play a conspicuous part are urticaria pigmentosa in the human (Lever, 1949), and mast cell tumors in dogs (Bloom, 1942).

This increase in the mast cells is interesting because of its probable association with the anticoagulant demonstrated in the blood of these patients. The intimate relationship between mast cells and heparin has been known for some time (Jorpes, 1946), as has been demonstrated by experiments which show that the heparin content of tissues parallels the number of mast cells seen in histological sections (Holmgren and Wilander, 1937; Wilander, 1938). Recent studies in which active heparin has been extracted from mast cell tumors in dogs (Oliver, Bloom, and Mangieri, 1947), and from a human scrotum which contained numerous mast cells but no thrombi (Ehrich, et al., 1949) support the contention that tissue mast cells contain heparin. It appears to be more than coincidence, then, that the mast cell content of the tissues of Cases 1 and 3 can be correlated with the presence or absence of a defect in the clotting mechanism of the blood. In Case 3, whose blood was uncoagulable at autopsy, there was a great increase in the number of tissue mast cells, while in Case 1, in whom there was no apparent increase in the blood clotting time at death, the mast cells were less numerous. From this observation, as well as the experimental evidence just reviewed, it seems likely that the anticoagulant in the blood of Case 3 was related to, if not derived from, the tissue mast cells.

It is necessary to consider how mast cell heparin reaches the blood stream. An increased coagulation time is not found either in dogs with mast cells tumors, or in patients with urticaria pigmentosa. Therefore, one must examine the conditions under which the mast cells discharge heparin into the blood stream. Observations on tissue cultures of mast cells from dog tumors indicate that the cells do not release heparin as long as the cultures are maintained in good condition (Paff and Bloom, 1949). When the cultures become old, however, extracellular heparin can be demonstrated. In vivo damage to tissues by irradiation with x-rays results in an increase in the metachromatic interstitial substance with a concomitant decrease in the number of metachromatic intracellular granules (Sylven, 1940). These observations together with the instances in the present cases of rupture of the cytoplasmic membrane with discharge of metachromatic granules into the interstitial spaces, if this can be accepted as an antimortem change, suggest that degenerating mast cells can release heparin into the body system.

It should be emphasized again that recent studies show that an increase in circulating anticoagulant is often not observed in irradiated animals (Cronkite, 1950). The mast cell content of the tissues of such animals has not been reported.

C. Skin

The histopathology of acute radiation damage to the skin has been well described and the pertinent literature discussed in a review by Montgomery (1946), as

well as in a summary by Warren (1942-3). The acute exudative and ulcerative dermatitis following x-ray, gamma ray, or neutron irradiation involves both the epidermis and its appendages and the supporting tissues. As has been pointed out in Section IV, the cellular response in both the epithelium and in the dermis becomes manifest only after a latent period of a week or more except when the radiation dose is overwhelming. It is in this respect that radiation "burns" usually differ from thermal burns, in which gross pathologic changes are usually seen immediately after the injury. The usual type of acute radiation burn produced by moderately large doses of radiation, in contrast, is characterized by its late development and by multiple, successive waves of ulceration and repair, a process that may continue for many months or years.

Although the radiation received by Cases 1 and 3 differed considerably in amount, skin doses of the body in both cases were so large that considerable cell damage of a prompt character occurred; therefore, it seems reasonable to compare the pathology of the two cases directly and to speak of the evolution of certain lesions. The skin lesion in the abdomen of Case 3 nine days after exposure were characterized by: (a) cessation of mitotic activity in the epidermis and in the hair follicles; (b) necrosis and necrobiosis of epithelial cells with swelling, vacuolization of both the cytoplasm and nuclei, and loss of cellular differentiation; and, (c) thinning of the entire epidermis with a disappearance of the characteristic structural layers. These epithelial changes were accompained by edema of the underlying dermis and a resultant flattening of papillae, and by the early stage of separation of the epidermis from the dermis. In contrast to the relatively early yet striking and widespread changes of Case 3, the skin lesions in Case 1, twenty-one days after exposure, were advanced. Although severe ulcerative and exudative dermatitis was the predominant reaction, regeneration of epidermis and the formation of actual granulation tissue had begun at the edge of burned areas.

In both cases the striking metaplastic changes in the epithelium of the ducts of the sweat glands was the most unusual finding noted in the skin. Metaplasia of this type has not been described previously except for brief mention in an autopsy report of a Japanese in whom the process was believed to be related to the thermal component of the atomic bomb explosion (Liebow, Warren and DeCoursey, 1949). Although no dead or disintegrating cells were seen at the time of study in either of the cases under discussion, the metaplastic changes presumably occurred subsequent to destruction of the normal cuboidal epithelium of these ducts. These proliferative changes were in striking contrast to the degenerative and necrobiotic phenomena seen in other areas of the epidermis. Squamous cell proliferation of the ducts of the sweat glands was much further advanced in Case 1 than in Case 3, but no normal duct epithelium could be found in either case except in the skin from lightly irradiated areas. The progressive character of this squamous metaplasia is well illustrated by the way in which it had replaced the original lining and occluded the lumina of some of the ducts. This was evidence of the viability of these cells, which proliferated rapidly and aggressively enough, not only to fill the ducts, but also to replace or obscure the epithelial cells of the secreting portions of the sweat glands. It is possible that such occlusion of the ducts of the sweat glands may be responsible in part for the cessation of sweat production which is reported to occur in irradiated skin (Knowlton, et al., 1949).

It is interesting to speculate as to whether squamous metaplasia of epithelial cells of the ducts of the sweat glands has any bearing on the pathogenesis of carcinoma of the skin, a well known late sequel to irradiation injury. The cell proliferation in these cases is so active that one wonders if it might not have gone on to become a precancerous or cancerous lesion. Such changes, however, were not described by Wolbach (1925) in his studies of the pathogenesis of radiation-induced cancer.

482 80

D. Hematopoietic Tissue

Study of the histopathology of the radiosensitive hematopoietic tissue of Cases 1 and 3 brings out several interesting features:

1. In the first place, in both cases, which differed considerably not only in radiation dose but in the duration of life after exposure, the lymphoid tissue appeared to be less depleted of cells and more actively regenerating than the bone marrow. This is exemplified by the almost complete aplasia of the bone marrow of Case 3 in the face of hypoplastic but proliferating lymphoid tissue. Such a finding might seem paradoxical in view of the fact that lymphocytes are considered to be more radiosensitive than other blood cells. However, the lymphoid tissues are able to recuperate rapidly, and it must be assumed that the histopathological picture at the time of autopsy in each case represents recovery rather than absence of injury. This observation suggests that the stem cells in the marrow and lymphoid tissues, which are almost indistinguishable histologically, actually have different characteristics. Although both types appear to be relatively radioresistant, the stem cells in the lymphoid tissue apparently are somewhat less vulnerable. Such a difference in resistance between the stem cells of the bone marrow and lymphoid tissues is not in accord with the observations reported in the literature.

2. The proliferation of the lymphoid tissues brings up another interesting point, namely, that the mitotic activity of the lymphoid tissue is not normal with respect to the production of healthy, vigorous daughter cells which are capable of entering the blood stream. Morphologic evidence of the abnormality of cell division is found in the frequent chromosomal aberrations. It is apparent that many dividing cells die in mitosis, and that those which complete mitosis do not form normal-appearing daughter cells. Thus, many of the daughter lymphocytes have large, hyperchromatic, and even multiple nuclei, or they have developed into giant cells. Even the more normal appearing small lymphocytes often have double nuclei or an unusually large amount of basophilic cytoplasm. The failure of the lymphocyte count to rise in the face of proliferation of the parent tissue indicates either that the adult cells were not reaching the circulating blood stream, or that they were rapidly being destroyed once in the circulation.

3. A third interesting point about the hematopoietic tissue is that hyperplasia of the reticulum cells, which had been previously reported (Liebow, Warren and DeCoursey, 1949) was not observed in these cases. The radioresistant reticulum cells became more prominent in the lymphoid tissue and bone marrow because of depletion of the leucocytes, but hyperplasia of the reticulum was not evident. This is in contrast to observations in the Japanese which showed that reticulum hyperplasia had begun in the bone marrow of a few patients dying six days after exposure.

In addition to the interesting findings just noted, two facts stand out about the bone marrow response in Cases 1 and 3. First, the apparent damage to the reticulum cells in the irradiated vertebral bone marrow of Case 3 suggests that the radiation dose in this tissue was high, since the radioresistance of this type of cell is well known. This is in conflict with the concept that most of the radiation was

absorbed in the superficial layers of the anterior surface of the body (see Section II, Vol. I and Section X). Second, the great number of cells dying during mitosis in the marrow of Case 1 is an unexpected finding so long after exposure. Very likely, this is a manifestation of the cyclic destruction described in the bone marrow of animals (Bloom, 1948).

E. Reproductive Organs

The lesions in the reproductive organs are identical with those described in humans many years ago (Schinz and Slotopolsky, 1928) and with the well known testicular abnormalities in experimental animals, first described in 1903 (Albers-Schönberg). Sections of the testes of the two fatal cases clearly illustrate the sequence of events that takes place in germinal tissue exposed to an overwhelming dose of irradiation. It is irrelevant that the radiation dose probably differed in the two cases because, in both instances, it was undoubtedly much higher than that necessary to produce complete and probably permanent sterility.

In Case 3, a few poorly preserved and highly abnormal spermatocytes and even a few spermatogonia were observed among the clumped necrotic cells and debris within the lumina of the seminiferous tubules. In contrast, there was little evidence of cellular debris in Case 1, although a few swollen, degenerating germinal cells were seen. The lining cells of the tubules in the latter case consisted wholly of abnormally large Sertoli cells. In addition, there was hyalinization of the basement membrane, an indication of involution and atrophy of this organ. The sequence of events in these cases is in accordance with the histologic pattern noted in the Japanese. Sloughing and disintegration of the epithelium were evident in a Japanese patient dying four days after exposure. In other patients, the destruction of germinal epithelium continued well beyond the second week. In contrast to the tubular atrophy, the interstitial tissue in the Japanese appeared normal or hyperplastic.

Even though the testicular changes can be explained by the direct damaging action of radiation, indirect factors must be considered since these tissues are known to be very sensitive in their response to many diverse and generalized stimuli and injuries such as malnutrition, and acute or chronic infectious processes. It is highly unlikely, however, that such systemic factors played a primary role in the pathogenesis of the lesions noted in the testes of Cases 1 and 3, although they may have contributed to the over-all picture seen at autopsy.

The prolonged effect of radiation on the spermatogenic cells is shown by the sperm counts and biopsies in Cases 4 and 5. Why regeneration should be so markedly delayed in this tissue is not clear. Such a delay is in marked contrast to the other radiosensitive, actively proliferating tissues such as the lymphoid tissue, bone marrow, and intestinal epithelium which show prompt recovery (Liebow, Warren and DeCoursey, 1949).

F. Gastro-Intestinal Tract

The unfortunate loss of most of the intestinal tissues of Case 1 prevents a comparison of the histologic appearance in these two cases. However, it can be said from the examination of the gross specimens that the most intense reaction in Case 3 was in the jejunum and ileum, while that in Case 1 occurred in the colon. This observation in Case 3 is in keeping with the findings in the Japanese dying after the second week in whom the large bowel was the site of the most intense reaction.

The essential lesion in Case 3, however, was the almost complete loss of intestinal epithelium, particularly marked in the small intestine; there was also mononuclear cell infiltration, edema, and hyperemia of the submucosal tissues. The few remaining epithelial cells were mostly in the intestinal crypts and appeared to be severely injured. The most obvious abnormalities were the marked enlargement of the cell and the chromosomal aberrations. In a few portions of the jejunum and ileum, the ulceration had extended beneath the epithelium to the underlying submucosal tissues. It is possible that some of these ulcerations may have occurred in

disintegrated Peyer's patches. Myriads of bacteria were present on the denuded surface of the gut, but there was little bacterial invasion of the underlying tissues. This delay in the invasion of the gut by the intestinal bacteria, even in the absence of a cellular reaction, has been observed previously in irradiated animals, although not as late as the ninth day after exposure (Warren and Whipple, 1923).

In contrast to the almost complete destruction of intestinal epithelium in Case 3, the gastric mucosa showed relatively little damage. This is consistent with the observations in the injured Japanese patients.

G. Heart

Except for a spotty endocardial thickening of Case 1 and the petechial hemorrhages in Case 3, there were no significant lesions of the valves, the endocardium, or the myocardium in either case. The hemorrhagic fibrinous pericarditis of Case 1 appeared to be acute in some places and subacute, with early organization of the fibrinous exudate, in others. The process thus appeared to be at least a week to ten-days old, and must have started about the time of the tachycardia on the fifteenth post-exposure day. Except for the absence of bacteria and cellular reaction, there was nothing specific about the appearance of this pericardial reaction. The histological picture was compatible with uremic pericarditis, but it is known that there was no true azotemia at the start of the process and the good renal output even in the terminal stages of the illness suggests that nitrogen retention of a significant degree did not occur. Pericarditis of this nature has not been reported as a consequence of thermal or radiation injuries or as a concomitant symptom of the Wolff-Parkinson-White syndrome. The cause of this pericarditis, therefore, is not apparent although infection cannot be excluded even in the absence of demonstrable organisms in section stained with bacterial stains.

H. Adrenals

Since certain of the clinical and metabolic responses of these patients were assumed to be dependent upon adrenal cortical dysfunction, the study of the adrenals becomes a subject of great interest. Atrophy of the gland was described in the Japanese while increase in weight and loss of cholesterol have been reported in animals given lethal or sublethal doses of total-body radiation (Patt, et al., 1947). It is surprising, therefore, that the adrenal cortex of Case 1 showed no morphologic changes, particularly in view of the very high levels of excretion of corticoid-like substances. In contrast, the adrenal cortex of Case 3 showed evidence of marked damage, characterized by vacuolization and necrosis of cortical cells in both the zona glomerulosa and fasciculata, and edema of the cords. The similarity of this reaction to the "tubular degeneration" of the adrenal cortex observed in patients dying in a state of cardiovascular collapse resulting from overwhelming infection has been pointed out (Rich, 1944).

It is appropriate to bring up the role of the adrenal glands in the acute radiation syndrome at this time. This subject has been reviewed recently (Selle, 1949) and there seems to be little doubt that adrenal dysfunction contributes to the clinical response in irradiated animals. Thus, studies of the histologic changes in the heavily irradiated adrenal gland of rabbits show evidence of cell damage (Englelstad and Torgerson, 1937). Recent observations on the organ weight and cholesterol content (Patt, et al., 1947) attest to the hyperactive state of the gland in animals exposed to doses of x-rays in the median lethal range. Hypophysectomy, or the previous injection of adrenal cortical extract in rats wipes out the adrenal response (Patt, et al., 1948-b). In the terminal stage of the illness of fatally injured animals, the adrenal activity, as indicated by cholesterol content, may decline. This is in keeping with the concept of the exhaustion stage of the General Adaptation Syndrome. Although it has been reported that pretreatment with desoxycorticosterone protects mice against lethal doses of x-rays, recent experiments do not confirm this observation (Ellinger, 1947;

Straube, et al., 1949). Thus, it has been shown the the mortality rate for irradiated normal and adrenalectomized mice maintained on a salt diet is the same, even though the mean survival time for the intact animals is somewhat longer (Straube, et al., 1949). Indeed, this evidence suggests that the adrenal glands play a prominent role in the response of animals to irradiation, but probably not one of primary importance in determining whether or not the irradiated animals survive the acute stage of their illness.

I. Liver

There was no microscopic evidence of liver damage in Case 1. In Case 3, however, occassional dead liver cells were seen, and there was edema of the cords. The changes in the latter case are compatible with the clinical evidence of frank liver insufficiency. Although the liver parnechyma is said to be highly radioresistant, evidence of liver damage has been described in animals which have received large doses of radiation in the region of the abdomen (Pohle and Bunting, 1932; Warren, 1942). It has been stated recently that the histological changes seen in the liver in animals subjected to lethal doses of total-body radiation (not exceeding 1200 roentgens or its equivalent) are probably secondary in nature (Bloom, 1948). In Case 3, however, the heavy irradiation of the abdomen may have resulted in direct damage to this organ in addition to the indirect or secondary reaction of the liver to the injury of the body as a whole.

J. Kidneys

Aside from the infarct in Case 1, the only kidney lesion of significance was the moderate degenerative change noted in cells high in the tubular system of Case 3. This lesion, together with the extrarenal shunt (Trueta, et al., 1947), was probably responsible for the terminal anuria and nitrogen retention of this patient. In view of the widespread tissue breakdown, one might wonder if the kidney lesion described in this patient is similar in nature to that seen in the crush syndrome. Actually, the tubular damage in these cases is quite different from that seen in traumatic shock. The kidney changes in the latter syndrome usually occur lower in the tubules, are focal in nature, and are usually associated with precipitation of blood pigments within the tubules (Mallory, 1947).

K. Summary

Certain generalizations about the histopathological response of irradiated tissues were made, and the unusual features of the pathology of individual organ systems were discussed. The two types of cell death--that due to apparent acceleration of the usual type of cell death, and that which occurs during mitosis--were related to the experimental cytogenetic observations mentioned earlier (Section IV). The apparent relationship of the mast cells to the anticoagulant in the circulating blood was discussed. The most unusual lesion in the skin was the metaplasia of the duct cells of the sweat glands. There was an apparent difference in sensitivity of the stem cells of the lymphoid and hematopoietic tissues in these cases, and hyperplasia of the reticulum cells, a conspicuous finding in the Japanese, was absent. The possible causes of the pericarditis of Case 1 were considered, but no conclusion as to the biologic factor was reached. The kidney response was comparted to that observed in cases of the crush syndrome and found to be different in appearance.

X. RECONSIDERATION OF THE CALCULATED RADIATION DOSES IN TERMS OF THE OBSERVED BIOLOGICAL RESPONSE OF THE PATIENTS

The discussion of the radiation doses in Section II was intentionally limited, in so far as was possible, to the physical problem, and consideration of biological variables was kept to a minimum. Now that the clinical response of the patient has been presented, it is desirable to review the problem of radiation doses emphasizing the biological aspects. Therefore, the reliability of the dose computations will be re-assessed; those characteristics of the radiation which may be expected to influence the clinical response will be re-evaluated; and finally the quantitative aspects of specific tissue reactions will be compared with those produced by known amounts of x-rays or gamma rays. Such a correlation of physical and biological data is of utmost importance because of the paucity of accurate information about radiation doses in humans with acute radiation injury.

A. Reliability of the Radiation Dose Calculations

The uncertainties encountered in the computation of radiation doses in these patients were stressed in Section II (Vol. I). It seemed appropriate to re-examine the postulates on which the calculations were based. In this way, the reliability of the doses can be assessed and an idea of the direction of possible error may be obtained in some instances.

1. Doses in Terms of Energy Units

Consider for the moment the radiation dose in terms of energy dissipated in the body (Table I, Section II, Vol. I). It has been pointed out that the validity of these computations rests on the basic assumption that the incident neutron flux can be calculated from the specific activity of the serum sodium. The introduction of biological variables in the form of possible differential handling by the body of radiosodium poses a problem not implicit in calculations based on the physical interaction between neutrons and inert sodium. However, the simultaneous in vivo production of radiosodium by neutrons in all body sodium compartments, and the known ability of radiosodium to reach a prompt equilibrium with body sodium argue against errors due to biological factors. This view is substantiated by the constancy of the sodium specific activity which provides, in itself, strong experimental evidence against selective handling of the radioisotope by the body. Taking these facts into consideration, the use of sodium specific activity for the determination of neutron flux appears to be sound on both theoretical and experimental grounds.

Assuming that the basis used for neutron dose calculations is valid, the major sources of error in the dose calculations are the lack of good experimental data about incident neutron energy and the improper interpretation of their nuclear reactions. In considering the first of these sources of error, it should be pointed out that the arbitrary values for average neutron energy were chosen so that the errors in dose are likely to be high rather than low. This means, therefore, that the total energy dissipated in the body may be less than that indicated in Table I. Corrections for the use of unduly high neutron energies would result in an increase in the surface doses even though the total-body doses would be diminished. The second of these possible errors, that due to misinterpretation of nuclear reactions, deals with unknown factors which must await further experimentation.

In evaluating the fraction of the radiation dose due to gamma rays, it should be realized that considerable experimentation was undertaken to determine the self-dosage of the body by the capture gamma rays (column 2 in Table I). A mock-up of the body containing radiosodium was utilized in these studies and the reciprocity theorem was employed (Hoffman, 1947). It seems reasonable to accept with confidence these doses and the prompt primary gamma doses as being in the proper range. In contrast to these gamma-ray doses, the delayed gamma-ray dose (column 4, Table I) depends

upon an estimate of how long the operator remained in a region of known gamma ray intensity. It is conceivable that this dose estimate, particularly in Case 1, may not be accurate.

2. Doses in Terms of X-Ray Equivalent

The possible errors encountered in converting energy dissipated in the body into x-ray equivalent doses has been emphasized so strongly that little need be added here. It should be pointed out again that the over-all R.B.E. used for the neutrons in these calculations may be low for selected effects in specific tissues. Thus, the effective dose in such tissues as the gonads and lens, organs known to be highly sensitive to repeated neutron exposure, might be higher for an acute exposure than that indicated in Table II. The difference between the actual depth dose curves for neutrons of this energy and 80-kv x-rays should also be stressed again. Tissue ionization caused by irradiation with neutrons of this energy does not fall off so rapidly below the skin surface as does that produced by soft x-rays. Thus, for a given amount of energy dissipated in the body, the skin dose, in the case of the neutrons, would be less than that for the x-rays.

B. Reconsideration of the Characteristics of the Radiation Mixture and its Biological Implications

Now that the validity of the computed doses has been discussed, it is fitting to reconsider the unusual characteristics of the radiations which might influence the clinical response of the patients. Although much of this information has been stated before, the biological implications of these radiation characteristics will be reiterated and summarized.

1. Uneven Distribution of the Radiation in Cases 1, 3, and 4

It has been pointed out repeatedly that the doses quoted in Table I represent the average dose of radiation distributed over the entire body of the individual. In these cases where the incident radiation was non-uniform, the value for the average dose cannot be taken literally. Thus, the abdomen of Cases 1 and 3, and the head and left arm of Case 4 received a greater amount of radiation than that designated by the average dose. The heavy irradiation of these parts of the body must be kept in mind when the reactions of the abdominal skin, the intestines, and the superficially placed hematopoietic tissues of Cases 1 and 3 are studied and when the epilation of Case 4 is considered.

2. The Depth Dose of the Radiation

The limited powers of penetration of moderately fast neutrons has been stressed and the difference in the shape of the depth dose curves of neutrons of this energy and 80-kv x-rays has been pointed out. The distribution of the radiation dose in the body is important in determining the response of the individual. Preliminary reports of current investigation indicate that, in swine, the same dose of 200-kv x-rays is more injurious if given half to one side of the body and half to the opposite side, than if the entire dose is administered to one side (J.L. Tullis, personal communication). In the case of the more heavily irradiated survivors of these radiation accidents, the superficial nature of the body dose undoubtedly accounts for the fact that the patients did not show more serious constitutional reactions. This is analogous to animal experiments which show that protection of part of the hematopoietic tissue diminishes the damaging effects of ionizing radiation on the formed elements of the blood (Jacobson, et al., 1949-b).

It should be emphasized again that the superficial radiation dose due to beta rays and secondary x-rays was not taken into consideration except in the case of the hands. Such radiations may also have contributed to the damage of the exposed skin, e.g., of the face, particularly in the patients nearest the assembly. Furthermore,

482 86

the dose calculations do not take into account scattered slow neutrons of thermal energy. Slow neutrons would have contributed to the induced radioactivity in the body tissues but would produce maximum tissue ionization only 0.3 cm below the surface (Capron, Faes and Tavenier, 1949). The unknown amounts of such soft radiations, i.e., beta rays, soft x-rays and thermal neutrons would have enhanced the skin effects with relatively less influence on the systemic response of the patients.

The sharp localization of the epilation of Cases 4 and 5 and the effective shielding of the lens of the right eye of Case 4 by the bridge of the nose show the soft quality of much of the incident radiation.

3. Dose Rate of the Incident Radiation

It is well known that the biologic effect of radiation depends, to some extent, upon the dose rate. For the range of dose rates often used in biological experimentation, it has been shown that decreasing the dose rate lessens the biologic effect. Perhaps it is not valid, then, to compare the clinical reactions produced by radiation delivered at a rate of thousands of roentgen equivalents per minute with the response of patients and experimental animals irradiated by x-ray tubes, the usual output of which is about 50 roentgens per minute.

A recent symposium reviews the literature concerning the effect of dose rate on biological reactions (Gray, 1944; Ellis, 1944). It considers the results of irradiating various biological materials at vastly different dose rates ranging from very low radiation intensities to 40,000,000 roentgens per minute. The data indicates that the quantitative biological effects of radiation are dependent on dose rate at low intensities, but not at high intensities. The rate above which the biological effects become dose-rate-independent is 5-10 roentgens per minute for most animal reactions. In contrast, the reactions of some plants (algae) show dose-rate-dependence up to intensities of 1000 to 2000 roentgens per minute. Although it has been reported that the response of human skin is dose-rate-dependent at intensities up to 500 roentgens per minute, this has not been established by recent investigation (Ellis, 1944; Schottelndreyer, 1949).

The evidence just reviewed suggests, then, that the very high dose rate responsible for the injury of the present cases should not be expected to be inordinately effective in influencing clinical responses of these patients.

4. Specific Effect of Neutron Irradiation

The variability in the biological effectiveness of neutrons as compared with other radiations was mentioned in Section IV. Studies of the response of skin to neutron and x-ray irradiation provide an example of the inconsistancy of the R.B.E. for different effects in the same tissue (Stone, 1948). Thus, although one rep of neutrons delivered in a single dose is equivalent to 2.5 rep of x-rays in producing erythema, it is equal in effectiveness to 4 reps of x-rays in causing epidermolysis and late skin changes. An example of the variation of the R.B.E. for neutrons and x-rays in different tissues is found in an experiment in which the effects of localized acute irradiation of rabbit testes was studied (Lampe and Hodge, 1943). When compared to x-rays, fast neutrons are relatively more damaging to the germinal epithelium than to skin. This is shown by the fact that sterilization can be produced by neutron irradiation in the absence of permanent injury to the overlying skin, whereas, in the case of x-rays similarly applied, a comparable testicular effect can be achieved only by doses which cause lasting skin damage.

Besides the variations just mentioned for acute irradiation, it has been found that the R.B.E. of neutrons is dependent on the dose rate. When the lethal effect in mice of neutrons and x-rays are investigated at different radiation intensities, it is found that the effectiveness of neutrons as compared to x-rays or gamma rays is much greater for chronic than for acute exposures (Henshaw, Riley and Stapelton, 1947; Mitchell, 1947; Evans, 1949). Similarly, the selective damaging action of neutrons

in specific tissues is exaggerated when exposures are protracted. Thus, chronic neutron irradiation of mice causes more damage to the gonads and lens than to other tissues (Evans, 1948). No studies have been done on the R.B.E. of neutrons and x-rays or gamma rays at high intensities. If the dose rate data just mentioned is extrapolated to the present situation, one would expect the R.B.E. to be relatively low.

This discussion of the specific effect of neutrons emphasized the difficulties encountered in choosing a single value for the R.B.E. for use in neutron dose calculation. The evidence just presented, however, suggests the best approximation of the R.B.E. for the over-all response of the individual can be made by using a relatively low value. Since the range of the R.B.E. in acute biological responses is from 4 to 10, the factor of 5 used in the present computations does not seem unreasonable.

C. Consideration of the Quantitative Aspects of Selected Tissue Response

It is appropriate to conclude this section by considering specific tissue reactions; also, the dose estimates in the present cases will be compared with the x-ray doses known to produce comparable tissue responses. Except for the estimates of lethal doses, most of the clinical data presented below was obtained from studies using local rather than total-body irradiation.

1. Lethal Doses of Total-Body Radiation in the Human

The median lethal dose of radiation is not known with accuracy for humans, but it may be assumed to be about 400 $\pm$ 100 roentgens of gamma rays.* The mixed x-ray equivalent doses listed in Table II (Section II, Vol. I) obviously cannot be compared with this value. If one ignores the important consideration of the depth distribution of tissue ionization and converts megagram-roentgen dose into its gamma ray equivalents (using an R.B.E. of 5), the radiation doses turn out to be 300 roentgens for Case 1, 830 roentgens for Case 3, and 112 roentgens for Case 4. Although such an interpretation of the data is not strictly acceptable, the computation suggests that, in terms of energy loss in the body, the present doses are of the same order of magnitude as that which has been estimated to be the lethal radiation dose in man.

2. Epilation

There is considerable individual variation, particularly with age, in the epilation reaction of the scalp of man. Therefore it is difficult to express the minimal epilation dose as a single value. However, the dose of soft x-rays recommended for temporary therapeutic epilation in the treatment of ringworm of the scalp is not less than 300 roentgens of soft unfiltered x-rays (MacKee and Cipollaro, 1946). Recently it has been stated that complete temporary epilation is not achieved except by administering 75-kv unfiltered x-rays in such a way that the dose in a given field varies between 375 and 500 roentgens (Pendergrass and Mahoney, 1948). Thus, it appears that the calculated dose in Case 4 approximates the x-ray dose known to produce comparable clinical effects. The calculated doses in Case 5 may be slightly lower than the x-ray dose which would produce the same degree of epilation.

3. Sterilization

The dose of radiation which causes temporary and permanent sterilization in the male is not known with certainty (Dunlap, 1947). However, it has been stated that 250 roentgens of x-rays (energy undesignated) will cause temporary, but not permanent sterility (Ellinger, 1941; Glucksmann, 1947). The response of the germinal tissues noted in Cases 4 and 5, then, is not unexpected in view of the calculated radiation doses. The fact that irreversible damage did not occur suggests that, unlike chronic

*This figure is based on the opinion of a group of twenty-four experts in radiology and radiobiology (R. R. Newell, personal communication).

482 88

exposure, acute neutron irradiation in these cases was not inordinately effective in damaging germinal tissue. As has been pointed out, however, animal experiments suggest that neutrons delivered as a single dose are more damaging to germinal epithelium than are doses of x-rays which produce comparable skin effects (Lampe and Hodge, 1943). Thus, it may be true that the effective dose of the gonads in the patients under discussion was somewhat greater than that indicated in the dosage tables.

It should also be pointed out that the partial shielding of Case 4 may have resulted in the gonadal dose being lower than the average body dose.

4. Delayed Damage to the Lens of the Eye

The dose of x-rays required to produce cataracts in animals varies directly with age (Warren, 1943). The minimal cataractogenic dose of x-rays in adult humans is not known, but it has been reported that 875 roentgens of 130-kv x-rays and 1000 roentgens of gamma rays cause cataracts in patients treated for cancer of the eyelid (Hunt, 1947). The radiation dose of the ten Japanese who developed radiation cataracts is not known exactly, but an idea of its order of magnitude is gained from the fact that all were epilated (Cogan, Martin and Kimura, 1949). The cataractogenic properties of acute exposures to neutrons has not been studied, but it seems clear that repeated neutron exposure is more damaging to the lens than to most other tissues. The development of the cataract in the left eye of Case 4 is not unexpected in view of the fact that his head probably received more radiation than that indicated by the average body dose.

5. Initial Reaction or Radiation Sickness

This response is so variable that it is not worth while considering seriously as an index of radiation dosage. Nevertheless, it has been shown that 100 roentgens of 200-kv x-rays can be delivered to the entire body of a patient without causing symptoms of any sort (Cantril, et al., 1947). If the important factor of the distribution of tissue ionization in the body is ignored again, and the megagram-roentgen dose of Case 6 is converted into its gamma ray equivalent, a value of 92.4 roentgens is obtained. As has been pointed out the significance of such a dose conversion is of limited value, but it does suggest that total-body radiation approximating 100 roentgens or below (as was received by Cases 6 and 7) need not cause symptoms of radiation sickness.

6. Bone Marrow Damage

Experimental studies in animals and clinical experience in man, indicate that the blood forming tissues are exceedingly radiosensitive. The lymphoid tissue is considered to be of the same order of radiosensitivity as the erythroblastic tissue of the bone marrow (Bloom and Bloom, 1947). Histologically, the damage in the marrow and lymphoid tissues produced by whole-body irradiation with either gamma rays or fast neutrons in equivalent doses is identical (Bloom, 1947). For this reason, the marked damage of the deeply placed vertebral marrow of Case 3, which is out of proportion to the lymphatic tissue reaction, represents a distinctly unexpected finding. This observation cannot be explained on the basis of enhanced scattering of neutrons by the relatively heavy bone tissue with the resultant increased energy dissipation in the bone and adjacent tissues such as is known to occur with soft x-rays (Spiers, 1946). The well known scattering properties of calcium and phosphorus for neutrons suggest that increased energy absorption does not occur in bone. Nor can severe marrow effects be explained by induced radioactivity in bone phosphorus which would have accounted for only a fraction of the total ionization in the bony tissue.

Hence, one cannot interpret the marrow damage on the basis of any known phenomenon occurring as a result of the interaction of neutrons and bone. The

extreme damage, even to the radioresistant reticulum cells of the deep-seated vertebral marrow of Case 3, is difficult to reconcile with the present concept of depth dose of the radiations as presented in Section II (Vol. I).

D. Summary

After considering the tenets on which the radiation dose computations are based, it is concluded that the use of the specific activity of the serum sodium is justifiable. The values assumed for the average energy of the incident neutrons are such that errors in the neutron dose are believed to be on the high side. Reconsideration of the characteristics of the incident radiations suggest that in so far as the quantitative aspects of the biological reactions are concerned, unusual clinical responses are not to be expected in the present cases. When specific tissue reactions of the present cases are considered, it is noted that they are in agreement with the x-ray doses known to produce a comparable response, with one exception -- the severe damage of the vertebral bone marrow of Case 3. The degree of epilation in Case 5 is also probably greater than that which could be expected for the calculated dose.

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