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PATHOLOGIC CHANGES OBSERVED IN COBALT-60 UPPER-BODY IRRADIATED SHEEP

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AIR FORCE WEAPONS LABORATORY
Air Force Systems Command
Kirtland Air Force Base
New Mexico

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PATHOLOGIC CHANGES OBSERVED IN COBALT-60 UPPER-BODY IRRADIATED SHEEP

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FOREWORD

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Inclusive dates of research were September 1967 through February 1969. The report was submitted on 15 July 1969 by the Air Force Weapons Laboratory Project Officer, Captain Thomas C. DeFeo, USAF MC (WLBR).

The authors express their sincere appreciation to the staff of the Radiobiology Laboratory, Biophysics Division, Air Force Weapons Laboratory.

Research was conducted according to the principles enunciated in the "Guide for Laboratory Animal Facilities and Care," prepared by the National Academy of Sciences-National Research Council.

This technical report has been reviewed and is approved.

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ABSTRACT

(Distribution Limitation Statement No. 2)

Pathologic examination was made of 11 sheep after spontaneous death caused by upper-body midlethal dose range exposure to Cobalt-60 gamma radiation. The examination revealed that death was caused by pulmonary insufficiency arising from radiation pneumonitis with varying degrees of alveolar septal fibrosis. Gross and microscopic pathologic changes were evaluated in terms of the pathogenetic sequence of events culminating in death from respiratory insufficiency. Additional studies giving more detailed data will be published as a result of this experiment.

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SECTION I

INTRODUCTION

This report presents the pathologic findings in sheep after spontaneous death following acute midlethal dose range exposure of the upper (cephlad) half of the body to Cobalt-60 gamma radiation. This is only part of a multifaceted lethality study designed to evaluate the effects of partial body shielding following exposure of sheep to Co-60 gamma radiation. Other parameters, including death frequency distribution, $LD_{50/60}$, hematologic and biochemical values, will be presented in separate technical reports, together with greater detail of the exposure setup and dosimetry.

SECTION II

PROCEDURES

1. MATERIALS AND METHODS

Adult female domestic sheep of the Columbia-Rambouillet cross were used in this study. There were eight dose groups (1600, 1800, 2000, 2200, 2300, 2400, 2500, and 2600 R midline air dose) containing ten sheep each, plus a control group.

The Cobalt-60 gamma ray source used in this study is housed in the Large Animal Irradiation Facility (LAIF); the design and operating characteristics of this facility are described in detail in Air Force Weapons Laboratory Technical Report 68-16.

Sheep in each dose group were placed radially around the cobalt source, receiving the entire gamma dose to the right side of the upper half of the body. The lower half of the body, below the level of the xiphoid process, was shielded by 2-1/4 inches of lead (95 percent dose reduction factor). Details of the dosimetry, exposure configuration, and animal husbandry practices before and after exposure will be reported in other AFWL technical reports, together with the lethality, biochemical, and hematologic data. All doses referred to in this report are calculated midline air doses, based on the effective half-life of the Cobalt-60 source employed. The dose rate for all groups was 11 R per minute.

Following exposure, all sheep were observed for a period of 60 days. The 11 sheep that died spontaneously during this period were autopsied. Representative samples from all organs and tissues were fixed in formalin, and embedded in paraffin blocks. Sections approximately 6 micra in thickness were prepared from the paraffin blocks, placed on glass slides, and stained with hematoxylin and eosin. In addition, duplicate sections of lung were stained with Masson's trichrome stain for evaluation of fibrous tissue change in the lungs.

2. RESULTS

The gross and microscopic pathologic alterations noted represent the end-stages of a disease process initiated by delivery of Co-60 gamma radiation to the portion of the body above the level of the xiphoid process. Distribution

of pathologic changes, irrespective of dose group, is shown in table I.

Table I

PATHOLOGIC CHANGES NOTED IN 60-DAY NONSURVIVING SHEEP RECEIVING UPPER-BODY COBALT-60 GAMMA RADIATION

Lesion	Number of sheep with lesion
Radiodermatitis	11
Central Nervous System Lesions	0
Cardiac Dilatation	11
Radiation Pneumonitis	11
Fatty Metamorphosis of the Liver, Slight	5
Acute Gastritis, Focal	1
Hypoplasia of Bone Marrow, Upper Half of Body	11
Hyperplasis of Bone Marrow, Lower Half of Body	11
Hemosiderosis of Lymph Nodes and Spleen, Mild	11
Evidence of Systemic Bacteremia	0

Epilation of the upper body was prominent in all decedents; the gross and microscopic features of radiation dematitis were present.

There were no gross or microscopic lesions noted in the central nervous system.

The gastrointestinal tract was essentially unremarkable, except for one sheep with a nonspecific focal acute gastritis and five sheep with slight fatty metamorphosis of the liver; probably because these sheep did not eat during the last days of life.

Panmyocardial cardiac dilatation was the only abnormality noted in the hearts of the decedents. This was felt to have developed secondary to the pulmonary lesions (cor pulmonale) described below.

Hypoplasia of upper body foci of bone marrow was offset by hyperplasia of lower body bone marrow foci. Macrophages in the lymph nodes and the spleen were increased in number and contained increased amounts of hemosiderin pigment.

There was no evidence of systemic bacteremia.

The most significant pathologic changes were found in the lungs of each decedent (table II). In each case, the hallmarks of the early phase of radiation pneumonitis were present, consisting of varying degrees of focal hemorrhage, vascular congestion and edema (in the absence of inflammation), hypertrophied and distorted alveolar lining cells, and deposition of a fibrinous exudate within alveolae (figures 1 through 4). In some animals, particularly the four decedents receiving the largest dose of radiation (table II), a more advanced stage of radiation pneumonitis was superimposed with significant fibrosis of alveolar septae associated with perivascular fibrosis (figures 5 and 6).

Bacterial colonies were conspicuously absent in the pulmonary lesions described above. Grossly, in all cases, the lungs were abnormally heavy, owing to the presence of edema fluid. They also exhibited significant decreases in crepitance and had a rubbery consistency. The four decedents that received the highest doses of radiation (table II) exhibited striking additional gross pulmonary findings in the posterior portions of both lower lobes. These findings consisted of marked increase of pulmonary tissue consistency involving the posterior portions of both lower lobes. There was a striking contrast between the greyish-white pallor of these areas when compared with the reddish congested areas of lung, anteriorly. The pulmonary parenchyma in the affected areas was virtually bloodless, had a hyalin-like appearance and cut with increased resistance. The gross pulmonary findings mentioned above appeared slightly more severe on the right side than on the left side. Microscopically, these areas reflected extensive alveolar fibrosis superimposed on a typical picture of radiation pneumonitis as noted above (figures 5 and 6). An important point that should be emphasized is that in every case the most severe pulmonary lesions were found in the posterior portions of the lower lobes. Although the number of decedents per dose group was small, one does get the impression that the sheep receiving the higher doses (2500 and 2600 R) exhibited greater degrees of alveolar and perivascular fibrosis superimposed on existing radiation pneumonitis (table II). In addition, the degree of intra-alveolar fibrinous exudate in the lungs of these high-dose-group decedents with increased pulmonary fibrosis appears to be somewhat less than that in the lower dose group decedents. Because of the limited number of decedents, correlations between dose, time of death, and pathologic changes are admittedly difficult to make.

Table II

PULMONARY PATHOLOGIC FINDINGS OF COBALT-60, UPPER-BODY IRRADIATED 60-DAY NONSURVIVORS

Dose group (R)	Animal No.	Time of death (days post exposure)	*Specific gross lung lesions	Congestion and edema (without inflammation)	Fibrinous exudate within alveolar spaces	Hypertrophied & distorted alveolar lining cells	Alveolar wall fibrosis	Perivascular fibrosis
2600	1021	33	+	+	+	+	+ + +	+ + +
2600	1082	44	+	+	+	+ + +	+ +	+ +
2500	1887	40	+	+	+	+ +	+ + +	+ +
2500	1689	42	+	+	+ +	+ +	+ +	+
2400	931	26	-	+	+	+ +	+	-
2400	922	28	-	+	+	+	+	-
2400	1313	32	-	+	+ +	+	+	-
2400	1320	33	-	+	+	+ + +	+	-
2400	1133	52	-	+	+ + +	+	+ +	+
2300	1836	24	-	+	+ + +	+	+	-
2200		No deaths						
2000	1131	47	-	+	+ + +	+ + +	+ +	+
1800		No deaths						
1600		No deaths						

*Increased pulmonary tissue consistency with decreased crepitus, palor, and hyalin-like appearance of pulmonary parenchyma most prominent in the posterior portions of both lower lobes.

Figure 1. Alveolar Septal Thickening Secondary to Vascular Congestion and Edema, H & E, X 125 (Sheep No. 931)

Figure 2. Hypertrophied and Distorted Alveolar Lining Cells; Alveolar Septal Thickening Secondary to Vascular Congestion and Edema, H & E, X 500 (Sheep No. 1082)

Figure 3. Fibrinous Exudate within Alveolar Spaces
Masson Trichrome, X 125 (Sheep No. 1133)

Figure 4. High-Power View of Intra-Alveolar Fibrinous
Exudate, Masson Trichrome,
X 500 (Sheep No. 1133)

Figure 5. Alveolar Septal Fibrosis Associated with Intra-Alveolar Fibrin and Distorted Alveolar Lining Cells, H & E, X 500 (Sheep No. 1131)

Figure 6. Advanced Pulmonary Fibrosis with Associated Perivascular and Peribronchiolar Fibrosis, Masson Trichrome, X 125 (Sheep No. 1021)

SECTION III

DISCUSSION

During the last 30 years, many good articles have been published concerning the clinical, pathologic, and pathophysiologic changes in the pathogenesis of radiation pneumonitis, in both man as well as a variety of experimental mammalian species (Refs. 1 through 12).

The changes described in the respiratory system of the nonsurviving sheep exposed to various doses (2000 to 2600 R) of upper-body Co-60 gamma radiation in this experiment reflect the typical response (at this general dose level) of mammalian pulmonary tissue to radiation injury culminating in death from respiratory insufficiency.

The pathological changes leading to death as described above for sheep exposed under the conditions of this experiment is clearly different from that of sheep exposed to whole-body midlethal dose range (300 to 415 R) Co-60 gamma radiation as reported in a previous publication (Ref. 13).

Unfortunately, because of the nature of this study and the small numbers of fatalities, correlation of dose rate, total dose, and time of death post-exposure with pulmonary pathologic changes was not possible with any degree of statistical significance. Others have, however, related these parameters as well as additional ones to the degree of radiation injury (Refs. 8, 10, 12).

An interesting occurrence observed in this experiment was the presence of the severest pulmonary lesion in the posterior (dorsal) portions of both lower lobes of sheep receiving right unilateral Cobalt-60 irradiation to the upper half of the body. With the knowledge of the role of oxygen in potentiation of the effects of radiation on tissue, it has been suggested that uneven alveolar ventilation during exposure to ionizing radiation may account for the fact that some areas of pulmonary parenchyma are less severely injured than others (Refs. 11 and 14). From the overwhelming pulmonary pathologic changes in the absence of other more significant lesions in other organ systems, death in each case must be attributed to the pulmonary insufficiency arising from radiation pneumonitis with varying degrees of alveolar septal fibrosis.

Briefly, current concepts of the pathogenesis of radiation pneumonitis relate to initial injury of cells comprising the alveolar septae, namely, alveolar lining cells, aveolar septal cells, and capillary endothelial cells. As has been pointed out, suppression of mitotic division by irradiation results in a gradually progressive cellular depletion atrophy of the alveolar septum (Ref. 9). This is due to an upset of the steady-state system of alveolar lining cells because cells fail to renew after radiation-induced cellular depletion (Ref. 15). Such an occurrence undoubtedly weakens the delicate aveolar septum, and together with injury of the capillary endothelium which increases vascular permeability, allows exudation of fibrin-rich edema fluid into the alveolar spaces (Refs. 10 and 16). Normally, when fibrin is deposited within the alveolus, an insoluble enzyme--lung plasminogen activator--acts locally, converting circulating plasminogen to plasmin, thus effecting local fibrinolysis, with clearing of fibrin from the alveolar spaces (Refs. 6 and 17). Abnormalities of the fibrinolytic system of the lung have been described in hyalin membrane disease of the newborn (Ref. 17) as well as in experimentally induced radiation pneumonitis where decreased lung plasminogen activator was demonstrated to be associated with persistence of alveolar fibrin deposition and later hyalin membrane formation resulting from polymerization of intra-alveolar fibrin deposits (Ref. 6). The surviving alveolar epithelium rapidly grows as an attenuated layer to cover the surface of the contracting mass of fibrin so that the latter becomes interstitial in position (Ref. 18). This occurs in the absence of resolution of the intra-alveolar fibrinous exudate by the actions of fibrinolysins (plasmin) or digestion by tryptic enzymes released from degenerating polymorphonuclear leukocytes (leukocyte production is considerably depressed following irradiation). This process of organization and fibrous repair is unaccompanied by capillary proliferation (Ref. 18). Thus, the ultimate outcome if resolution of intra-alveolar fibrinous exudate fails to occur is fibrosis of an already compromised alveolar wall, across which critical gas exchange normally occurs.

Spencer (Ref. 18) has pointed out that the lung as well as other organs may incur damage in a number of ways and that these differing injury processes may result in end-stage changes that are quite similar (figure 7). In the lung, interstitial fibrosis (or chronic interstitial pneumonia according to Spencer's terminology, Ref. 18) may be caused by microbial, chemical, or physical agents, circulating immune complexes, circulating metabolites, and possibly other unrecognized initiating agents or events (Ref. 18). Ionizing

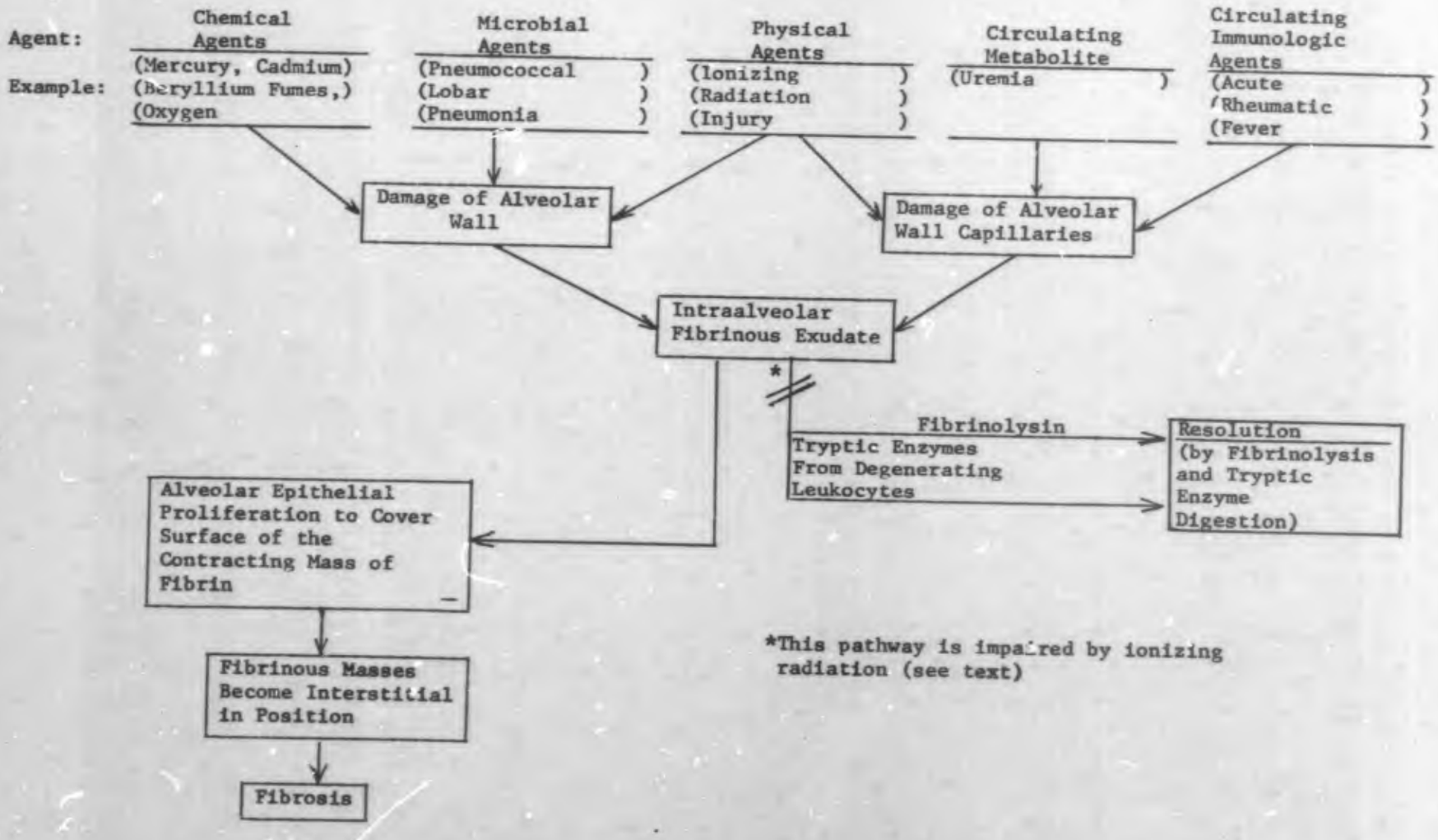

Figure 7. Pathogenesis of Chronic Interstitial Pneumonia (Interstitial Fibrosis of the Lung) Arising from Multiple Etiologic Agents Including Ionizing Radiation

radiation can be considered a physical agent initiating a disease process that culminates in radiation pneumonitis (figure 7). Thus, we see that the lung with its specific structural composition has only a limited number of ways in which it can respond to a variety of injurious agents, including ionizing radiations.

SECTION IV

CONCLUSIONS

Pathologic examination of sheep after spontaneous death following midlethal dose range exposure to the upper-half of the body by Cobalt-60 gamma radiation reveals that death is caused by pulmonary insufficiency arising from radition pneumonitis with varying degrees of alveolar septal fibrosis. Generally, radiation pneumonitis with extensive intra-alveolar fibrinous exudate is more severe in the lower dose groups, while pulmonary fibrosis is more severe in the higher dose groups. This may be due to accelerated fibrosis of fibrinous masses as the dose increases. The role of the intra-alveolar fibrinous exudate in the pathogenetic sequence is found to be very important.

REFERENCES

1. Warren, S., and Gates, O., "Radiation Pneumonitis--Experimental and Pathologic Observations," Arch. Path. 30: 400 (1940).

2. Warren, S., and Spencer, J., "Radiation Reaction in the Lung," Am. J. Roentgenol. 43: 682 (1940).

3. Whitfield, A. G. W., Bond, W. H., and Arnott, W. M., "Radiation Reactions in the Lung," Quart. J. Med. 25: 67 (1956).

4. Stone, D. J., Schwartz, M. J., and Green, R. A., "Fatal Pulmonary Insufficiency Due to Radiation Effect Upon the Lung," Am. J. Med. 21: 211 (1956).

5. Jennings, F. L., and Arden, A., "Development of Experimental Radiation Pneumonitis," Arch. Path. 71: 437 (1961).

6. Fleming, W. H., Szakacs, J. E., and King, E. R., "The Effect of Gamma Radiation on the Fibrinolytic System of Dog Lung and Its Modification by Certain Drugs: Relationship to Radiation Pneumonitis and Hyalin Membrane Formation in Lung," J. Nucl. Med. 3: 341 (1962).

7. Jennings, F. L., and Arden, A., "Development of Radiation Pneumonitis--Time and Dose Factors," Arch. Path. 74: 351 (1962).

8. Smith, J. C., "Radiation Pneumonitis--A Review," Am. Rev. Resp. Dis. 87: 647 (1963).

9. Smith, J. C., "Experimental Radiation Pneumonitis," Am. Rev. Resp. Dis. 87: 656 (1963).

10. Rubin, P. and Casarett, G. W., Clinical Radiation Pathology, Ch. 12, W. B. Saunders Company, Philadelphia, Pennsylvania (1968).

11. Goldenberg, V. E., Warren, S., Chute, R., and Besen, M., "Radiation Pneumonitis in Single and Parabiotic Rats," Lab. Invest. 18: 215 (1968).

12. Cooper, G., Jr., Guerrant, J. L., Harden, A. G., and Teates, D., "Some Consequences of Pulmonary Irradiation," Am. J. Roentgenol. 85: 865 (1961).

13. Mobley, T. S., and DeFeo, T. C., Cobalt-60 Gamma Irradiation of Sheep: Correlations of Sixty-Day Median Lethal Dose Studies with Selected Biochemical, Hematologic, and Pathologic Findings, AFWL-TR-68-83, Air Force Weapons Laboratory, Kirtland AFB, New Mexico (1968).

14. Leroy, E. P., Liebner, E. J., and Jensik, R. J., "The Ultrastructure of Canine Alveoli After Supravoltage Irradiation of the Thorax," Lab. Invest. 15: 1544 (1966).

15. Bertalanffy, F. D., and LeBland, C. P., "The Continuous Renewal of the Two Types of Alveolar Cells in the Lung of the Rat," Anat. Rec. 115: 515 (1953).

REFERENCES (cont'd)

16. Szirmai, E. (ed.), Nuclear Hematology, Academic Press, New York (1965). (Chapter 14 by Iossifides, I. A., and Geisler, P. H.: Radiation Effects on Blood Coagulation, Paragraph 3. Effects of X Rays on Vascular Permeability.)

17. Lieberman, J., "Clinical Syndromes Associated with Deficient Lung Fibrinolytic Activity: I. A New Concept of Hyaline Membrane Disease," New Eng. J. Med. 260: 619 (1959).

18. Liebow, A. A. (ed.), The Lung, The Williams and Wilkins Company, Baltimore, Maryland (1968). (Chapter 10: "Chronic Interstitial Pneumonia," by H. Spencer.)

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13. ABSTRACT

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