ISCHEMIA OF THE LUNG DUE TO IONIZING RADIATION: QUANTITATIVE STUDIES

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The response of the human lung to therapeutic levels of ionizing radiation is well established pathologically and corresponds to the pattern of radiation-induced injury and repair observed in other organs (*1,2*). In the acute stage there is hyperemia, bronchial epithelitis and depletion of lymphoid elements. Later there is a degenerative stage of parenchymal cellular damage or death. This stage is marked by fibrino-purulent pleuritis, swelling and sloughing of alveolar lining cells, and the appearance of fibrinoid material within alveoli. The latter may form a continuous hyaline membrane. The final, regenerative stage is characterized by metaplasia of bronchial epithelium and alveolar lining cells, connective tissue proliferation and obliterative pulmonary arteriolitis. Grossly, the lung exhibits loss of elasticity and volume. Recent studies with light and electron microscopy have shown striking changes in the pulmonary capillaries following large radiation doses (*3*). There is an early increase in capillary permeability, followed by sloughing of capillary endothelium and obliteration of the endovascular space by cells and acellular debris. Arterioles exhibit intimal thickening and proliferation of adventitial fibrous tissue. This is accompanied by interstitial parenchymal fibrosis.

The clinical counterparts of these events may emerge in the variable syndrome of so-called radiation pneumonitis and radiation fibrosis of the lung. Post-irradiation pulmonary fibrosis has been observed radiographically in 70% of long-term survivors with carcinoma of the breast (*4*); usually asymptomatic, it may be associated with disabling pulmonary insufficiency (*5*). Cor pulmonale is another severe, if rare, complication of late radiation fibrosis (*6*).

Several investigators have studied the functional changes caused by radiation-induced injury of the lung using classical methods of respiratory physiology in man and in the dog. There is general agreement that the maximum breathing capacity, residual lung volume, pulmonary compliance and diffusing capacity are reduced, with varying degrees of alveolocapillary block (*7,8*). However, several uncontrollable factors have often mitigated against the sensitivity of clinical data. These include the presence of intercurrent chronic pulmonary disease such as emphysema or bronchitis, persistence or progression of neoplasia and the difficulty of detecting regional dysfunction in measurements of total lung function.

Using serial pulmonary perfusion scans, we have shown that pulmonary blood flow is reduced regionally in the irradiated segment of the lung in patients who have undergone radiation therapy (*9*). These patients also exhibit pulmonary fibrosis, pleural thickening or both, but these radiographic findings are less extensive than the corresponding deficit in pulmonary flow.

In view of evidence that pulmonary ischemia may follow irradiation of the lung, an experiment was designed to measure its incidence and severity. Advantage was taken of the fact that the distribution of radioactive macroaggregates in the lungs is flow-dependent and that the partition ratios of the particles and of pulmonary arterial blood between the lungs are equivalent.

MATERIALS AND METHODS

Adult female albino Swiss mice underwent irradiation of the entire right lung using orthovoltage equipment. Factors were: 250 KVP, 15 mA, no added filtration and half-value layer—1.5 mm of copper. The dose rate was 178 R/min at the treatment distance of 60 cm.

The following steps were taken to insure uniform irradiation of the right lung. The projected surface area of the right lung in the coronal plane was measured in radiographs of representative animals of similar weight. This could be approximated by a right triangle of specific dimensions (Fig. 1). Using

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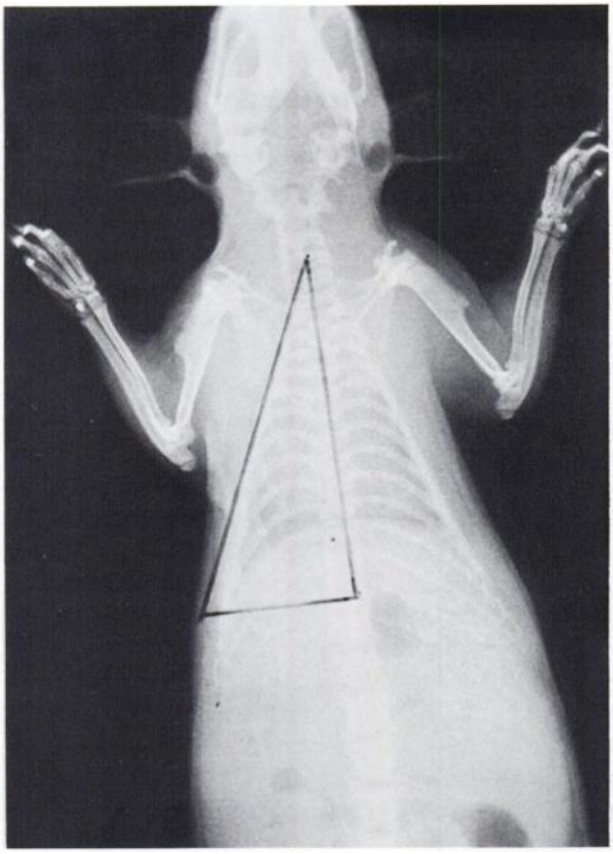

FIG. 1. Roentgenogram of thorax of mouse in prone position. Black triangle outlines region of thorax exposed to ionizing radiation and includes right lung.

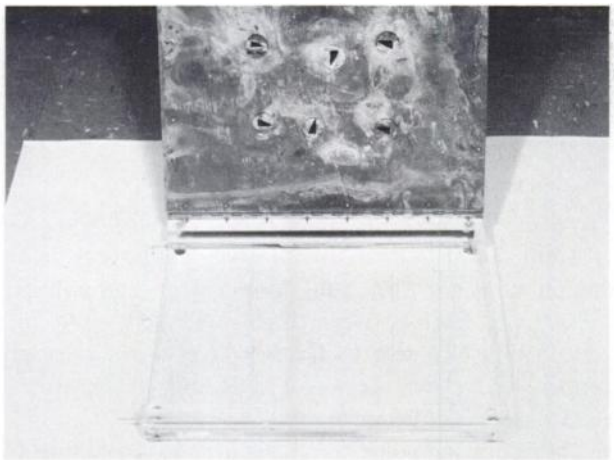

FIG. 2. Photograph of open irradiation jig. Hinged lead cover contains six triangular apertures through which incident radiation passes. Lucite plate in fixed part of jig contains marks which correspond to centers of adjacent apertures when lid is closed.

these dimensions, six identical triangles were cut in a sheet of lead ¼-in. thick which formed the top of the irradiation jig. These apertures (Fig. 2) were spaced so that six mice could be irradiated in the prone position at one time. Additional lead strips were attached at the medial and inferior margins of each aperture to reduce penumbra. Ionization chamber measurements established that the intact lead cover transmitted less than 1% of incident radiation. Further, radiation passing through each aperture was uniform in amount per unit time at each treatment locus. These points were marked on a removable Lucite plate.

Prior to irradiation, groups of mice were anesthetized with pentobarbital intraperitoneally. The mice were then taped prone on the marked positions on the Lucite plate. A nonscreen radiograph of the loaded jig was developed and inspected at once. When correctly positioned, the right hemithorax of each mouse was visible through the treatment aperture (Fig. 3). Initially, minor changes in position were often required to relocate the right hemithorax directly beneath an aperture. The irradiation jig was then placed in a predetermined position beneath the orthovoltage tube head and treatment given.

Five groups of 42 mice received single radiation doses of 100 R, 500 R, 1,000 R, 2,000 R and 4,000 R, respectively. After irradiation, mice were housed in separate cages according to radiation dose.

Subgroups of six mice were sacrificed at fixed intervals following irradiation. These intervals were 4 days, 16 days, and 1, 2, 4, 6 and 10 months. Immediately before sacrifice a suspension of ^{131}I-labeled macroaggregated human serum albumin containing 5–20 μCi in a volume of 0.1–0.2 ml was injected via tail vein. One minute later the mouse was decapitated. The lungs were removed en bloc, severed at the hilum, blotted and weighed separately. The whole lung was then placed in a counting vial,

TABLE 1. MEAN VALUES OF PULMONARY PERFUSION RATIO IN EXPERIMENTALLY IRRADIATED MICE

Interval irradiation to sacrifice	Radiation dose to lung (rads)				
	100	500	1,000	2,000	4,000
4 days	1.02	1.07	1.01	1.01	1.04
(s.d.)	(±0.16)	(±0.21)	(±0.24)	(±0.50)	(±0.20)
16 days	1.09	1.00	0.96	0.99	0.92
(s.d.)	(±0.15)	(±0.26)	(±0.30)	(±0.28)	(±0.12)
1 month	1.01	—	1.08	0.93	0.81
(s.d.)	(±0.05)	—	(±0.27)	(±0.12)	(±0.28)
2 months	1.04	0.96	0.82	0.71	0.64
(s.d.)	(±0.11)	(±0.17)	(±0.24)	(±0.16)	(±0.31)
4 months	1.09	0.88	0.81	0.84	0.57
(s.d.)	(±0.38)	(±0.20)	(±0.34)	(±0.12)	(±0.25)
6 months	0.79	0.91	0.70	0.69	0.58
(s.d.)	(±0.44)	(±0.16)	(±0.18)	(±0.09)	(±0.05)
10 months	0.73	0.74	0.88	—	0.45
(s.d.)	(±0.22)	(±0.36)	(±0.48)	—	(±0.50)

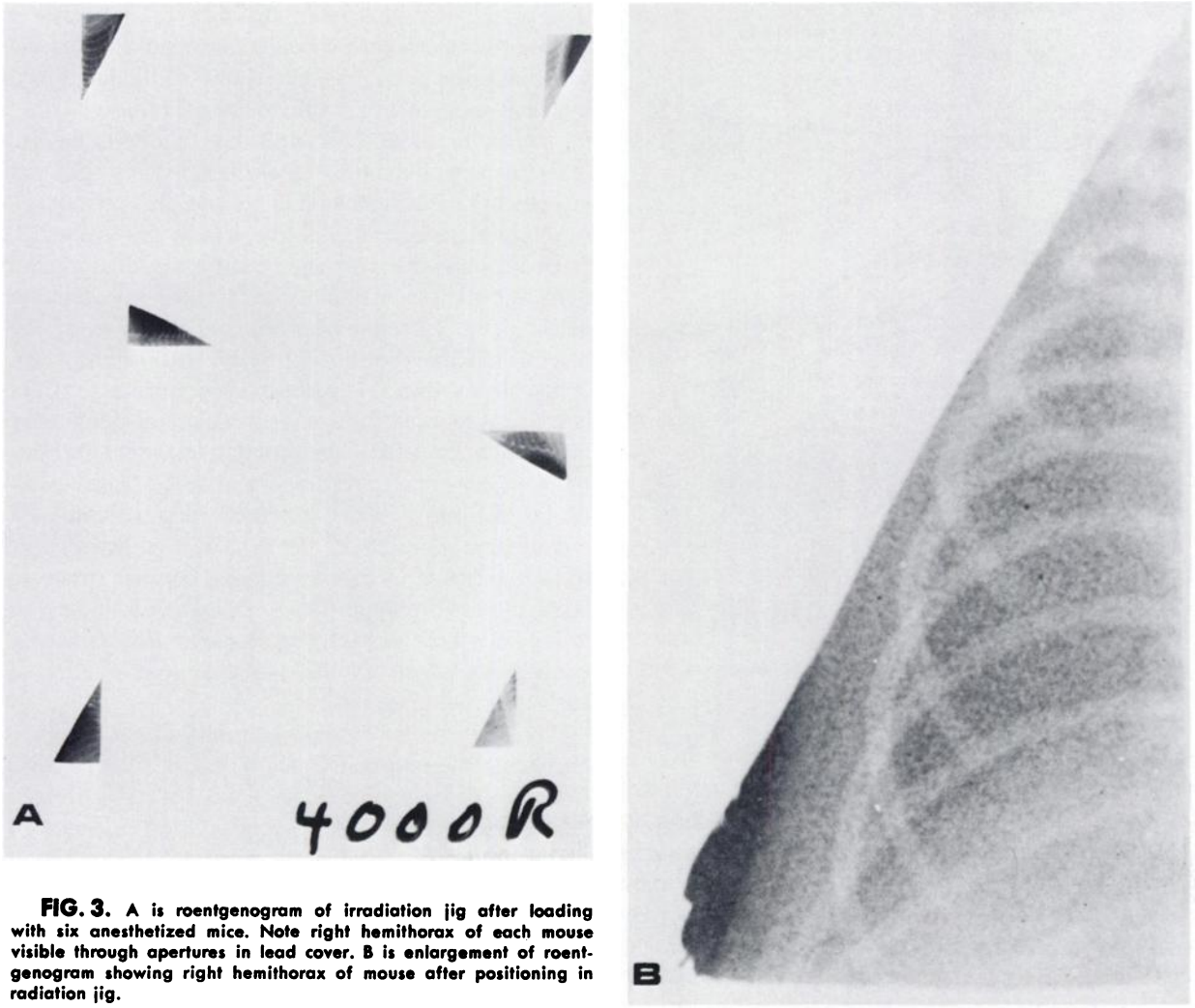

FIG. 3. A is roentgenogram of irradiation jig after loading with six anesthetized mice. Note right hemithorax of each mouse visible through apertures in lead cover. B is enlargement of roentgenogram showing right hemithorax of mouse after positioning in radiation jig.

and the radioactivity content was measured in a well scintillation counter with pulse-height analyzer.

Animals whose lungs exhibited gross consolidation or atelectasis were considered to have intercurrent pulmonary disease and were not included in the investigation. However, histological examination of the lungs was not performed.

For analysis of results, the number of counts per minute of each lung was divided by lung weight in milligrams, giving a value whose units were counts per minute per milligram of wet lung tissue. The ratio of these values for each pair of lungs, one of which was irradiated, was expressed as the pulmonary perfusion ratio (PPR). Thus,

$$\text{PPR} = \frac{\text{cpm/mg irradiated lung}}{\text{cpm/mg control lung}}.$$

RESULTS

The pulmonary perfusion ratio in the normal (nonirradiated) mouse was 0.99 ± 0.06 (s.d.). Values of PPR after irradiation are listed in Table 1 and reproduced graphically in Fig. 4. The data demonstrate reduced PPR values regardless of radiation dose in all groups sacrificed more than 1 month after treatment. Reduction in PPR was evident not only as a function of the irradiation-to-sacrifice time interval but also as a function of total radiation dose. Thus the greatest drop in PPR occurred in groups receiving the largest doses and having the longest survival until sacrifice.

Epilation of the thoracic fur was routinely observed in mice receiving doses of 2,000 and 4,000 R. Fine pleural adhesions were frequently present in these animals, and the irradiated lungs were stiffer and smaller than their normal counterparts. No case of massive pleural effusion was seen even in animals sacrificed at short intervals after large doses of radiation.

DISCUSSION

The validity of our experimental method of quantifying the intrapulmonary partition of right ven-

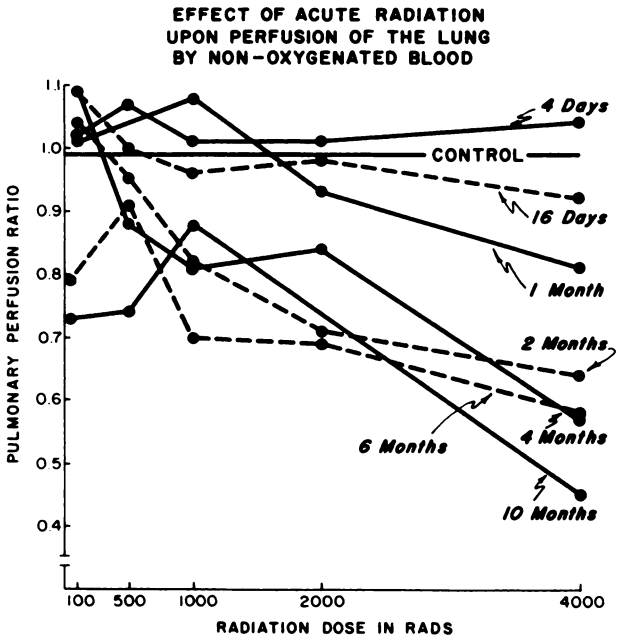

FIG. 4. Graphic presentation of values for pulmonary perfusion ratio in all experimental groups.

tricular output has been established by two investigations. Differential bronchospirometry in man has shown that the percent of oxygen uptake of one lung, which is a measure of the percent of pulmonary blood flow to that lung, correlates closely with the fraction of radioactive particles in that lung (*10*). It has also been shown that radioactive particles and labeled red cells undergo a nearly identical distribution in the lung (*11*).

Our results confirm the finding that radiation injury causes pulmonary ischemia. They indicate that the degree of ischemia is influenced both by the duration of survival following irradiation and by the magnitude of the radiation dose.

Our observations were made over a 10-month interval, which is about half the life span of the Swiss mouse. We did not observe evidence of a leveling-off or reversal of the intensity of ischemia. Therefore we conclude that the process is irreversible although further studies through an entire life span would be required to establish this point irrefutably.

We regard the consistency of the data as satisfactory for an experiment of this type. We consider the high standard deviations associated with certain PPR values to result from (1) the relatively small number of mice in each subgroup, (2) the problems associated with accurate weighing of wet tissue and (3) the possibility that unrecognized pulmonary disease may have been present in some animals.

Findings similar to ours have been reported by Teates who evaluated the effect of massive irradiation of the hemithorax in the dog (*12*). Using a densimetric method to quantify serial lung scans, he found that the percent of blood flow to the irradiated lung fell from a pretreatment mean value of 50% to a value of about 20% 400 days after irradiation.

Pulmonary ischemia impairs respiratory gas exchange by reducing blood flow and the pulmonary capillary volume, critical to respiratory gas exchange. It thus alters the balance between ventilation and perfusion, whose maintenance is a primary function of the lung. Ischemia also impairs nutrition of the alveoli and metabolism of the alveolar lining cells. It has been shown that surface activity of lung extract is sharply reduced 3 days after unilateral pulmonary artery ligation (*13*). Surfactant is essential for stability of alveolar structure. Its absence leads to alveolar deformity with decreased lung volume and pulmonary compliance. Therefore it is reasonable to postulate that radiation damages alveolar structure both directly by interstitial fibrosis and injury to lining cells and indirectly by alveolar ischemia with consequent effect on the synthesis and quality of surface active material.

The late effects of radiation injury of the lung are significant because of the greater aggressiveness with which carcinoma of the lung and breast is treated by the radiotherapist. Our data provide evidence of the profound irreversible effect on pulmonary blood flow caused by the acute administration of therapeutic levels of ionizing radiation to the entire lung. Further studies are needed to quantitate the effect of identical doses administered incrementally.

SUMMARY

The effect of graded doses of ionizing radiation on pulmonary blood flow was measured quantitatively in the Swiss mouse. The experimental data indicate that pulmonary blood flow is reduced following radiation doses ranging from 100 to 4,000 R. The intensity of radiation-induced pulmonary ischemia is affected by the size of the radiation dose and the length of survival following treatment. There was no evidence of a limiting process nor of recovery from radiation-induced pulmonary ischemia during survival periods extending to half the life span of the experimental animal.

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