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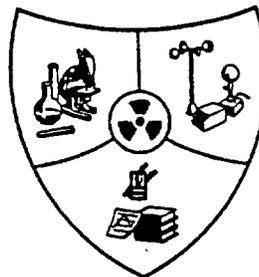
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HISTOPATHOLOGY OF SHEEP THYROID IN
CHRONIC ADMINISTRATION OF I¹³¹

By

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Biology Section
Radiological Sciences Department

August 23, 1955

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ABSTRACT

• The pathologic effects in the thyroid glands of sheep receiving chronic oral administration of I^{131} are reported. Dose levels employed ranged from 0.15 μc to 1800 μc per day.

No pathologic alterations were observed with a daily feeding level of 0.15 μc after a maximum period of observation of four years. The principal early manifestations of thyroid damage were interfollicular fibrosis in adults receiving 5 μc daily and interfollicular edema in their offspring. Focal inflammation was the initial effect resulting from a daily dose of 480 μc . At feeding levels between 5 and 480 μc per day, various combinations of edema, inflammation and fibrosis occurred. Prolonged administration of higher levels of I^{131} caused necrosis of follicles or infarcts. Progressive arterial damage was observed, including intimal thickening, changes in the internal elastic membrane, destruction of the smooth muscle of the media and eventual obliteration of the lumens.

The pathologic changes observed involve the interstitial tissue, epithelium and blood vessels. The effects on the interstitial tissue may be considered of primary significance in the early diagnosis of low-level chronic exposure to I^{131} in the species investigated.

HISTOPATHOLOGY OF SHEEP THYROID IN
CHRONIC ADMINISTRATION OF I¹³¹

INTRODUCTION

A study of the effects of the chronic administration of I¹³¹ on sheep has been in progress at this installation for over four years. (1-3) Since I¹³¹ is an important product of atomic fission and is present in the gaseous wastes of chemical processes in the manufacture of plutonium, the investigation of its toxicology when administered chronically is a matter of immediate interest. The primary purpose of this experiment was to furnish data indicating a maximum level of chronic administration of I¹³¹ which is not productive of harmful effects in sheep, a range animal common to this geographic area, such data also being of potential accessory value in the defining of similar levels in other large animals. In the course of this investigation an opportunity was afforded to observe pathologic effects resulting from the chronic administration of I¹³¹ at various levels of dosage. The present report is concerned with the pathologic effects in the thyroid gland.

The pathology of the thyroid gland following the acute administration of I¹³¹ experimentally has been described in dog, (4) rabbit, (5) rat, (6-8) and mouse. (9-11) The pathologic effects in thyroid tissue of man following therapeutic clinical use of I¹³¹ have been reported for euthyroid patients (12, 13) and for hyperthyroid patients. (13-15) However, no studies involving the chronic administration of I¹³¹ either clinically or experimentally have been reported.

•
METHODS

The principal phase of the experiment has involved the daily oral administration of carrier-free I^{131} in feed pellets to Suffolk sheep. The sheep were maintained on a marginally goitrogenic diet to insure maximum uptake of I^{131} by the thyroid. The method of administration insured ingestion of the total dose at each feeding. Details of the experimental design and instrumentation are described by Bustad *et al.*⁽¹⁶⁾ in a separate report. Necropsies were performed on the animals after sacrifice by exsanguination under sodium pentobarbital anesthesia or after spontaneous death. Organs showing significant autolysis were deleted from the study. The tissues were fixed in Bouin's fluid and stained routinely with hematoxylin and eosin. Mallory's aniline blue stain and Romeis' orcein stain were used when needed,

In order to achieve the purpose of defining a maximum daily level of administration causing no manifest abnormality, a wide range of feeding levels of the I^{131} was originally employed, including doses of 0.005 μ c, 0.15 μ c, 5.0 μ c, and 240 μ c daily, as well as controls. Subsequent daily feeding schedules of 0.5, 1.5, 15, 30 and 45 μ c were added to further narrow the range of uncertainty as to the maximum nontoxic level. Auxiliary experiments were conducted to explore problems of special interest, furnishing supplementary pathologic data as a by-product. The number of animals in the various feeding levels employed in this study appear in Table I.

TABLE I

Number of Animals in Various Feeding Levels

Daily Dose of I^{131} in μc	Control	0.15	5	15	30	45	135	240	480	1800
Adults	16	12	9	3	6	0	0	10	16	3
Offspring	52	2*	18**	9	6	5	7	27	0	4

*Does not include 35 lambs sacrificed at or before 4 months of age.

**Does not include 39 lambs sacrificed at or before 4 months of age.

In an attempt to approximate conditions on the range for the purpose of evaluating the hazard to grazing sheep from chronic range contamination, the exposure was carried out throughout the cycle of reproduction. The original feeding of I^{131} to adult animals was continued through the period of gestation and lactation so that the offspring were exposed to I^{131} in utero and during the suckling period by way of the maternal milk. The daily oral feeding of I^{131} was then begun on the offspring at weaning (about four months of age) and continued for the duration of the experiment. The regimen employed thus provided the offspring with an exposure comparable to what they might be expected to receive under natural conditions throughout their life cycle if their dams and, subsequently, they were exposed to the given quantity of I^{131} .

The inclusion of the fetal period and first year of life in the exposure of the offspring necessarily leads to a considerable variability in the intake of I^{131} . The intake in the fetus is dependent upon the quantitative partition of the ingested I^{131} between the ewe and fetus and the stage of functional development of the thyroid gland in the fetus. ⁽¹⁷⁾ Subsequent to birth, it is dependent upon the quantity of milk ingested by the lamb and the concentration of I^{131} in the milk. After weaning the intake is constant, since the

lambs are then fed the spiked pellets used for each group. However, the ratio of I^{131} intake to body or thyroid weight is somewhat higher in the weaned lambs until maturity than in the adults.

The photomicrographs are accompanied by graphs of the variation of the concentration of I^{131} in the thyroid with time for each animal. The estimated cumulative thyroid exposures in rad units are also included. The following equation was used in the calculation of thyroid exposure:

$$\text{Dose (rad)} = \frac{51 \times E \times q \times t}{W}$$

51 = disintegrations/ μc /day x ergs/Mev x rad/erg

E = average energy of beta particles in Mev

q = mean of quantity deposited in thyroid in μc as determined by external monitoring or estimated in the case of in utero exposure.

t = days exposed to I^{131}

W = weight of thyroid gland in grams estimated on the basis of average organ weights at necropsy for various ages and seasons.

This formula assumes that all of the energy of the beta particles is absorbed within the thyroid gland, ignoring the loss to surrounding tissue. The gamma component was ignored since it was estimated to be less than 10% of the beta component. In view of the number of inexact estimates required, the calculated dosages are considered to be crude approximations. Periods of feeding during which measurable amounts of radioiodine were not concentrated because of thyroid ablation were deleted from the graphs.

RESULTS

The variation in I^{131} intake of the offspring compared with the adult as well as differences in response between mature and immature tissues make it necessary to describe the findings in the adult animals separately from their progeny in several instances.

Histology of the Normal Sheep Thyroid

The normal thyroid gland of the sheep shows the basic pattern observed in the human and in common experimental animals. The follicles are intimately apposed to each other, being separated only by a rich capillary network, which is not readily apparent in the microscopic sections, and a delicate reticular stroma. The lability of the follicular epithelium is demonstrated in the sheep thyroid. The histologic pattern varies from that of large follicles having low cuboidal epithelium to an appearance of smaller follicles lined by high columnar epithelium with peripherally vacuolated colloid. The nuclei are usually round to oval and contain finely dispersed chromatin. Fibrous septa of moderate size, which contain blood vessels and nerves, traverse the parenchyma. The periphery of some of the adult thyroids show fibrous tissue between the follicles, a finding most striking in the outermost layers and present only in relation to septa in central portions of the glands. This normal histologic feature was a source of difficulty in evaluating slight histologic damage, since fibrosis was the principal evidence of damage in the adults of the groups receiving low levels of I^{131} .

A high percentage of the glands contain islands of stratified squamous epithelium which are frequently cystic, resembling small epidermoid cysts of human skin (Figure 1). Internal parathyroid tissue is frequently present and thymic tissue rarely so (Figure 2). Lymphocytic infiltrations and adenomatous nodules were not encountered in the normal thyroid gland.

Adults Administered 0.15 or 5 μ c Daily

Histologic damage was not observed in animals receiving 0.15 μ c of I^{131} for a maximum period of four years. Members of the groups introduced later at levels between 0.15 and 5.0 μ c as well as offspring of the 0.15 μ c group of an age greater than six months have not yet been sacrificed.



FIGURE 1.
CYSTIC ISLANDS OF STRATIFIED SQUAMOUS EPITHELIUM
IN NORMAL SHEEP THYROID. (150x)

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FIGURE 2.

INTERNAL PARATHYROID GLAND AND ECTOPIC THYMIC TISSUE WITHIN NORMAL THYROID. PARATHYROID TISSUE PARTIALLY SURROUNDS THYMIC TISSUE. HASSALL'S BODY PRESENT WITHIN THYMIC TISSUE NEAR RIGHT MARGIN. (150x)

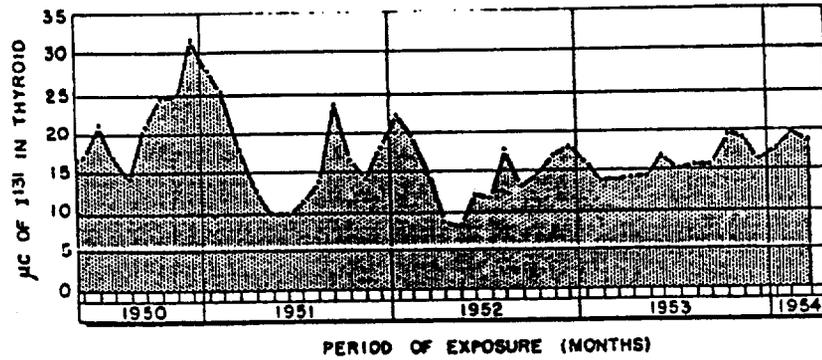
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The lowest daily dose productive of pathologic damage was 5 μ c. (18) Seven of nine adult animals in this group showed interfollicular fibrosis, which was the principal pathologic effect observed. The fibrosis was slight in six glands (Figure 3) and mild in one (Figure 4). In view of the previously mentioned normal occurrence of fibrous connective tissue between follicles in the periphery of the glands and adjacent to septa in deeper locations, especially in older animals, differentiation between the fibrous connective tissue observed as a feature of the histology of the gland and that occurring pathologically was extremely difficult in cases of minimal damage. As a manifestation of damage, the fibrous tissue had a random distribution and a rather compact texture. When present in minimal form, the distribution of the scar tissue was focal, but the generalized nature of the distribution became obvious with the developing severity of the process. The only deviation from the normal pattern in the epithelium was the presence of a rare enlarged nucleus in an epithelial cell. However, there was a diminution in the average size of the follicles in affected glands. The presence of damage could not be established in two glands.

Vascular effects of a minimal character were observed in a few thyroids of this group. The arteries involved were of medium size and showed focal thickening of the intima due to fibrosis with splitting and fraying of the internal elastic lamina. The media and adventitia were unchanged, and the arterioles showed no evidence of pathologic alteration.

The initial damage was observed at 35 months of feeding. Animals of this group who expired prior to this age exhibited significant autolysis and were not included in the results. For this reason 35 months may not represent the minimal period of feeding of 5 μ c per day at which morphological effects can be initially observed in an adult animal. Additional animals are to be sacrificed at earlier intervals in a continuation of the experiment.



PERIOD OF EXPOSURE (MONTHS)
ESTIMATED TOTAL DOSE TO THYROID = 22,000 RAD.

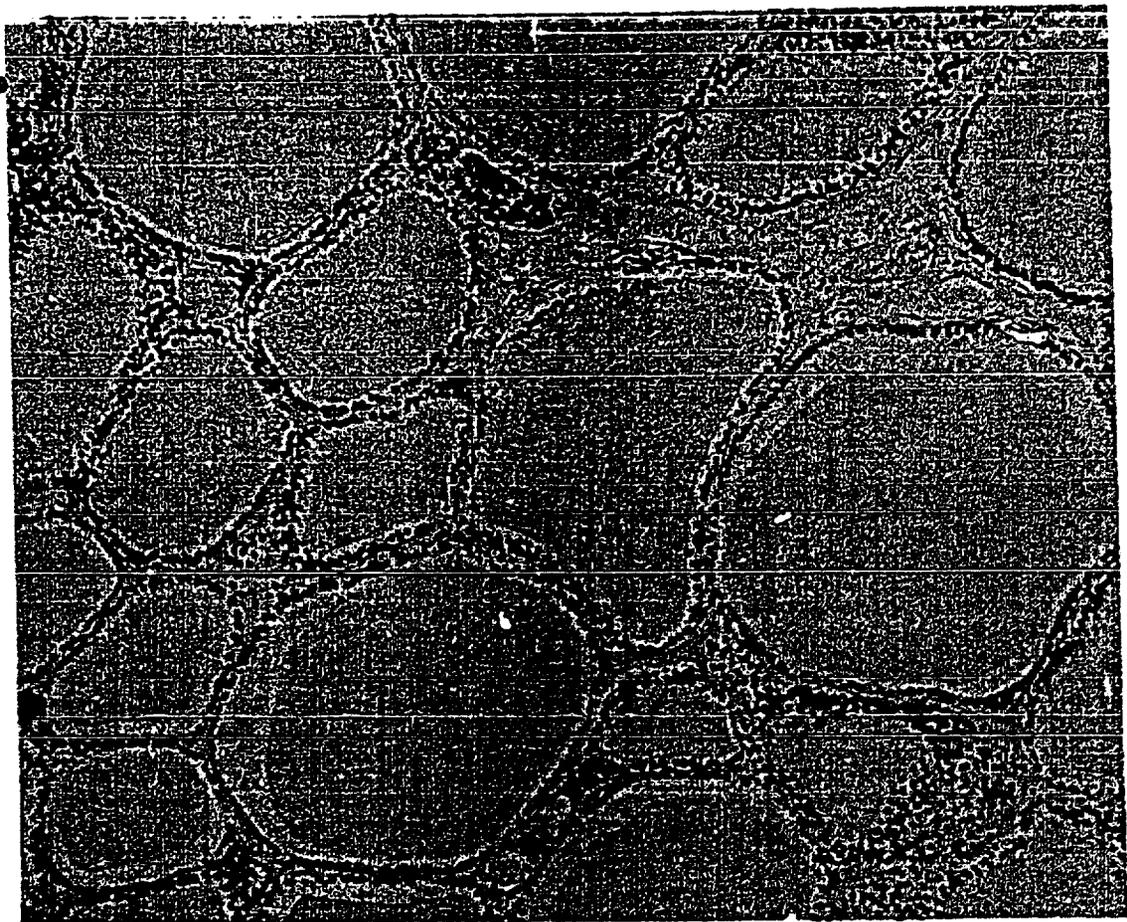


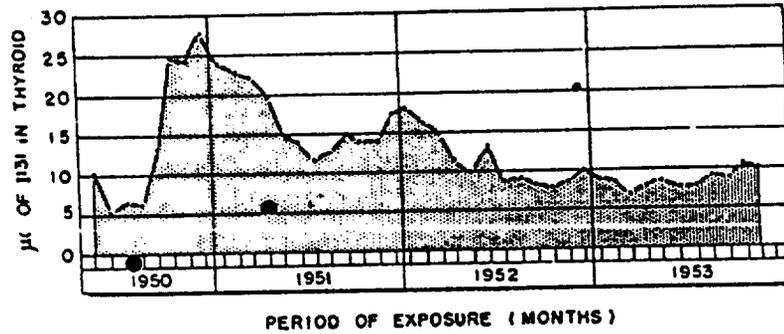
FIGURE 3.

MINIMAL INTERFOLLICULAR FIBROSIS IN THYROID OF ADULT
EWE RECEIVING 5 μc ^{131}I /DAY FOR 47 MONTHS, (135x)

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ESTIMATED TOTAL DOSE TO THYROID = 26,000 RAD.

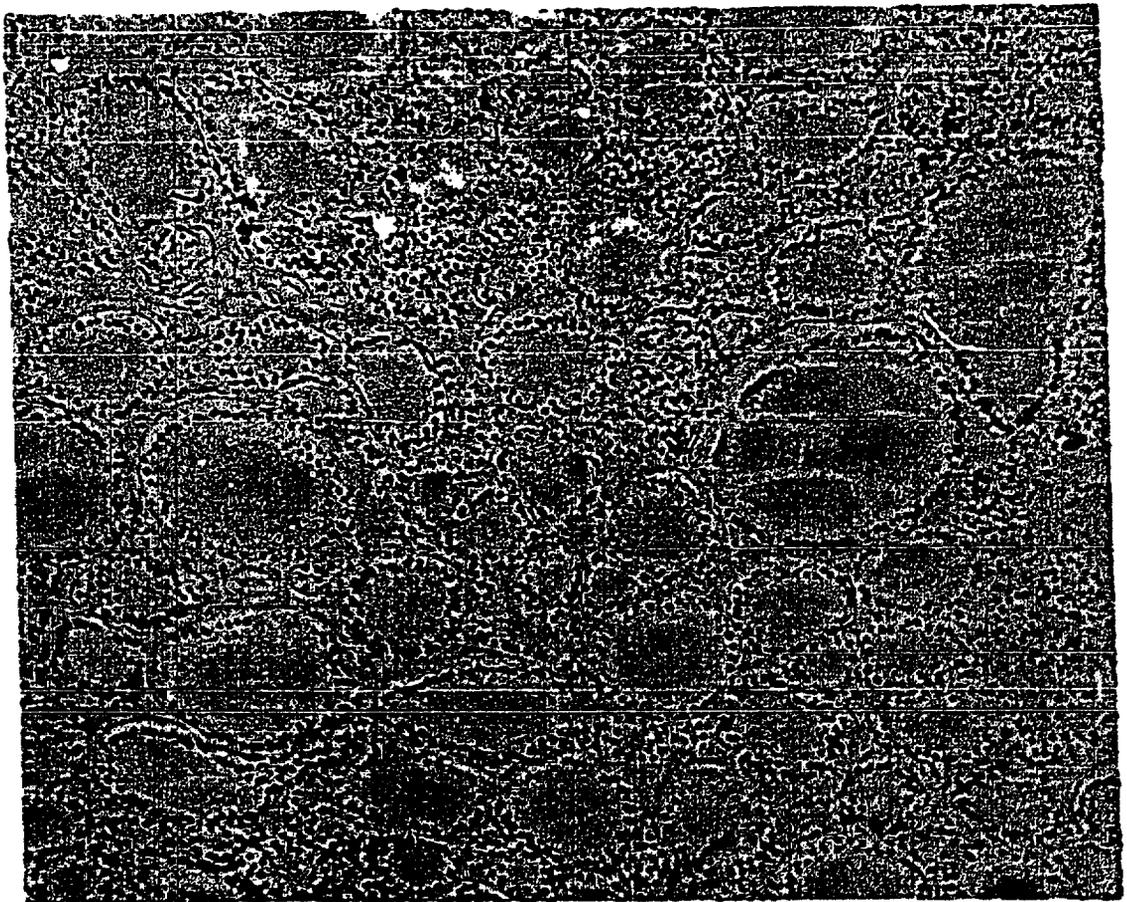


FIGURE 4.

MILD INTERFOLLICULAR FIBROSIS IN THYROID OF ADULT
EWE RECEIVING 5 μc ^{131}I /DAY FOR 43 MONTHS. (135x)

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Offspring Administered 5 μ c Daily

Tissue damage was observed in the thyroid glands of 12 of 15 of the offspring of 5 μ c animals at an age of eight months or older. The principal effect was focal interfollicular edema in contrast to the fibrosis described in the adults. Within edematous areas the follicles were rounded and appeared to be suspended in a fluid medium (Figure 5). In early cases, the interfollicular fluid was essentially devoid of fibers, but a fibrous component appeared in the glands exposed for longer periods of time. Mild infiltrations of lymphocytes were observed in a few glands.

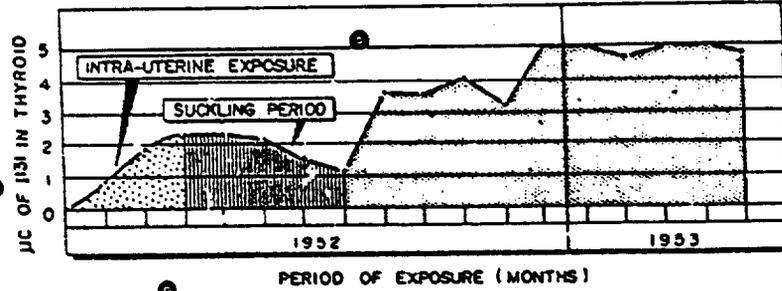
Intermediate Feeding Groups

The thyroid glands of three ewes fed 15 μ c per day for a period of two years showed generalized mild effects. Fibrosis was present between the follicles, being rather loose and tending to an edematous appearance in one gland (Figure 6). The pattern of the follicles was undisturbed. However, the nuclei of the epithelial cells showed mild variation in size with slight clumping of the chromatin to form small chromatin knots. The arterial effects, which were questionable in the 5 μ c ewes, were more definite at 15 μ c, the intima of certain arteries showing distinct fibrous thickening (Figure 7).

Two offspring of the above 15 μ c ewes, sacrificed at an age of four months, had normal thyroid glands. Four lambs which were sacrificed at 15 months of age showed edema and fibrosis of moderate severity. Occasional lymphocytes were observed in the stroma. In the lambs born to the same ewes a year later, two of three thyroids of lambs sacrificed at four months were damaged, showing definite edema.

Six ewes at 20 months of age received 30 μ c/day of I¹³¹ for a period of six to seven months. All members of this group showed minimal evidence of damage. Slight interfollicular fibrosis was observed in five glands.

The sixth gland revealed a small accumulation of lymphocytes without evident fibrosis.



ESTIMATED TOTAL DOSE TO THYROID = 10,000 RAD.

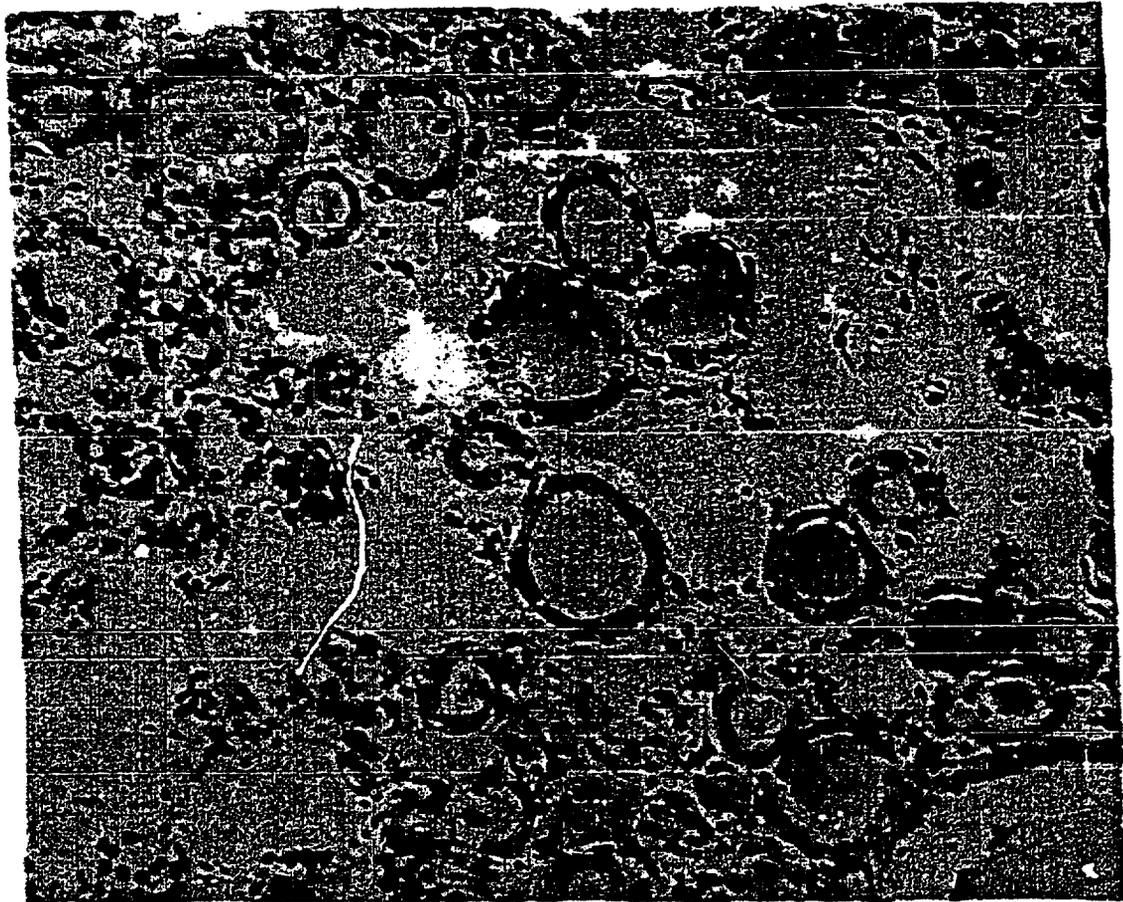


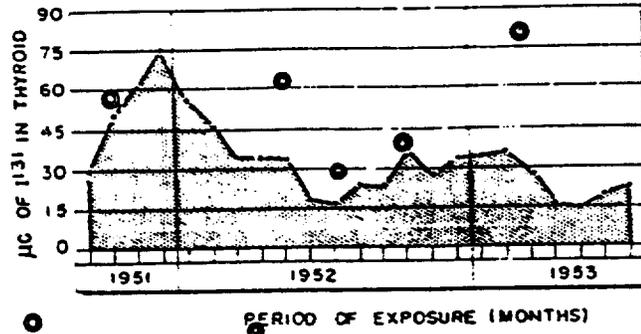
FIGURE 5.

MODERATELY SEVERE EDEMA BETWEEN FOLLICLES IN THYROID OF 14-MONTH-OLD 5 µc OFFSPRING. (135x)

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ESTIMATED TOTAL DOSE TO THYROID = 26,000 RAD.

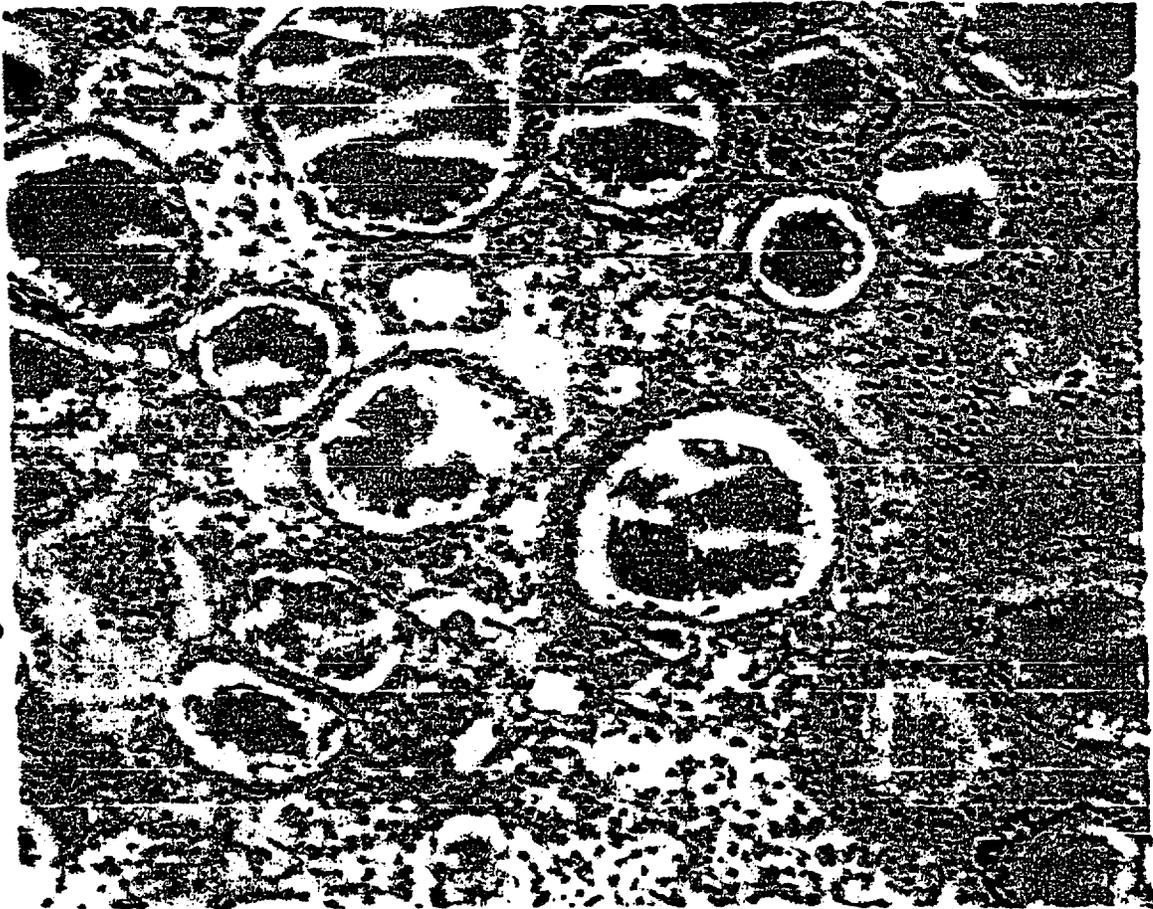


FIGURE 6.

FIBROSIS AND FOCAL EDEMA BETWEEN FOLLICLES IN
THYROID OF EWE RECEIVING 15 µc ¹³¹I/DAY FOR TWO YEARS.
(135x)

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FIGURE 7.

ENDOTHELIAL HYPERPLASIA AND INTIMAL FIBROSIS IN
ARTERY OF THYROID SHOWN IN FIGURE 6. (150x)

Six offspring of ewes fed 30 $\mu\text{c}/\text{day}$ of I^{131} were exposed in utero during the last two trimesters of pregnancy and subsequently during the period of suckling. They were sacrificed at periods between 10 and 130 days after birth. The thyroid gland of the lamb sacrificed 10 days after birth was normal. The thyroid glands of the remaining five lambs, including one sacrificed at 29 days, were damaged, all but one showing edema. Fibrosis and lymphocytic infiltration were associated findings in two glands. Only fibrosis was encountered in the fifth gland.

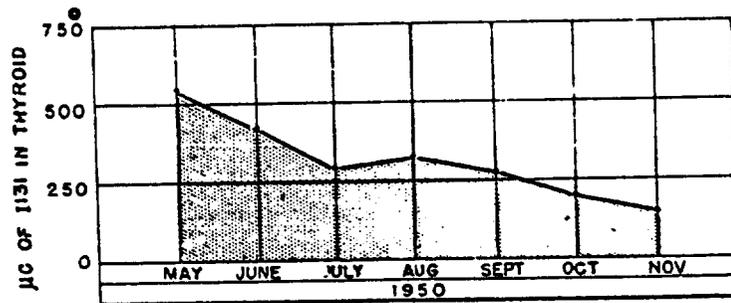
Five offspring of ewes fed 45 μc per day showed histologic damage. Two lambs were newborn, and the remaining three were sacrificed at an age of four months. Of the two newborn lambs, one showed minimal and the other severe edema. Edema also predominated in the glands of the four-month lambs, being accompanied by mild fibrosis in one and by slight inflammation in another.

Seven offspring of ewes fed 135 μc per day showed thyroid damage. Three lambs were newborn, two were one month and two were four months of age. Edema of mild to moderate severity was present in all glands, being accompanied by fibrosis in two of the seven.

Adults Administered 240 μc Daily

The effects of prolonged administration of high levels of I^{131} were observed in the group of two-year-old animals which received 240 μc per day for periods up to 450 days.⁽¹⁹⁾ Two animals of the group died during the period of administration while eight others died or were sacrificed subsequent to the termination of I^{131} feeding.

The first necropsy on an animal of this group was performed six months after the feeding was initiated. Microscopically, the gland showed a widespread separation of follicles by fibrous tissue (Figure 8). The fibrous tissue was predominantly mature and occasionally had an acellular,



ESTIMATED TOTAL DOSE TO THYROID >80,000 RAD.



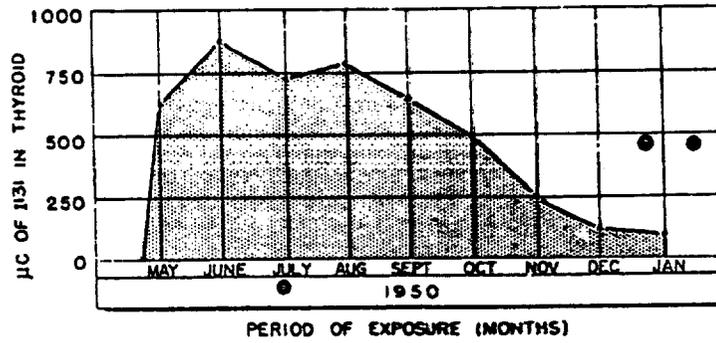
FIGURE 8.

MODERATE INTERFOLLICULAR FIBROSIS IN THYROID OF ADULT EWE RECEIVING 240 µc ¹³¹I/DAY FOR SIX MONTHS. FEW DESQUAMATED EPITHELIAL CELLS IN LUMENS OF FOLLICLES. (135x)

hyalinized appearance, although loose, edematous areas were also noted. Inflammation occurred in the fibrous stroma in the form of scattered infiltrations of lymphocytes. The follicles showed a moderate reduction in size in central areas. The epithelial cells were generally normal in appearance, apart from a mild variation in nuclear size with slight condensation of chromatin. A few cells showed hydropic degeneration of the cytoplasm, causing a ballooned appearance. Similar desquamated cells were present in the lumens of a few follicles.

After a year of feeding, the thyroid gland of a second ewe showed dense fibrosis. The follicles were substantially reduced in size and showed a frequent absence of distinct lumens. The epithelial cells varied in size and shape. Nuclear irregularity was prevalent, many nuclei being moderately enlarged and showing prominent chromatin knots. A central pallor was observed in some nuclei while others were pyknotic. The fibrotic stroma contained scattered neutrophilic leukocytes, their presence at this advanced stage of damage being an isolated finding in this gland.

The administration of I^{131} in a daily dose of 240 μ c was continued for 450 days (about 15 months). No essential difference was observed in the histologic pattern between those animals who died or were sacrificed at the end of 15 months and others who lived on after the cessation of feeding for periods up to 24 additional months. A marked reduction in the quantity of surviving parenchyma with the residual follicles surrounded by hyalinized fibrous tissue was the usual finding in this group of severely damaged thyroids. The follicles were severely constricted (Figure 9), although a few follicles at the surfaces of the glands were normal in size and contained colloid. Large central areas were completely devoid of epithelial cells, the parenchyma being entirely replaced by the fibrous connective tissue. The epithelial cells showed a more advanced degree of alteration than that observed at earlier intervals. Many nuclei were enlarged and displayed a coarse chromatin structure. However, no bizarre or gigantic cells were



ESTIMATED TOTAL DOSE TO THYROID = 200,000 RAD.

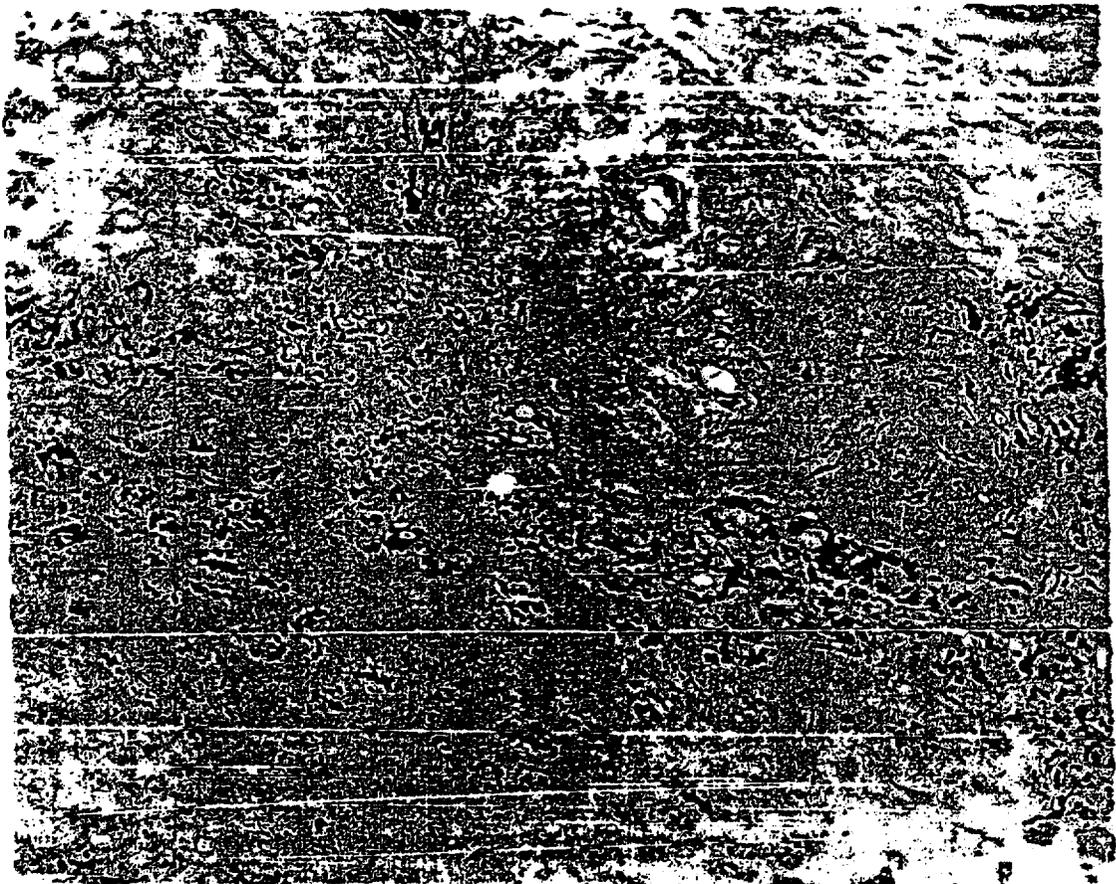


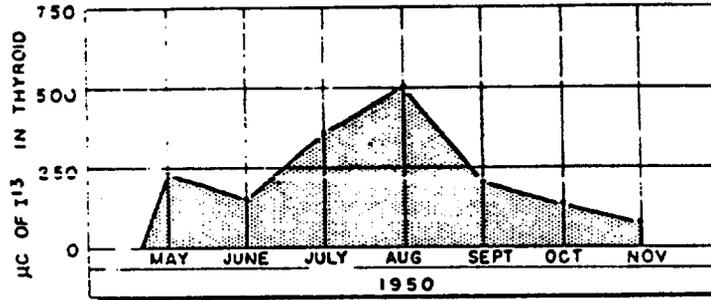
FIGURE 9.

HYALINIZED FIBROSIS WITH MICROFOLLICULAR PATTERN
IN THYROID OF ADULT EWE RECEIVING 240 µc I¹³¹/DAY
FOR 450 DAYS. (135x)

observed. Inflammation was virtually absent. One gland displayed small areas of necrosis which had an amorphous, glazed appearance. One such focus was surrounded by a layer of foreign body giant cells. These distinctive necrotic areas may have been derived from stratified squamous epithelial cysts.

- Vascular effects were pronounced in the thyroid glands of this group.
- The intimal fibrosis and changes in the internal elastic membrane mentioned previously were accentuated. The intimal thickening was present in various stages of severity, culminating in complete obliteration of the lumen. In the earlier stages, the thickened intima was moderately cellular, some of the cells being pyknotic or distorted. Subsequently, the nuclei disappeared, leaving only the connective tissue fibers. The internal elastic lamina had a frayed or disorganized appearance. Later, a gradual attenuation of the smooth muscle in the media occurred. The adventitia fused with the surrounding hyalinized fibrous tissue. In the end stage of damage the vessels could be distinguished only by remnants of elastic tissue which formed crude outlines of the pre-existing arteries.

Evidence for the importance of the direct action of radiation on arteries was obtained in two specimens of this group. In both cases large arteries immediately external to the thyroid glands of ewes fed 240 μc of I^{131} for prolonged periods showed asymmetrical damage. In the cross section of each artery, the proximal half showed typical severe damage while the distal half showed a gradation to a normal appearance. One artery showed severe endothelial proliferation in the intima with patchy areas of loss of nuclear stain in the media of the proximal half (Figure 10). In the second artery a loss of nuclear stain was present throughout the thickness of the proximal segment. The adventitia was not obviously altered in either vessel.



PERIOD OF EXPOSURE (MONTHS)
ESTIMATED TOTAL DOSE TO THYROID >70,000 RAD.

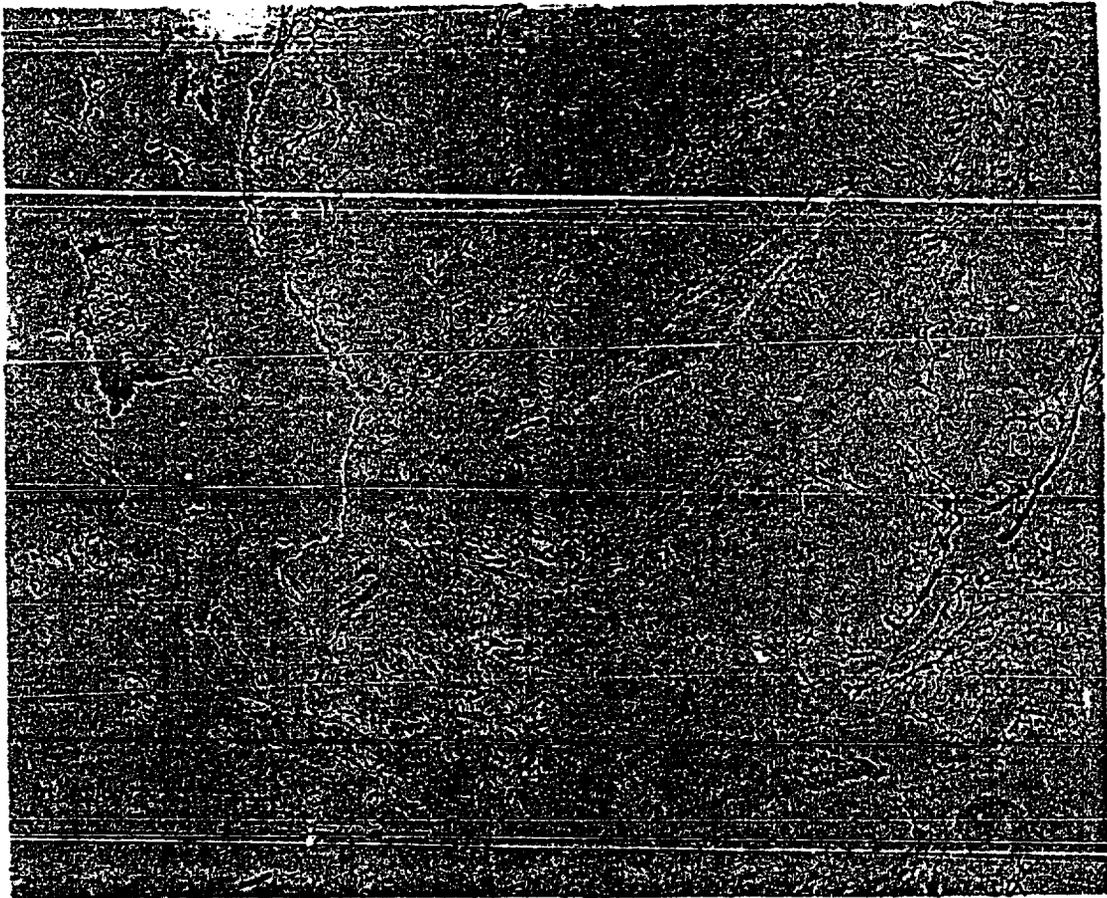


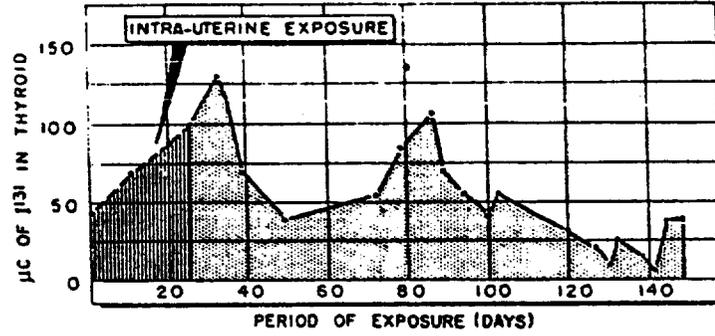
FIGURE 10.

LARGE ARTERY EXTERNAL TO THE THYROID SHOWING SEVERE INTIMAL THICKENING AND FOCAL REPLACEMENT FIBROSIS OF MEDIA IN PROXIMAL HALF OF VESSEL (LOWER LEFT) IN THE CASE OF AN ADULT EWE WHO RECEIVED 240 µc ¹³¹I/DAY FOR 450 DAYS (27x)

Offspring Administered 240 μ c Daily

The dosimetric considerations in the case of the offspring of ewes fed 240 μ c per day were complicated by the fact that the administration of I^{131} was begun at the onset of the third trimester of pregnancy. The first year offspring thus received I^{131} via the placenta only during the last third of fetal life; after birth they were exposed further by way of the maternal milk and, after weaning, by direct feeding. None of the first year offspring survived beyond 17 months of age. The original 240 μ c ewes were already severely hypothyroid at the time of delivery of the second year offspring, and the lambs were exposed to I^{131} throughout their fetal life. This group of lambs was either stillborn or lived a maximum period of five days.

The effects observed in the first year offspring were generally similar to those described in the adult. However, more specimens illustrating the earlier stages of damage were available in the offspring than in their dams. Two animals who died at birth showed no abnormal findings in the thyroid gland. Three lambs were sacrificed at four months of age. One gland showed central areas of necrosis and replacement fibrosis. Surrounding these areas a relatively abrupt transition to thyroid follicles having a normal configuration occurred, the surviving parenchyma comprising the bulk of the cross section of the gland (Figure 11). The follicles were closely arranged in the periphery and revealed cellular abnormalities. The cells were distorted, and the nuclei showed clumping of chromatin. The second gland showed fibrosis between follicles of normal size in the periphery with necrosis of the entire center of the gland. Small lymphocytic infiltrations were observed. Necrosis was absent in the third gland, but loose fibrosis between follicles was uniform throughout the sections. The follicles were only slightly reduced in size in central areas. Lymphocytes were scattered widely within the stroma and were accompanied by a few neutrophilic leukocytes. A mode of destruction of follicles by localized rupture of the follicle wall (Figure 12) and foreign body giant cell reaction



ESTIMATED TOTAL DOSE TO THYROID = 60,000 RAD.

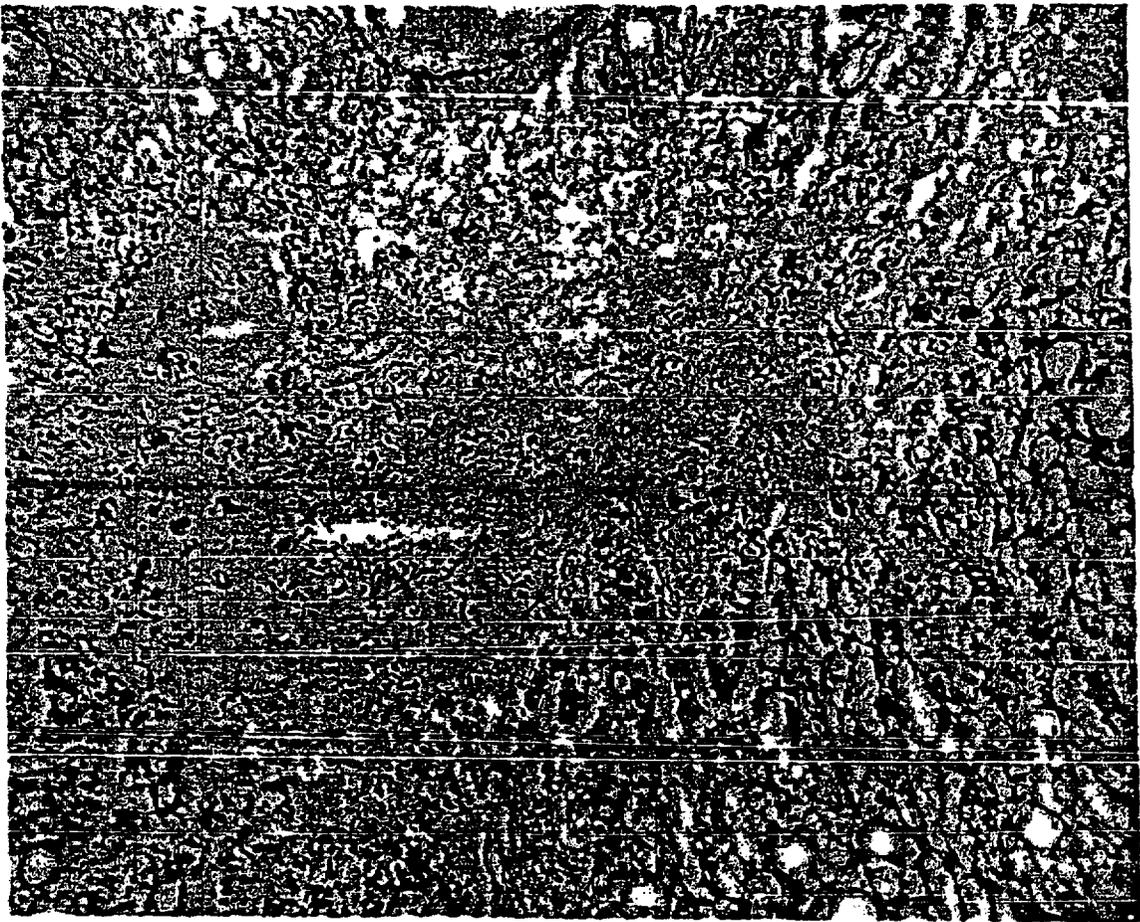
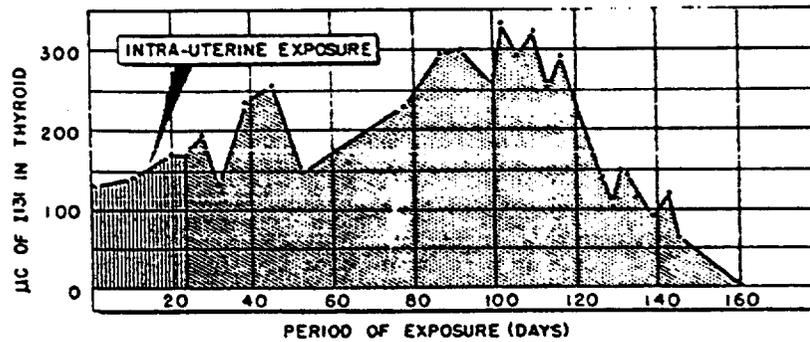


FIGURE 11.
CENTRAL NECROSIS AND FIBROSIS WITH RELATIVELY
NORMAL PERIPHERY IN THYROID GLAND OF 120-DAY-OLD
OFFSPRING OF 240 µc EWE. (90x)



ESTIMATED TOTAL DOSE TO THYROID = 100,000 RAD.

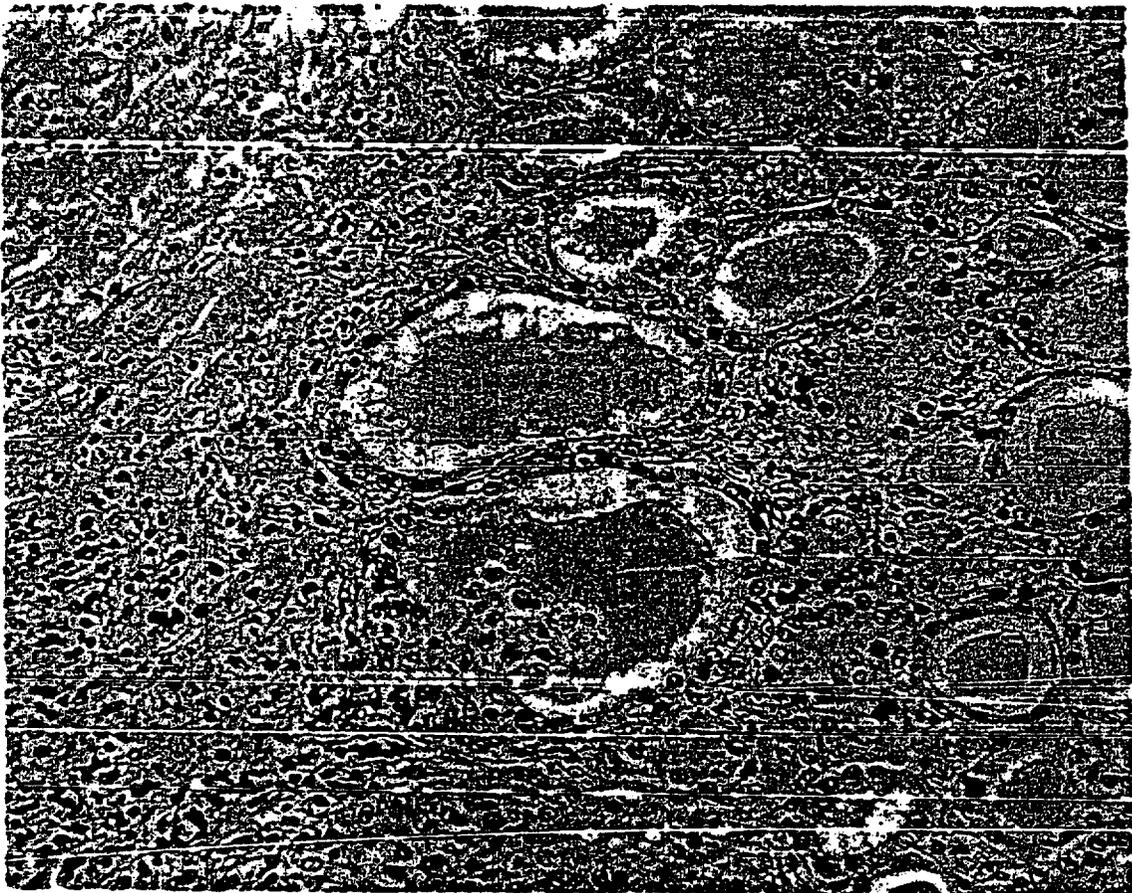


FIGURE 12.

MILD FIBROSIS AND CHRONIC INFLAMMATION IN THYROID OF 7-MONTH-OLD OFFSPRING OF 240 µc EWE. EARLY STAGE IN DISINTEGRATION OF FOLLICLE BY RUPTURE OF FOLLICULAR WALL AND INVASION OF EPITHELIAL AND STROMAL CELLS INTO LUMEN IS ILLUSTRATED. (270x)

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to follicular disintegration (Figure 13) were illustrated in this thyroid. The three glands showed a variation of effects ranging from complete central necrosis or fibrosis with no peripheral fibrosis to an absence of necrosis with diffuse interfollicular fibrosis.

The thyroid glands of seven lambs who subsequently died spontaneously at various ages up to 17 months showed more severe damage. Fibrosis was dense, and the follicles were markedly constricted. The surviving microfollicles often had a minute lumen or none at all (Figure 14). The cells showed no evident cell boundaries, the cytoplasm merging into the enveloping fibrous tissue. The nuclei were usually somewhat larger than normal and showed clumping of chromatin. Vascular changes were similar to those described in the thyroid glands of the 240 μc adult animals (Figure 15).

Twelve second-year offspring of the original 240 μc ewes, which were exposed to I^{131} via the fetal circulation throughout their fetal life, were stillborn or died during the neonatal period. The thyroid glands were reduced in size. The majority of the follicles were inordinately small in comparison with those of a normal neonatal gland, the lumens being minute. However, follicles of normal size were often present in the periphery of the glands. Definite interfollicular edema was observed in central areas (Figure 16). The edema was not accompanied by fibrosis or inflammation.

Adults Administered 1800 μc Daily and Their Offspring

Identical histologic patterns were observed in the thyroid remnants of adult ewes fed 1800 μc of I^{131} daily for 5 and 14 months. The gland in each case was replaced by a hyalinized fibrous scar with only a few scattered shrunken and distorted follicles remaining in the periphery. A third ewe received this dose level for 14 months but was not sacrificed until 19 months subsequent to the cessation of I^{131} feeding. The gland of this animal showed a most severe obliteration of the thyroid gland, the epithelial

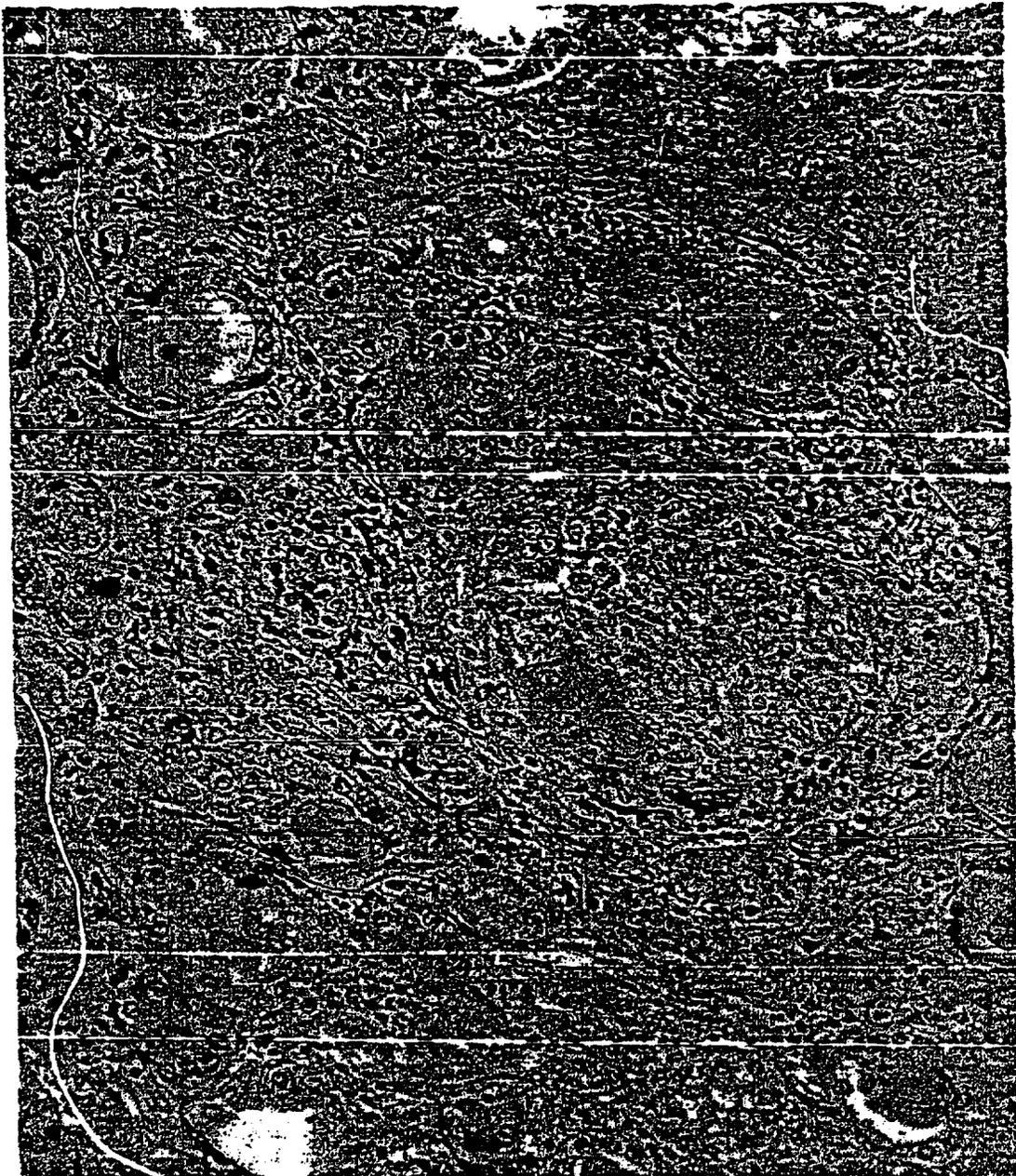


FIGURE 13.

GIANT CELLS WITHIN FOCUS OF INFLAMMATION AT SITE OF DISINTEGRATION OF FOLLICLE IN THYROID SHOWN IN FIGURE 12. (300x)

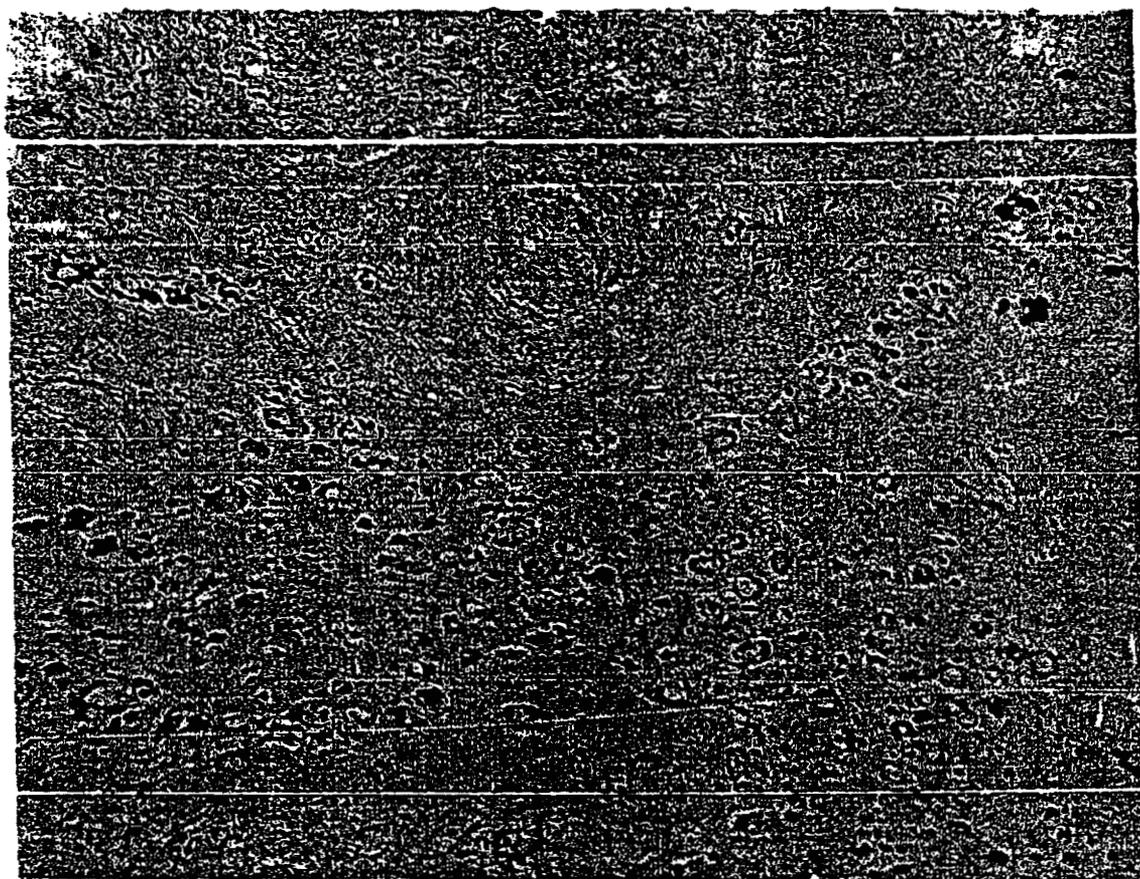
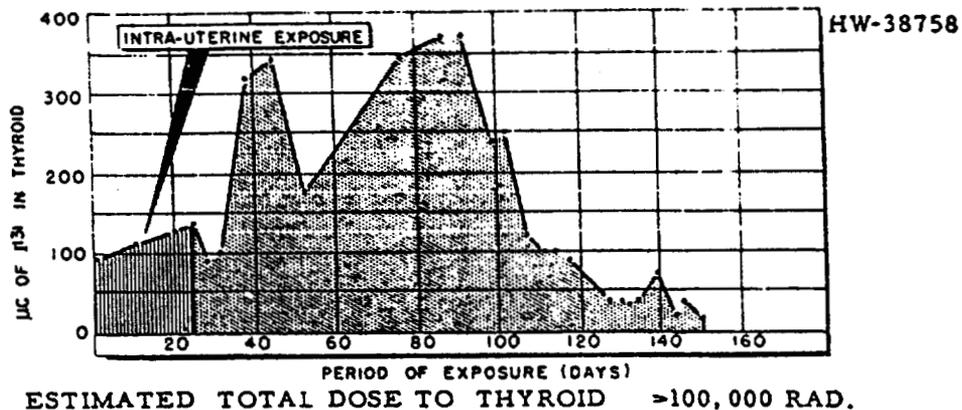


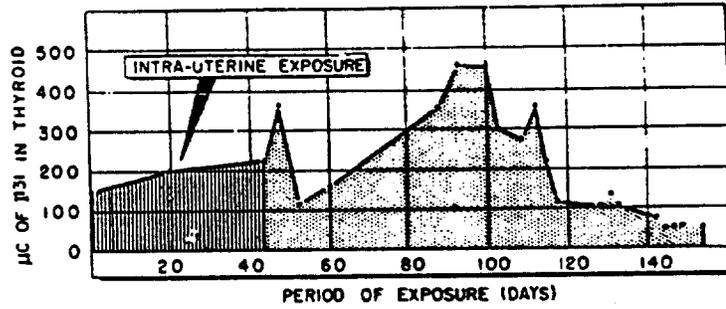
FIGURE 14.

MICROFOLLICULAR PATTERN IN 11-MONTH-OLD OFFSPRING OF 240 μc EWE. ARTERIES SEVERELY DAMAGED WITH ONLY ELASTIC TISSUE REMNANTS OF VESSELS NEAR MARGINS OF PHOTOGRAPH. (135x)

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ESTIMATED TOTAL DOSE TO THYROID >100,000 RAD.

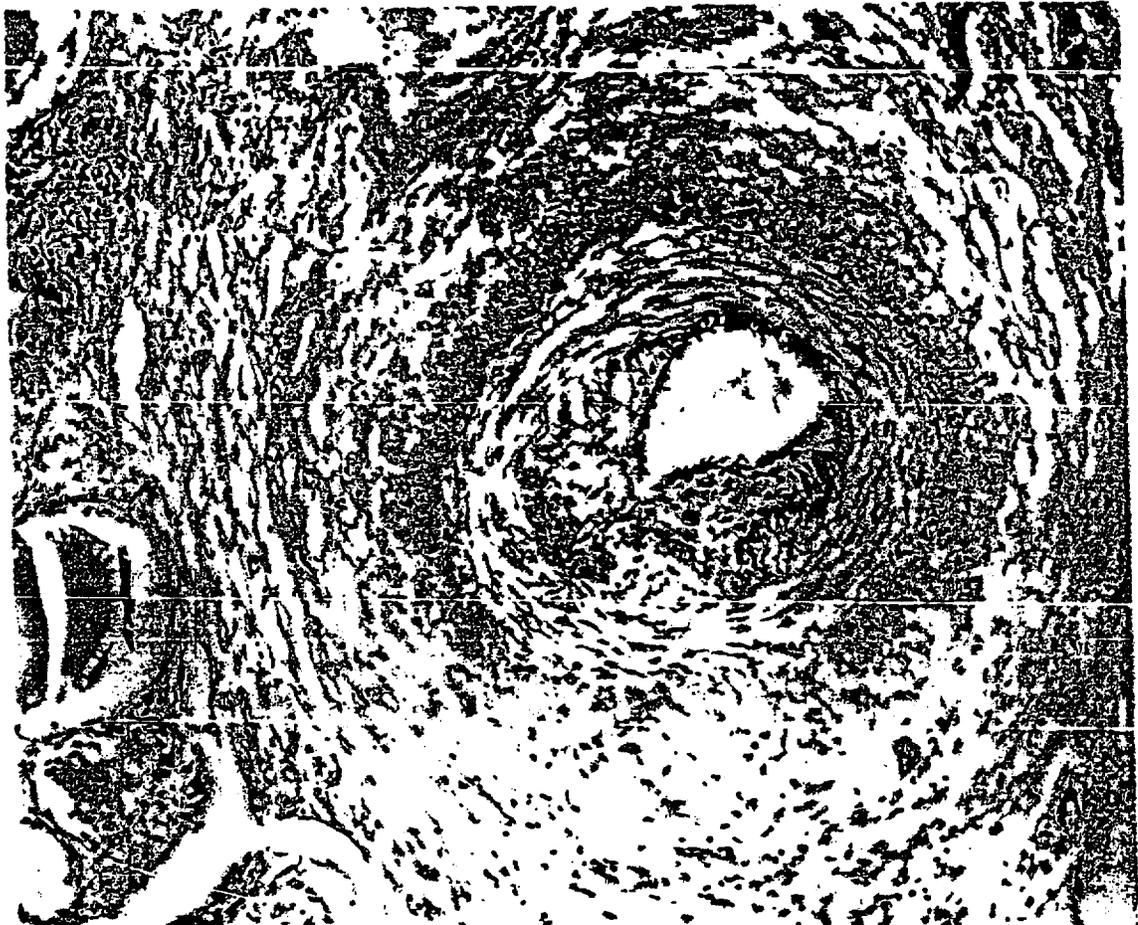
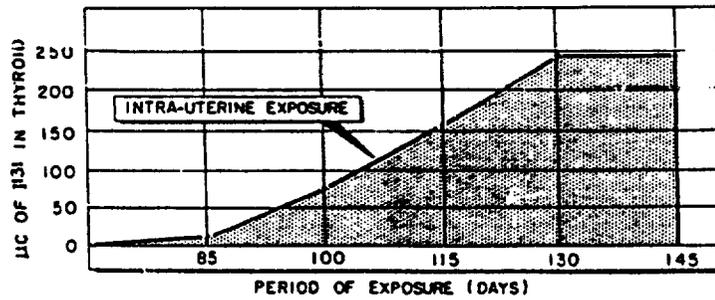


FIGURE 15.

INTIMAL THICKENING WITH NARROWING OF LUMEN OF SMALL ARTERY IN THYROID OF 4-MONTH-OLD OFFSPRING OF 240 µc EWE. (135x)



ESTIMATED TOTAL DOSE TO THYROID > 50,000 RAD.

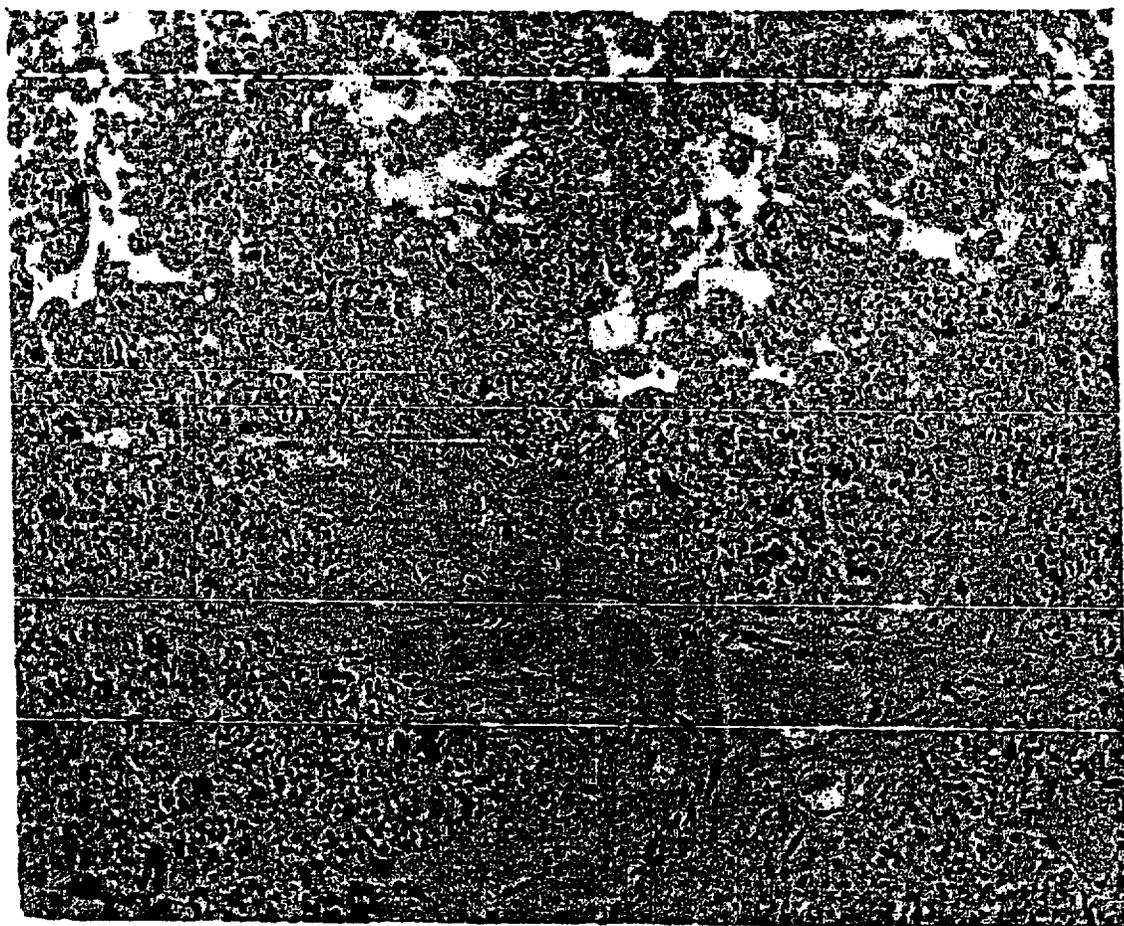


FIGURE 16.

EDEMA BETWEEN ABNORMALLY SMALL FOLLICLES IN THYROID GLAND OF NEWBORN SECOND YEAR OFFSPRING OF 240 µc EWE. (135x)

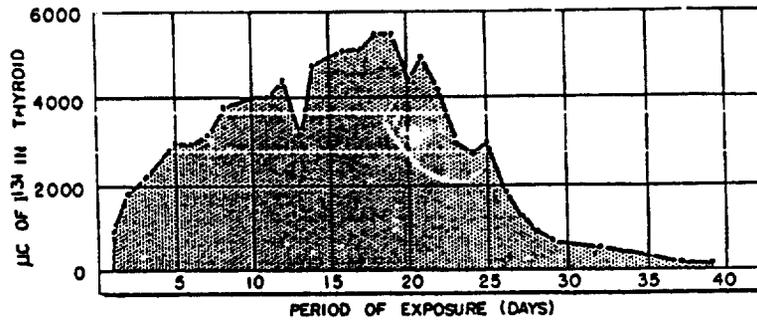
remnants consisting of only a few solitary epithelial cells and minute follicles within the hyalinized scar (Figure 17).

Thyroid glands were obtained from four offspring of the 1800 μc ewes, two being stillborn, one having lived for 15 hours and one for three days. The pathologic effects were severe in spite of the fact that the fetal exposure to I^{131} occurred only during the last trimester of pregnancy. Necrosis was observed in central areas, being accompanied by deposits of blood pigment (Figure 18). Fibrosis was observed in both central and peripheral locations.

Special Studies on Adults Administered 480 μc Daily

Two secondary experiments were performed utilizing a daily dose of 480 μc . The purposes of the first experiment were the determination of the shortest period of administration required to cause histologic evidence of damage and the observation of the early histopathologic effects at this level. ⁽²⁰⁾ In order to achieve this objective seven yearling rams were used as experimental subjects and were respectively sacrificed after 15, 19, 25, 29, 36, 43, and 70 days of feeding.

The thyroid gland of the sheep sacrificed at 15 days was normal. The first evidence of damage was observed at 19 days. Foci of inflammation were present within the parenchyma, the inflammatory cells being lymphocytes and fewer neutrophilic leukocytes. Small groups of cells suggestive of epithelial fragments were occasionally observed in the midst of the inflammation. The margins of these areas showed an invasion of inflammatory cells, principally neutrophils, into the stroma between the neighboring follicles with an early encroachment upon the follicles. The adjacent follicles occasionally showed the presence of desquamated epithelial cells within the lumens.



ESTIMATED TOTAL DOSE TO THYROID > 100,000 RAD.

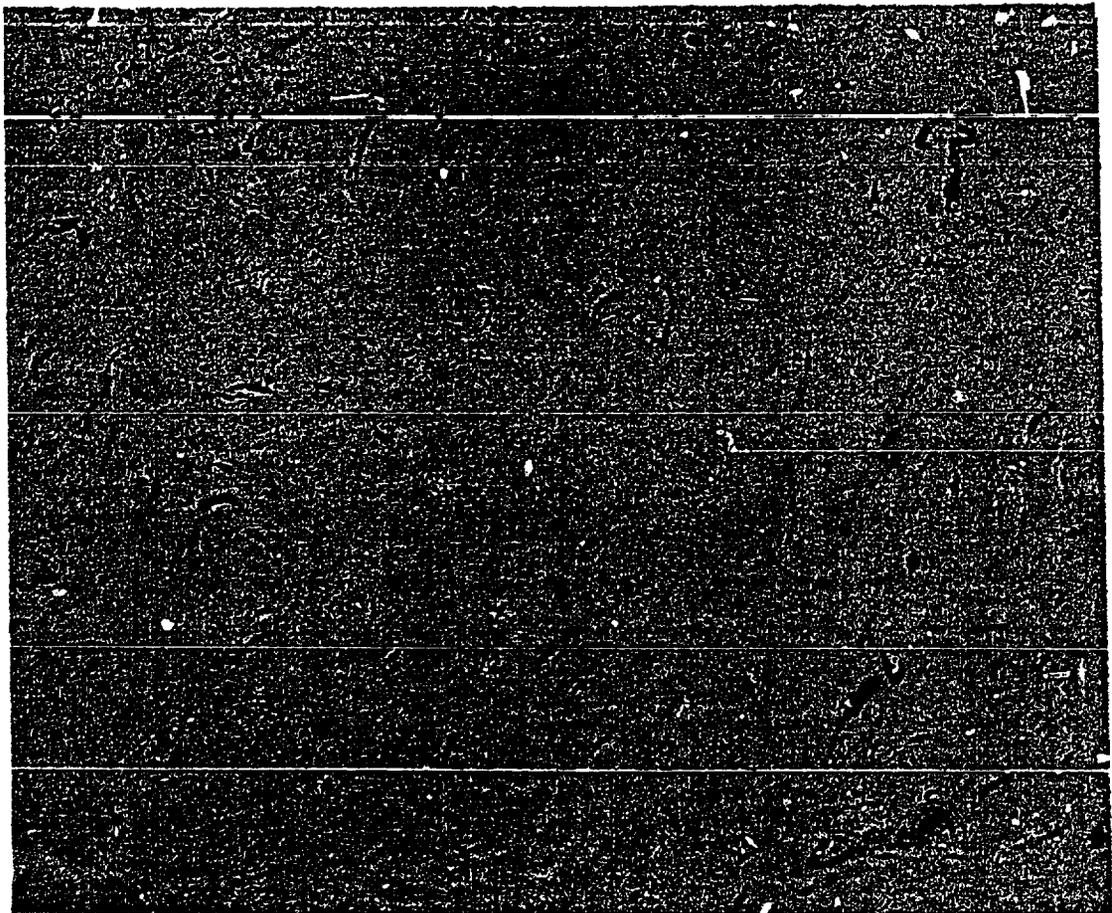
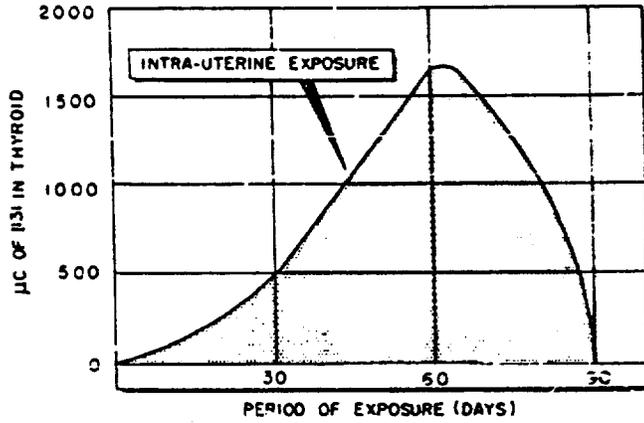


FIGURE 17.

MINUTE REMNANTS OF FOLLICLES WITHIN DENSE
HYALINIZED SCAR IN THYROID OF EWE RECEIVING
1800 µc I¹³¹/DAY FOR 420 DAYS. EPITHELIAL TISSUE
NEAR RIGHT MARGIN OF PHOTOGRAPH. (135x)



ESTIMATED DOSE TO THYROID > 100,000 RAD.

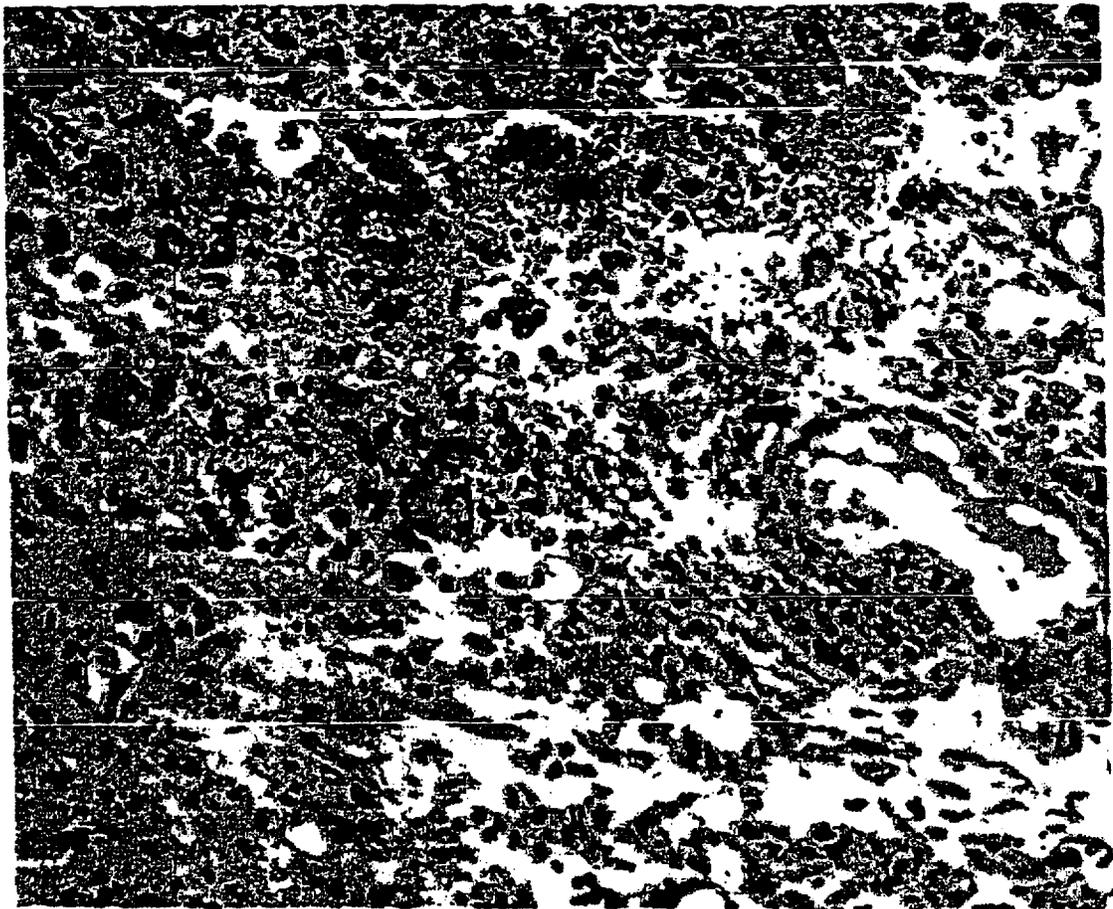


FIGURE 18.

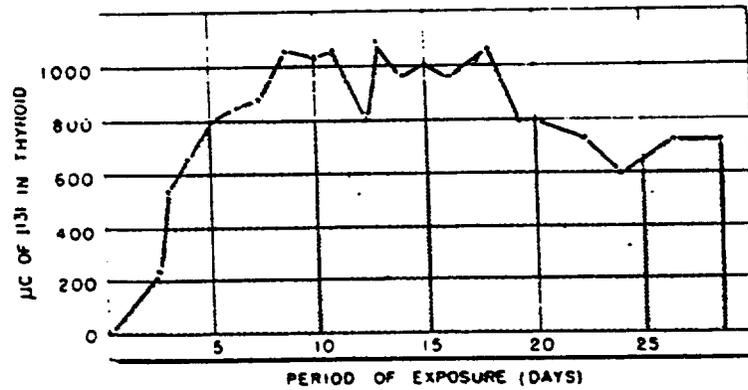
CENTRAL NECROSIS AND HEMORRHAGE IN THYROID
OF NEWBORN OFFSPRING OF 1800 µc EWE. (400x)

At 25 and 29 days, similar infrequent foci of inflammation were observed (Figure 19). However, the severity of the inflammation had increased substantially at 36 days, the areas of involvement being numerous and large. Slight fibrosis was present between follicles in areas devoid of inflammation. Early vascular damage was apparent.

At 43 days, the damage was diffuse, the follicles throughout the central areas being separated by a loose, edematous stroma containing scattered neutrophilic leukocytes and fibroblasts. An additional feature was the presence of multiple small infarcts. The severity of the process was less pronounced in the animal sacrificed at 70 days, presumably because of an individual diminished uptake of I^{131} .

The second experiment conducted at this level of administration of I^{131} was performed in order to observe differences in the effects encountered after withdrawal of the I^{131} in contrast to those observed after its continued administration. A second objective of this experiment of a clinical character was a comparison of changes in the ratio of serum iodide to protein-bound iodine (conversion ratio), in the external counting of the activity in the thyroid gland and in the histology of the thyroid gland in early damage. (21) A group of nine yearling rams was divided into three subgroups of three animals each. The first subgroup was fed I^{131} until clinical evidence of thyroid damage was obtained either by a decline in the iodide conversion ratio or in the external thyroid count and then sacrificed. The members of the second subgroup were sacrificed after being maintained on the feeding for a period of 60 days after damage was considered present by one of the above two methods. The third subgroup was taken off I^{131} feeding when the conversion ratio or thyroid count declined, but was not sacrificed until 60 days later. A progressively diminishing radiation exposure occurred in the third group, permitting a slight opportunity for recovery or stabilization of tissue damage.

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ESTIMATED TOTAL DOSE TO THYROID = 22,000 RAD.



FIGURE 19.

LARGE FOCUS OF LYMPHOCYTIC INFILTRATION BETWEEN FOLLICLES IN THYROID OF YEARLING RAM AFTER 29 DAYS OF FEEDING OF 480 µc ¹³¹I/DAY. DESQUAMATED EPITHELIAL CELLS PRESENT WITHIN LUMENS OF FOLLICLES. (270x)

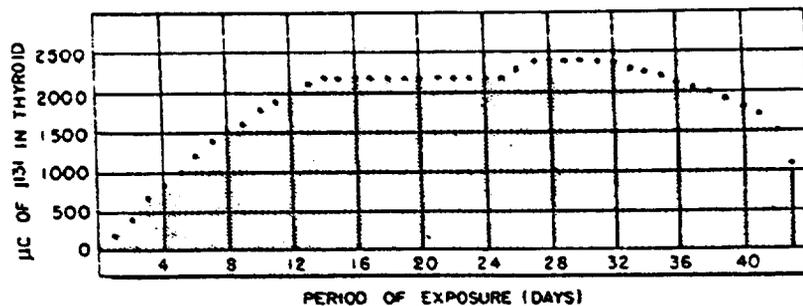
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The members of the first subgroup were respectively sacrificed at periods of 43, 44 and 57 days after the beginning of the experiment. The three glands showed a generalized separation of normal sized follicles by loose fibrous connective tissue. Lymphocytes and neutrophilic leukocytes were scattered in the stroma. Desquamated epithelial cells were often present in the lumens of the follicles. One gland also revealed multiple small infarcts (Figure 20).

In the second subgroup, the subjects were sacrificed at the end of 86, 104, and 108 days respectively, having received I^{131} continuously throughout these periods. One gland showed fibrosis with fairly good preservation of the follicles apart from large, sharply delimited infarcts. In the second gland, degeneration and distortion of the follicles were apparent in addition to fibrosis and infarction. Early calcification was evident within foci of infarction. The third gland revealed widespread necrosis, the great majority of the follicles being destroyed although their outlines were evident. Within the large areas of necrosis collections of preserved giant cells were observed (Figure 21). The giant cells were variable in size, each containing several to many nuclei in a homogeneous, eosinophilic mass of cytoplasm with irregular margins. Detritus was present within niches in the boundaries of giant cells.

The third subgroup was sacrificed 60 days after a decline in the external thyroid count or conversion ratio provided objective evidence of thyroid damage as described above. No I^{131} was administered during the 60-day period, and the animals were sacrificed 94, 115, and 121 days after the onset of the experiment. The damage was only slightly milder than that observed in the second subgroup. One gland showed generalized, moderately severe fibrosis without necrosis. Scattered foci of necrosis were present in the midst of fibrotic but reasonably well-preserved thyroid tissue in the second gland. The third gland showed a preponderance of necrosis with the presence of giant cells, the degree of damage being comparable with that of the most severely affected gland in the second subgroup.



ESTIMATED TOTAL DOSE TO THYROID = 80,000 RAD.

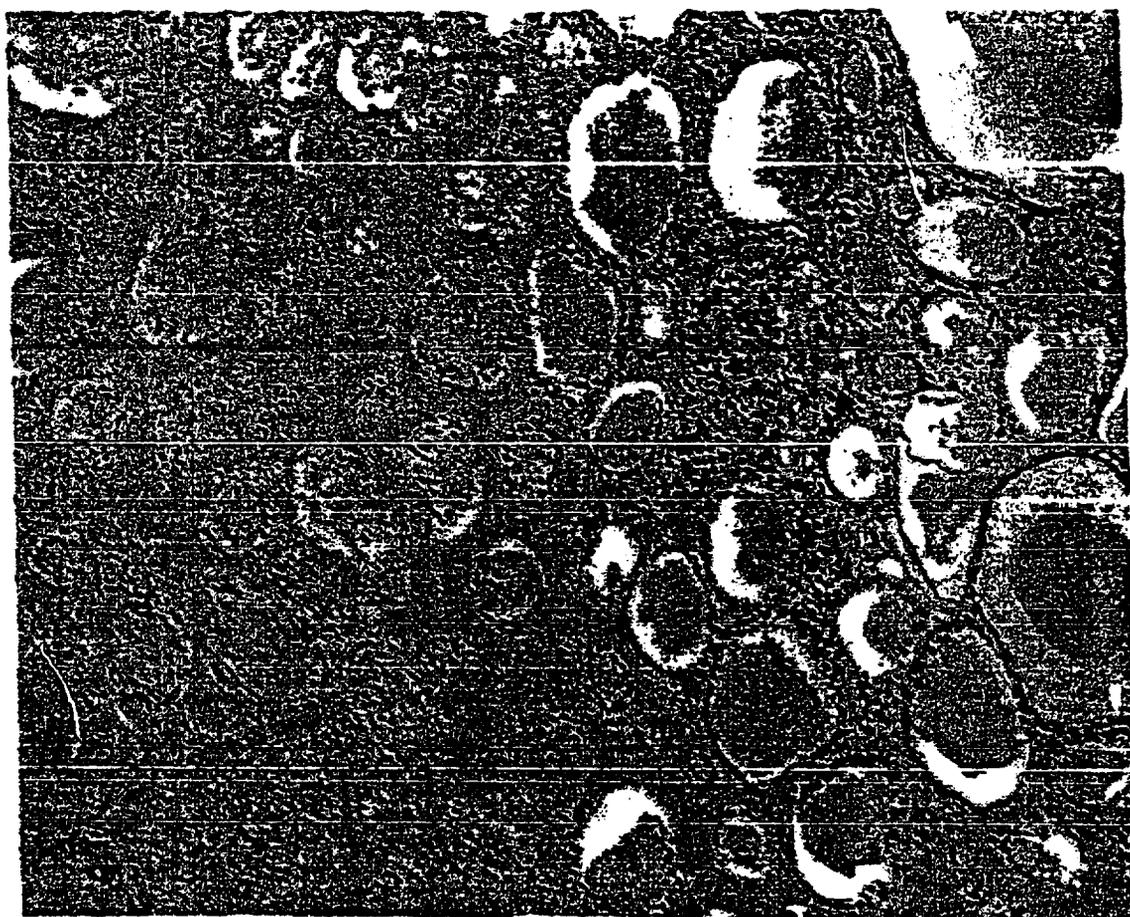
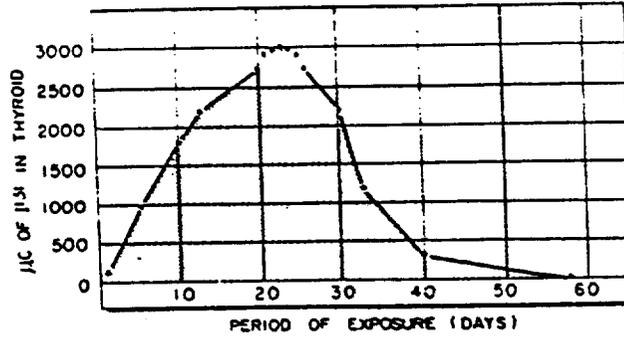


FIGURE 20.

INFARCT IN THYROID OF YEARLING RAM FED 480 µC ¹³¹I/DAY FOR 43 DAYS. (135x)



ESTIMATED TOTAL DOSE TO THYROID = 70,000 RAD.

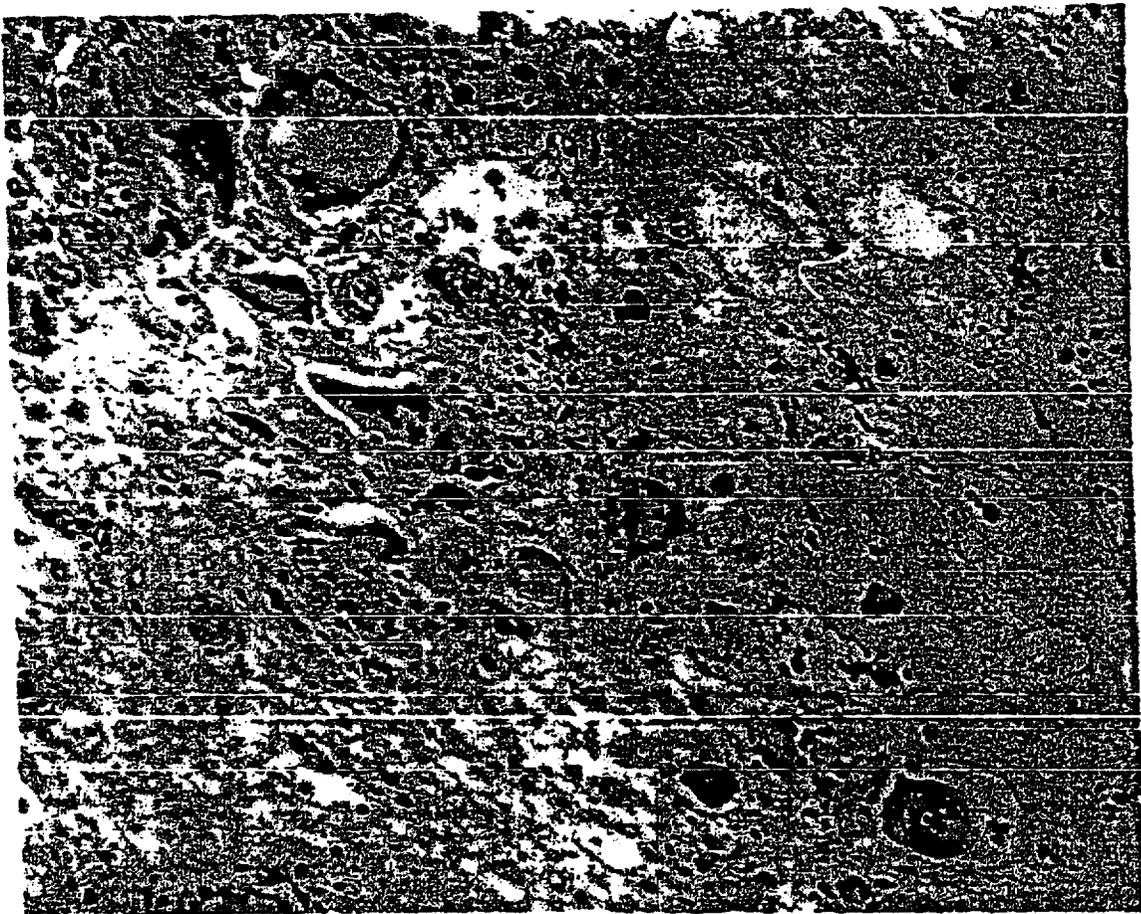


FIGURE 21.

GIANT CELLS WITHIN NECROTIC TISSUE IN THYROID OF YEARLING RAM FED 480 µc ¹³¹I/DAY FOR 108 DAYS. (270x)

DISCUSSION

The various elements of pathologic damage observed in this experiment are similar to those previously described in the clinical and experimental literature. However, the variety of controlled conditions employed permits a degree of correlation of specific effects with such factors as age and dose level. Secondly, the general histologic patterns differ in certain respects in acute and chronic administration of I^{131} .

The interstitial tissue was a more obvious site of pathologic change in the thyroid gland in this experiment. Although the epithelial effects are probably principally responsible for the functional decline of the gland, the diagnosis of early thyroid damage due to low level chronic exposure to I^{131} would best be attempted on the basis of the manifestations in the interstitial tissue.

The most characteristic tissue response in the thyroid gland of the young animal was edema. It was encountered at birth in fetuses exposed to high daily levels of I^{131} in utero, during the nursing or yearling periods in intermediate feeding levels and was the most frequent finding as late as the second year of life in the offspring of the 5 μ c group.

Interstitial fibrosis was the prominent feature of the pattern of damage in the adult animal. One may assume the fibrosis to be the end stage of preceding inflammatory or necrotizing processes. However, in the lowest injurious level, the 5 μ c group, the earlier stages postulated were not detectable, so that the fibrosis in effect assumed the significance of an initial indicator of damage. At higher levels of dosage, it was a more obvious sequel to inflammation or necrosis.

At the high level of daily administration of I^{131} of 480 μ c, foci of inflammation in the interstitial tissue constituted the earliest observed effect.

The early development of inflammation may also be expected at other high or intermediate levels where inflammation was frequently observed in association with edema or fibrosis.

The manifestations of epithelial damage ranged from slight nuclear abnormalities to extensive necrosis. Minimal nuclear alterations, such as clumping of chromatin and changes in nuclear size developed concomitantly with the early interstitial effects. However, the progression of the epithelial changes was relatively slow, making it difficult to evaluate the importance of primary as opposed to secondary epithelial damage. The former would stem from the direct action of ionizing radiation, and the latter from mechanical constriction by scar tissue and from impairment of blood supply due to vascular damage. The foci of inflammation observed early in high-level feeding may constitute a response to primary epithelial damage and early central necrosis would certainly seem to be a direct radiation effect. Evidence for a constriction mechanism is furnished by a correlation between the diminution in follicular size and the development of scar tissue. The occurrence of infarcts dramatically demonstrates one effect of vascular impairment. A phenomenon of potentially greater physiological significance at low levels is the deposition of inter-follicular fibrous tissue, interposing a possible barrier to exchange between the follicles and their capillaries. It seems likely that both primary and secondary damage contribute to the loss of functional epithelial tissue.

Pronounced cellular changes were observed in this experiment, especially in the presence of severe damage, but truly bizarre cells were not evident. Goldberg and Chaikoff⁽²²⁾ described the occurrence of carcinoma of the thyroid in rats 18 months after single large doses of I¹³¹. No tumors were encountered in the sheep thyroid glands in this experiment after a maximum period of four years*

*A metastasizing fibrosarcoma has developed in or immediately adjacent to the thyroid gland of a 53-month-old ewe offspring in the 5 μ c group during the fifth year of the experiment and will be reported separately.

Although 5 μc of I^{131} per day represents the lowest level causing damage in the autopsy material accumulated thus far, it is unlikely that 5 μc represents the absolute minimal toxic dose. An absolute minimal toxic level would be one causing a demonstrable effect only at the end of the animal's life span, the effective life span of the sheep being 6-7 years under experimental conditions. If any feeding level causes damage at a substantially earlier age, one may assume that a lower dose exists which would cause initial manifest damage at a later period in the animal's life. On the basis of the existing results, one may conclude that 0.15 $\mu\text{c}/\text{day}$ administered initially to the adult sheep is not injurious. However, the offspring of the 0.15 μc group and members of the 0.5 and 1.5 μc groups have not yet reached the age chosen for sacrifice. In view of the observation of pathologic effects in the offspring of 5 μc ewes at an age as early as 8 months, the occurrence of damage at a later age in the offspring of a group below 5 μc may be expected.

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