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EFFECTS OF RADIOSTRONTIUM ON THE BLOOD AND HAEMATOPOIETIC TISSUES OF MICE

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The general pattern of the effects of ionizing radiation, as external or internal irradiation, upon haematopoietic tissues is well known (1, 2, 3, 4, 5, 12, 13, 14, 17, 22, 25 and others). As early as 1903, *Heineke* (9) demonstrated that the changes in the circulating blood after exposure to external irradiation resulted from damage to the extremely radio-sensitive haematopoietic tissues. A similar effect for internal irradiation from radioactive isotopes was reported by *Martland* (16) in 1931.

The present report deals with changes in the blood and haematopoietic tissues observed during study of the occurrence and histogenesis of Sr^{90}-induced osteosarcomas in mice. Histologically and cytologically the bone marrow is a more sensitive indicator of radiation injury than is bone. Serial observations of the bone-marrow changes, especially shortly after the administration of Sr^{90}, can be expected to facilitate evaluation of radiation intensity and distribution within particular bones. The histological and cytological changes can also be correlated with the distribution pattern for Sr^{90} obtained by autoradiography (19). In these studies the opportunity was also taken of examining the place of the spleen in compensating for the defective bone marrow.

MATERIAL AND METHODS

Sr^{90} at 0.67 μC per gram body weight was given intraperitoneally to 200 male CBA mice, 75 to 85 days old. Two hundred male CBA mice of the same age served as controls. The mice used

in these experiments weighed between 21 and 22 g. and were fed and maintained under uniform conditions. Blood samples were obtained with a Pasteur pipette from the medial canthus of the eye of mice anaesthetised with mebumal® and were taken 6 hours, 12 hours, one, 2, 4 and 16 days and then at monthly intervals up to 10 months after the injection of Sr^{90}. Groups of ten mice given Sr^{90} and 10 control mice were sampled on each occasion. The spleen was weighed on a Mettler balance and then fixed in Stieve's fluid (23) together with a femur, tibia, humerus, and the thoracic and lumbar vertebrae. The bones were decalcified in 20 per cent formic acid under vacuum. The tissues were dehydrated and embedded in paraffin in the usual manner and then sectioned at 2—3 μ and stained with haematoxylin and eosin, van Gieson's stain, and with Lillie's (15) azure-eosinate.

Total leukocyte counts were made in a Spencer haemacytometer (American Optical) on samples diluted 1:20 in Türck's fluid. For thrombocyte counts by Nordensson's method (20), the blood samples were diluted 1:20, placed in a Bürker chamber, and allowed to stand in a moist atmosphere 30 minutes before counting under phase-contrast.

Haemoglobin levels were determined in a Ljungberg colorimeter (20 $mm.^3$ blood mixed with 3.5 ml. 0.1 per cent sodium carbonate solution) and the percentage values obtained were recalculated as g. per 100 ml. blood.

For differential counts, smears were dried in air, stained with May-Grünewald-Giemsa, and 200 cells were counted in each smear.

Conventional statistical methods have been applied to the results.

After the histological examinations of the bone marrow, representative azure-eosinate sections from a Sr^{90} mouse and a control mouse were chosen for each interval. These sections were then microphotographed in yellow light from a variocolour and enlarged 300 and 1000 times. Sections from the Sr^{90} mice and the respective control mice were then compared for cellularity, degree of sinusoid dilatation, and number of megakaryocytes. An impression could also be gained of the proportions between immature erythrocytes and leukocytes.

RESULTS

Bone marrow changes

In the distal portions of the femur (see Fig. 1), there was a distinct decrease in the cellularity of the bone marrow by 12 hours.

Cellularity in the distal femur reached a minimum after 16 days (Fig. 2) and at this time the bone marrow consisted of little more than fatty tissue. Injury to the bone marrow in the diaphysis and proximal femur was more slowly apparent. The first change in this region appeared after 24 hours and then rapidly became more severe in the proximal portion where the marrow was strongly hypoplastic but not aplastic after 16 days. In the femoral diaphysis an aplastic, oedematous, fatty marrow was not manifest before 6 or 7 months. Fig. 3 illustrates the gradual reduction in cell numbers in a portion of the distal femur following the injection of Sr^{90}.

Regeneration was apparent in some mice as early as 16 days and then in the proximal quarter or so of the femoral diaphysis. This regenerative activity appeared to be transitory and there was no distinct increase in cellularity before 2 months. All regions of the bone marrow then displayed this regeneration but again it was only temporary. In subsequent months cellularity decreased and continued to do so until, as mentioned above, aplasia was attained in the diaphysis after 7 to 9 months. Regeneration, however, was evident in the distal and proximal portions of the medullary cavity provided that tumour tissue did not fill out these regions.

Decrease in cellularity was accompanied by dilatation of the medullary sinusoids. The dilatation first appeared in the distal portion of the femur after 12 hours and by 24 hours extended over the entire medullary cavity. As the haematopoietic tissue decreased it was replaced by an increase in fatty tissue. After 16 days fatty tissue filled out the distal portion and by one month and for the rest of the observation period, the entire medullary cavity. Degenerative changes and haemorrhages were relatively slight. Oedema was commonly encountered and gave the bone marrow a gelatinous appearance. Haemosiderin was abundant.

There were no discernible shifts in the relative proportions between the granulocytic series and the erythroblastic series in the proximal femur and the diaphysis. In the small foci of regene-

ration about the blood vessels and along the endosteum in the distal regions the granulocytic series seemed to dominate between the second and fifth months.

Megakaryocytes were relatively radio-resistent. After 2 days cells in the distal marrow began to develop granular acidophilic patches in the otherwise normally basophilic cytoplasm. There was no demonstrable reduction in the number of megakaryocytes before 4 days. When it did occur, the reduction began and was most pronounced distally and did not involve the diaphysis before 8 days. Except during the second month, megakaryocytes were practically totally absent distally up to the fifth month. A great decrease in the number of megakaryocytes in the diaphysis was first evident after 6 to 7 months and by the ninth month they had disappeared from this region. By 10 months after injection the medullary cavity of most femurs was occupied by tumour tissue and persisting islands of bone marrow were usually aplastic.

Qualitatively, the changes noted in the marrow of the humerus, tibia and vertebrae followed the pattern of those in the femur. Cell depletion was greater in the femur and to some extent in the tibia than in the other bones which seldom attained complete aplasia of the marrow. Within the vertebral column cell depletion was greater in the lumbar than in the thoracic region. As was the case for the femur, bone marrow damage was initially most severe in the regions where Sr^{90} accumulation was greatest, i.e. close to the proximal epiphyseal plate in the humerus and tibia. With time, regeneration occured in the metaphysis while damage became more severe in the diaphyseal regions. Even the marrow of the vertebrae followed much the same pattern. Three to 4 months after injection, marrow hypoplasia in the humerus and tibia was especially apparent in a band about 2 mm. broad and located about 1 or 2 mm. distal to the epiphyseal plate (Fig. 4). This area of hypoplasia contrasted sharply with the often highly cellular regenerating marrow immediately adjacent to the epiphyseal plate.

Changes in the peripheral blood

An increase in the total leukocyte count occurred 6 hours after the injection of Sr^{90}. This increase was highly significant ($P < 0.001$) and represented an increase of all cell types without their relative proportions being affected to any extent. By 24

Table I.

Total number of leukocytes and differential counts expressed in absolute number per mm.³ blood. (n for each group = 10).

Time after injection of Sr^{90}	Injected				
	Total number of leukocytes Mean ± SE	Heterophils Mean ± SE	Lymphocytes Mean ± SE	Eosinophils Mean ± SE	Monocytes Mean ± SE
6 hours	***11360 ± 680	3045 ± 181.7	7625 ± 454.9	227 ± 13.6	489 ± 29.2
12 hours	6790 ± 420	1582 ± 98.8	4848 ± 302.7	190 ± 11.9	177 ± 11.0
24 hours	**5940 ± 320	2108 ± 133.3	2723 ± 172.3	77 ± 4.5	166 ± 10.5
2 days	***3880 ± 300	1214 ± 93.6	2447 ± 188.7	85 ± 6.6	132 ± 10.2
4 days	***2300 ± 200	490 ± 44.1	1757 ± 158.2	12 ± 1.0	41 ± 3.7
8 days	***2110 ± 110	346 ± 18.7	1712 ± 92.6	0 ± 0.0	51 ± 2.7
16 days	***1300 ± 150	39 ± 4.4	1238 ± 140.9	1 ± 0.2	22 ± 2.5
1 month	***1710 ± 124	530 ± 38.4	1060 ± 76.9	55 ± 4.0	65 ± 4.7
2 months	***2623 ± 772	753 ± 221.6	1692 ± 497.4	84 ± 24.7	94 ± 27.8
3 months	***1851 ± 216	740 ± 86.4	1016 ± 118.6	49 ± 5.8	44 ± 5.2
4 months	***2720 ± 450	979 ± 220.4	1659 ± 405.0	30 ± 10.4	52 ± 14.3
5 months	***2992 ± 213	1125 ± 80.1	1693 ± 120.6	108 ± 7.7	63 ± 4.5
6 months	***2708 ± 194	1145 ± 82.1	1416 ± 101.5	87 ± 6.2	62 ± 4.5
7 months	***2194 ± 169	941 ± 72.5	1174 ± 90.4	44 ± 3.4	39 ± 3.0
8 months	***1930 ± 145	872 ± 65.5	996 ± 74.8	21 ± 1.6	41 ± 3.1
9 months	**3010 ± 545	1800 ± 325.9	1141 ± 206.6	48 ± 8.7	27 ± 4.9
10 months	***1240 ± 159	605 ± 77.6	623 ± 79.8	6 ± 0.7	7 ± 0.9

Time after injection of Sr^{90}	Control				
	Total number of leukocytes Mean ± SE	Heterophils Mean ± SE	Lymphocytes Mean ± SE	Eosinophils Mean ± SE	Monocytes Mean ± SE
6 hours	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
12 hours	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
24 hours	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
2 days	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
4 days	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
8 days	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
16 days	7370 ± 510	1982 ± 136.4	4973 ± 342.2	147 ± 10.1	265 ± 18.3
1 month	9300 ± 109	1654 ± 19.7	6815 ± 83.4	279 ± 3.3	214 ± 2.5
2 months	7782 ± 396	1533 ± 78.0	5603 ± 285.1	335 ± 17.0	327 ± 16.6
3 months	8556 ± 778	1615 ± 150.2	6195 ± 563.3	368 ± 33.5	359 ± 32.7
4 months	8906 ± 670	1452 ± 107.6	7125 ± 528.0	303 ± 22.4	223 ± 16.5
5 months	9962 ± 563	2122 ± 119.9	7093 ± 400.9	488 ± 27.6	279 ± 15.8
6 months	8968 ± 149	2179 ± 36.2	6161 ± 102.4	395 ± 6.6	269 ± 4.5
7 months	8372 ± 474	1926 ± 109.0	5793 ± 328.0	234 ± 13.3	419 ± 23.7
8 months	8210 ± 530	2833 ± 182.9	4779 ± 308.5	328 ± 21.2	271 ± 17.5
9 months	5080 ± 409	1452 ± 117.0	3378 ± 272.0	163 ± 13.1	86 ± 7.0
10 months	5110 ± 367	1564 ± 112.3	3398 ± 244.1	102 ± 7.3	56 ± 4.0

Statistical analysis according to Student's t-test: total number of leukocytes in control group versus injected group.

** = $0.01 > p > 0.001$.

*** = $0.001 > p$.

hours, however, this leukocytosis gave way to a leukopenia, mainly because of a great drop in the number of lymphocytes (Table I). With few exceptions, this decrease in leukocyte count was highly significant ($P < 0.001$) for all leukocytes from the second day and throughout the entire observation period. The lowest value, 1300 cells (Table I), was recorded after 16 days at the time when the lowest absolute values for heterophils were also obtained. With small variations, there was a gradual increase in the heterophils up to 9 months and then a sharp decrease. The lymphocyte count decreased up to one month after injection and then remained on the whole fairly stable until the ninth month only to decrease greatly. Eosinophils reached their lowest values between 8 and 16 days then increased slightly only to decrease steadily after the sixth month. The monocytes decreased up to day 16, increased slightly until the second month and then decreased again.

Haemoglobin values had decreased by one to 2 days after the injection of Sr^{90} (Fig. 5). There was a steep drop until day 16 and then normal values were more or less regained after about a month. This increase continued during the second month and then the haemoglobin level, apart from small fluctuations, was much the same as for the control mice.

There was an initial increase in the thrombocyte count (Table II) and by 6 hours the count was significantly higher ($t = 3.635$) than for the control mice. Then a rapid decrease occurred and

Table II.

Thrombocytes, number per $mm.^3$ blood.
(n for each group = 10).

Time after injection of Sr^{90}	Control Mean ± SE	Injected Mean ± SE
0 hour	144400 ± 27500	—
6 hours		284000 ± 26900
12 hours		242700 ± 45900
24 hours		121800 ± 25500
2 days		65700 ± 8400
4 days		58600 ± 4900
8 days		71100 ± 23500
16 days		146000 ± 24300
1 month	152400 ± 25700	135200 ± 9200
2—10 months	206700 ± 15150	218700 ± 22263

by 4 days the thrombocyte count for the Sr^{90} mice was significantly lower. A rapid increase was evident from day 16 and throughout the remainder of the experimental period there were no significant differences between the groups.

Changes in the spleen

Changes in the weight of the spleen and differences between the Sr^{90} mice and the control mice are illustrated in Fig. 6.

At first there was a highly significant decrease in weight ($P < 0.001$) between 2 and 4 days after the injection of Sr^{90}. The weight of the spleen returned to normal between 8 and 16 days and then after one month increased greatly as hyperplasia occurred. Except for the second month, the significant increase in spleen weight persisted until the fourth month and then the weight of the spleen decreased slowly and fairly uniformly.

Cellular degeneration, particularly in the germinal centres, was evident by 12 hours after the injection of Sr^{90}. These changes were less severe at 24 hours and by this time practically all lymphocytes in the red pulp had disappeared. By 4 days erythropoiesis was apparent in the superficial subcapsular regions of the spleen and attained a degree greater than that seen normally. At the same time the follicles became less cellular about their periphery. By 8 days this appearance was more pronounced and the reticular framework of the follicles became quite visible. A large number of erythroblasts and other immature cell forms were emmeshed in the framework of the follicles. After 16 days the lymphatic tissue was still sparsely cellular and proliferation was still more evident with abundant erythroblasts, megakaryoblasts and other immature cells (Figs. 7, 8). The heavy erythro- and megakaryopoiesis and even some granulocytopoiesis was still evident 6 months after the injection of Sr^{90}. Regenerative activity in the white pulp was first manifest after 4 months.

DISCUSSION

There are no fundamental differences in the pattern of changes in the blood and haematopoietic tissues produced by external or internal irradiation. Those differences which can be discerned can be ascribed to the duration and distribution of radiation. External irradiation is usually of short duration and covers the whole body. Internal irradiation following upon the

injection of Sr^{90} is more protracted and through being concentrated within the skeleton particularly exposes the bone marrow. This makes it difficult to make direct comparisons between the damage occurring after the injection of Sr^{90} and that caused by external irradiation. Nor can the results of various Sr^{90} experiments be directly compared because of the wide variations in response of different animal species to Sr^{90}, means of administration, and amount of Sr^{90} administered.

In the femur, the site of marrow damage corresponded on the whole to the pattern of Sr^{90} accumulation (19). Initially the most severe changes appeared distally (Fig. 9) and to a lesser degree in the proximal femur just as Sr^{90} accumulated particularly strongly in these regions. The marrow damage persisted a long time in these regions. The signs of regeneration which appeared here after 5 or 6 months at the same time as the diaphyseal marrow became hypoplastic (Fig. 10) reflect the redistribution of Sr^{90} with time. A similar pattern of regeneration could be traced in the humerus, tibia, and vertebrae and in these bones as well, corresponded to the redistribution of Sr^{90}. In experiments with Sr^{89} on mice, *Bloom* (2) observed that regeneration usually began in the diaphysis and then appeared distally and proximally. The choice of isotope may account for this difference. Sr^{89} has a half-life of only 53 days which means that the residual radiation after 5 or 6 months is much less from Sr^{89} than from Sr^{90}.

The degree of marrow damage varied for different bones. Cell depletion was generally much greater in the femur and tibia than in the humerus and vertebrae. On the assumption that the bone marrow in various parts of the skeleton is uniformly radiosensitive, there seems to be some congruency between bone marrow damage and sites of tumour formation. Several factors, of course, probably influence the site of tumour formation (19). Many more tumours arise in the femur and tibia than in the humerus (18) and the total number of tumours in the 4 most commonly involved vertebrae is much lower than the number of tumours in the femur and tibia together. The size of particular bones and the volume of the medullary cavity may afford some explanation for the differences in the degree of marrow damage in different bones.

Within the femur there were regional variations in the pattern of cell depletion at various periods after the injection of Sr^{90}. At 16 days, cell depletion in the distal femur was as severe as that

in the diaphysis after 7 months. Megakaryocytes disappeared distally after 16 days but only after 9 months from the diaphysis. If *Finkel's* (6) calculations for total accumulated radiation are applied for the amount of Sr^{90} used in these experiments, then by 16 days the accumulated radiation in the metaphysis would be about 5000 rad. If the dose rate after the intravenous injection of 1 μC Sr^{90} per g. body weight is measured for a 10 μ cell in the femoral metaphysis it would decrease from 550 rad per day initially to 180 rad per day after 10 days (according to Finkel). It is quite conceivable that the initial rate of dose, as has been claimed by *Finkel* (6), *Owen* (21) and *Vaughan* (27), may be decisive for the occurrence of tumours. The initial damage to the bone marrow can then be taken as an indicator of the intensity of radiation in different regions of the skeleton. With this knowledge it should be possible to demonstrate the early histological changes in bone which herald the genesis of Sr^{90}-induced tumours.

Unlike the severe initial bone marrow damage from external irradiation (4), that resulting from the dose of Sr^{90} used in these experiments was slower in onset, and aplasia of the marrow did not occur before 16 days. Throughout the course of the experiments, frank degenerative changes were never more than relatively slight, probably because the cell damage that did occur was spread over a longer period of time, and in this respect differed from the more severe changes produced by external irradiation in large doses (2, 7, 11).

The increased marrow cellularity which became evident after 2 months (Fig. 4) is difficult to explain satisfactorily. At this time the Sr^{90} accumulation is high throughout the entire femur. From this point onwards there was some increase in the leukocyte count in the peripheral blood and the anaemia was gradually overcome. Extramedullary granulocytopoiesis in the spleen was now slight so that this increase in circulating blood cells undoubtedly reflected an increase in marrow activity, especially since this temporary regeneration was also seen in the tibia, humerus and vertebrae. This is probably an expression of a delayed abortive regeneration, much as has been described 4 to 11 days after external irradiation. *Jacobson* (13) considers this regeneration to reflect an increase in cells which were damaged by external irradiation but which could survive long enough to go through a limited number of divisions.

In some instances the marrow, instead of becoming aplastic, had a normal or even increased cellularity. One such example was seen in each group of Sr^{90} mice examined after 2, 4, 5 and 6 months. The marrow hyperplasia seemed to be generalised; a similar appearance was seen in all parts of the skeleton examined. The total leukocyte counts for these animals ranged between 6400 and 8680 and deviated significantly from the mean values for the respective groups. In 2 mice there was a lymphocytosis, in one a heterophilic granulocytosis, and in one the differential count was normal. The haemoglobin values for these animals fell within normal limits. All four mice had a significantly lower spleen weight than the other members of their groups. These deviations from the usual pattern of reaction may be associated with the capacity of Sr^{90} to induce leukaemia in some mice, in 5 per cent of another series (18).

The initial leukocytosis which appeared between 6 and 12 hours after the injection of Sr^{90} has not been reported earlier in conjunction with Sr^{90} but has been observed following external irradiation (3, 8, 11, 13 and others). The leukocytosis after external irradiation, according to *Harris* (8) and *Hulse* (11), is neutrophilic and reflects the mobilisation of neutrophils from the bone marrow. In these experiments there was no great increase in the heterophils in relation to the lymphocytes, probably because lymphocyte depletion did not occur as rapidly with the dose of Sr^{90} used here as after external irradiation in large doses. *Hulse* (10) has interpreted the early and rapid lymphocyte depletion after external irradiation as the result of the killing of these cells. According to *Hulse* (10) the neutrophilic granulocytosis increases with dose and is very strong at 5000 r, in part at least this probably depends upon the much greater radiosensitivity of the lymphocytes (26). Only after the initial peak had regressed somewhat at 24 hours was there a drop in the number of lymphocytes, evidence that the radiation dose was not sufficiently great initially to cause an immediate reduction in the lymphocyte count.

The great increase in spleen weight (Fig. 6) can largely be explained by the strong erythropoiesis in this organ, a point which has been made by *Jacobson* (13). The haematopoietic reserve offered by the spleen is one reason why the mice could survive since *Jacobson* (13) has shown that splenectomised mice die. Hyperplasia of the spleen occurs more rapidly and reaches a

greater extent (Fig. 6) after the intravenous injection of Sr^{90} than after intraperitoneal injection. Even although absorption is rapid, the Sr^{90} deposited in the abdominal cavity is apparently sufficient to produce a temporary depression of splenic function.

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SUMMARY

The effects of radiostrontium on the blood and haematopoietic tissues has been studied on 200 CBA mice injected intraperitoneally with 0.67 μC Sr^{90} per g. body weight. Groups of 10 mice given Sr^{90} and 10 control mice were killed at fixed intervals after the injection of Sr^{90}. Total leukocyte counts, differential counts, thrombocyte counts, and haemoglobin determinations were made. The spleen and bone marrow in the femur, tibia, humerus, and thoracic and lumbar vertebrae were examined histologically. Bone marrow damage — severe hypoplasia, dilatation of sinusoids, and an increase in fatty tissue — were observed in all bones examined. Damage was more severe in the femur and tibia than in the humerus and vertebrae. The site of marrow damage corresponded fairly well with the distribution pattern for Sr^{90} and the temporal changes in this pattern. Shortly after the injection of Sr^{90}, damage was most severe in the distal metaphyseal region of the femur and in the proximal metaphysis of the tibia and humerus. Later, the most severe changes were in the diaphyses of the long bones and regeneration gradually occurred proximally and distally in the long bones immediately adjacent to the epiphyseal plates. Close to the areas of regeneration, marrow damage was usually more persistent and more severe than in other regions with the exception of the initial damage in the metaphysis. Most tumours arise in this site.

There was an initial and significant leukocytosis which, by 24 hours, gave way to a leukopenia. Leukopenia then persisted throughout the experimental period (10 months). Haemoglobin levels fell up to the 16th day but from the second month onwards hovered about normal values. The drop in thrombocyte counts was not accompanied by a propensity to haemorrhage. The spleen increased greatly in weight, principally because of great increase in erythro- and megakaryocytopoiesis.

ZUSAMMENFASSUNG

Der Effekt von Radiostrontium auf Blut und blutbildende Gewebe bei der Maus.

Der Effekt von Radiostrontium auf Blut und blutbildendes Gewebe wurde an 200 CBA-Mäusen studiert, dit mit 0,67 μC Sr^{90} per g. Körpergewicht behandelt waren. In Zehnergruppen wurden die behandelten Mäuse sowie die Kontrollmäuse nach einem bestimmten Zeitintervall getötet. Blutausstrich, Rechnung von Leukozyten und Thrombozyten sowie die Bestimmung des Hämoglobingehalts wurden am Blut des venösen Sinus des medialen Augenwinkels ausgeführt. Milz sowie Knochenmark von Femur, Tibia, Humerus und Lendenwirbel wurden histologisch untersucht. Knochenmarkschäden in Form von mehr oder weniger hochgradiger Markhypoplasie, Sinusoiddilatation sowie Vermehrung von Fettgewebe wurden in sämtlichen untersuchten Knochen nachgewiesen. Am Femur und and der Tibia waren die Schäden deutlicher als am Humerus, and den Wirbeln sowie am Schädeldach. Die

Lokalisation des Knochenmarkschadens stimmte mit der, mit der Zeit auftretenden Umlokalisation von Sr^{90} überein. Demgemäss ist der Schaden, kurze Zeit nach der Injektion, am deutlichsten an den distalen Metaphysregionen des Femur sowie an den proximalen Metaphysen der Tibia und des Humerus. Lange Zeit nach der Injektion dagegen sieht man die hochgradigsten Veränderungen in den Diaphysen der langen Röhrenknochen, wobei eine allmählich eintretende Regeneration proximal und distal unmittelbar unter der Epiphysenplatte der Röhrenknochen beobachtet werden kann. In einem Bereich von 1—2 mm von diesen regenerativen Regionen ist der Knochenmarkschaden gewöhnlich persistent und deutlicher als in den übrigen Teilen, mit Ausnahme der initialen Schäden der Metaphyse. Von diesem Gebiet scheinen auch die meisten Tumoren auszugehen.

Im periferen Blut sieht man anfangs eine signifikante Leukozytenvermehrung. Die Leukozytose geht schon nach 24 Std. in eine Leukopenie über, die dann bis zum 10. Monat persistent ist. Die Hämoglobinkonzentration nimmt bis zum 16. Tage ab, aber vom 2. Monat an nach der Injektion sieht man, mit Ausnahme kleinerer Abweichungen, ein relativ stabiles Hämoglobinniveau nahe des Kontrollwertes. Die Thrombozytenwerte fallen nie so ab, dass eine erhöhte Blutungstendenz beobachtet werden kann. Milzveränderungen charakterisieren sich durch grosse Gewichtszunahme, vor allem durch eine starke Vermehrung der Erythro- und Megakaryozytopoese bedingt.

SAMMANFATTNING

Effekten av radiostrontium på blod och blodbildande vävnad hos mus.

Effekten av radiostrontium på blod och blodbildande vävnad har studerats hos 200 CBA möss, vilka behandlats med 0,67 μC Sr^{90} per gram kroppsvikt. De behandlade mössen samt kontrollmöss ha avlivats efter bestämda tidsintervall i grupper om 10 djur i vardera. Differentialräkning, räkning av leukocyter och trombocyter samt bestämning av hämoglobinhalten har utförts i blod uttaget från mediala ögonvinkelns venösa sinus. Mjälten samt benmärgen i femur, tibia, humerus samt ländkotor har undersökts histologiskt. Benmärgsskador i form av en mer eller mindre höggradig märghypoplasi, sinusoiddilatation samt ökning av fettvävnaden har påvisats i samtliga undersökta ben. Skadorna äro mera markanta i femur och tibia än i humerus, kotor samt skalltak. Benmärgsskadans lokalisation överensstämmer väl med den med tiden skeende omlokalisationen av Sr^{90}. Korta tider efter Sr^{90}-injektionen är sålunda skadan mest framträdande i distala metafysregionen i femur samt i proximala metafysen i tibia och humerus. Långa tider efter injektionen ses däremot de mest höggradiga förändringarna i de långa rörbenens diafyser, då en successivt inträdande regeneration kan iakttagas i rörbenen proximalt och distalt omedelbart under epifysplattan. I ett område 1—2 mm från dessa regenerativa partier, är benmärgsskadan vanligen persistent och mera framträdande än i övriga områden med undantag för de initiala skadorna i metafysen. Från detta område synes även flertalet tumörer utgå.

I det perifera blodet ses initialt en signifikant leukocytökning. Leukocytosen förbytes redan efter 24 timmar i en leukopeni, vilken sedan är persistent fram till 10:e månaden. Hämoglobinkoncentrationen minskar fram t. o. m. 16:e dygnet men fr. o. m. 2:a månaden efter injektionen ses med undantag för smärre fluktuationer en relativt stabil hämoglobinnivå i närheten av kontrollvärdet. Trombocytvärdena faller aldrig så lågt att någon ökad blödningstendens kan iakttagas. Mjältförändringarna karakteriseras av en stark viktökning framför allt beroende på en mycket stark ökning av erytro- och megakaryocytopoesen.

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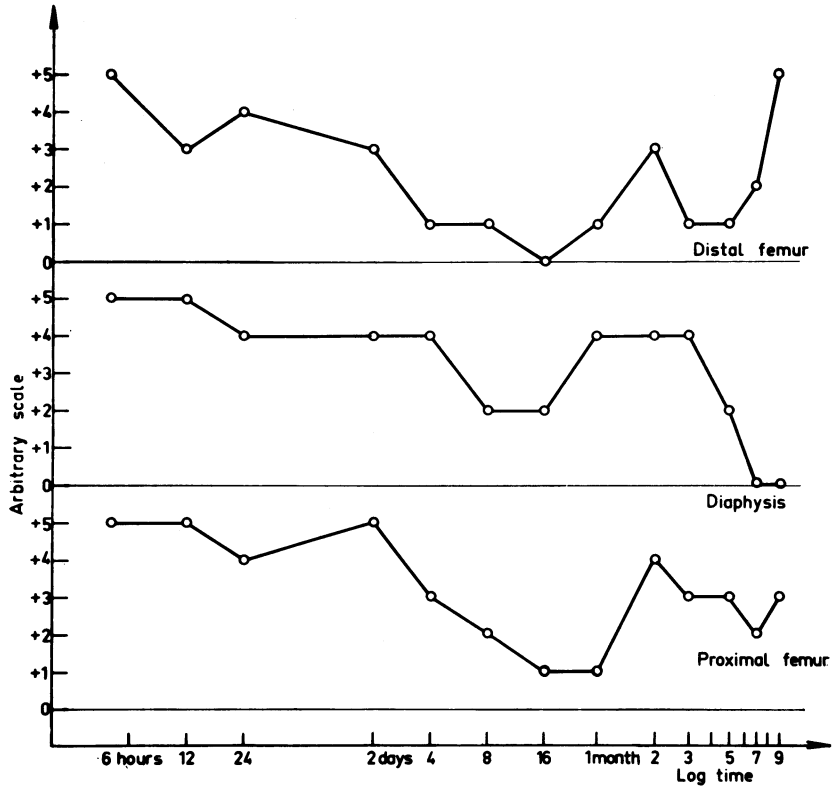
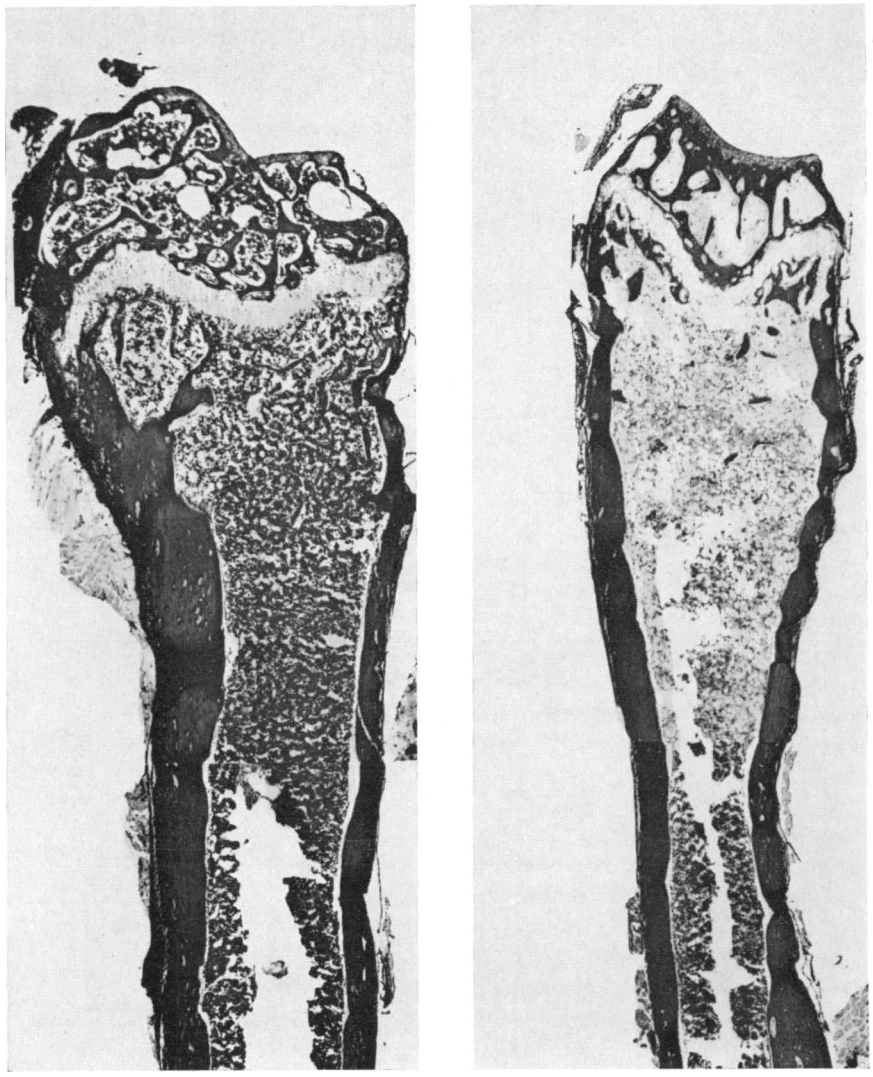


Fig. 1. Cellularity of femoral bone marrow of mice after the intraperitoneal injection of Sr^{90}, 0.67 μC/g. body weight. Marrow cellularity is graded from + 5 (normal) to 0 (aplastic marrow). Compare with Fig. 3.

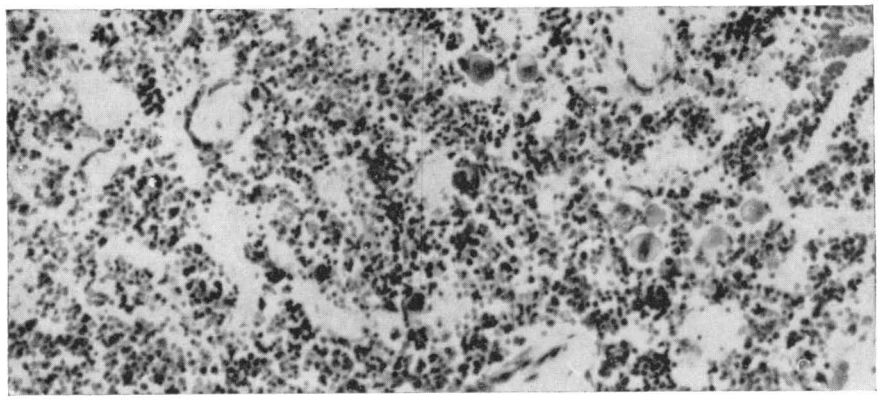


A B

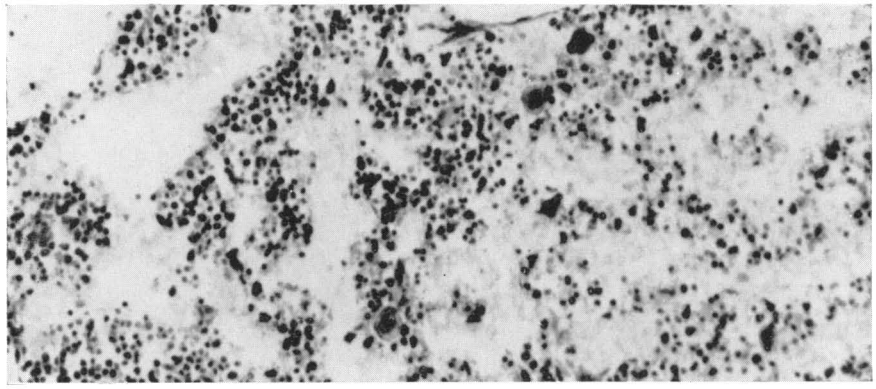
Fig. 2 A. Distal end and part of diaphysis of a normal mouse femur.

Fig. 2 B. Corresponding section, 16 days after injection of Sr^{90}, showing aplasia of the distal marrow. (van Gieson × 20).

A



B



C

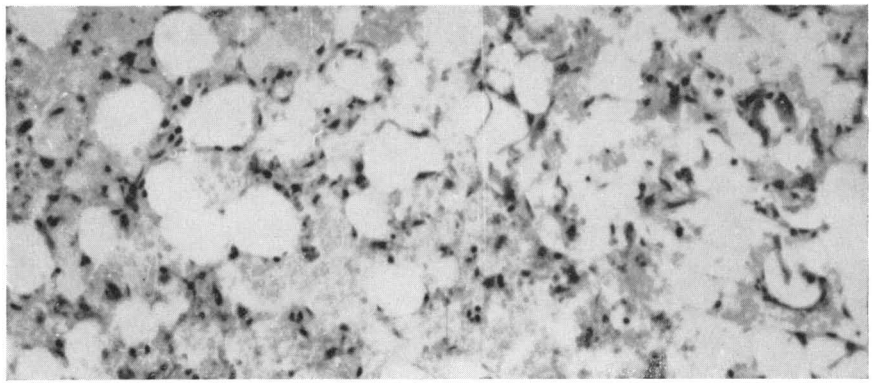


Fig. 3. Changes in bone marrow cellularity following the injection of Sr^{90}. The sections illustrate the grades used in Fig. 1.

A. Normal marrow, + 5.
B. Marrow 48 hours after Sr^{90}-injection, + 3.
C. Marrow 16 days after Sr^{90}-injection, 0

(Azure-eosinate, × 165).

Fig. 4. Humerus 5 months after Sr^{90}-injection showing regeneration of marrow just below epiphyseal plate. Note the characteristic location of the aplastic area in upper 1/4 of the diaphysis. (Haematoxylin-eosin × 40).

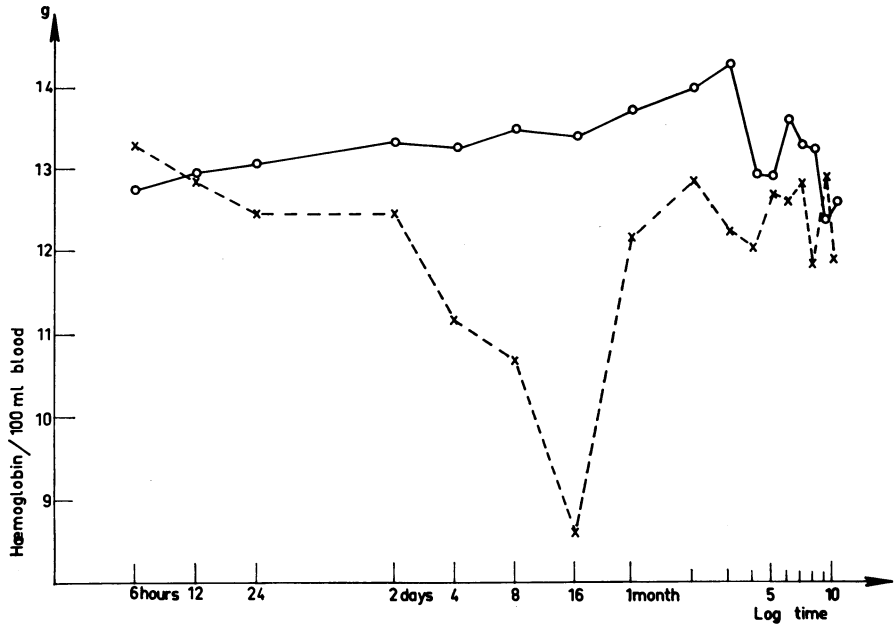


Fig. 5. Haemoglobin levels (g. per 100 ml. blood) after the intraperitoneal injection of Sr^{90}. Each value represents the mean for 10 mice. Broken line, Sr^{90}-injected mice; solid line, control mice.

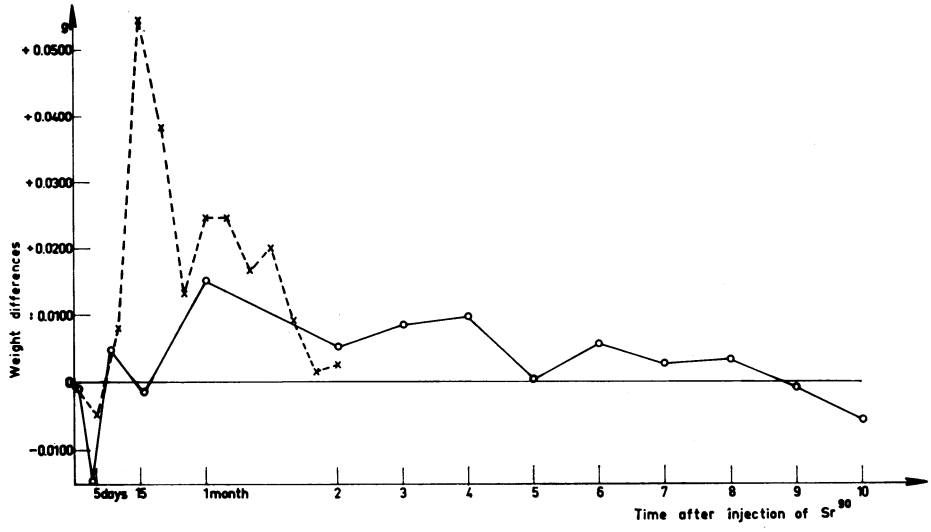


Fig. 6. Difference in spleen weights between Sr^{90}-injected mice and control mice. The reference line represents mean spleen weight for groups of 5 control mice, the solid line the mean weight for groups of 10 mice injected intraperitoneally, and the broken line the mean weight for groups of 10 mice injected intravenously.

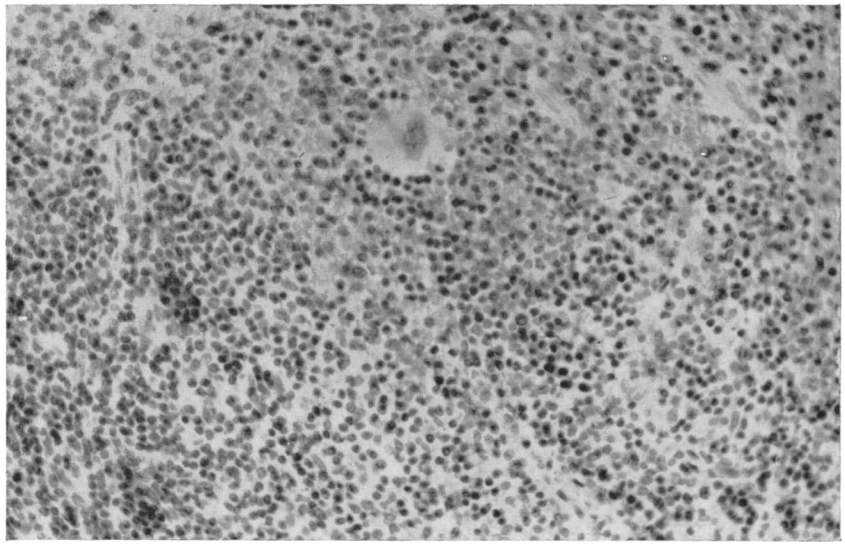


Fig. 7. Normal spleen of a 90-day-old CBA-mouse (haematoxylin-eosin, × 250).

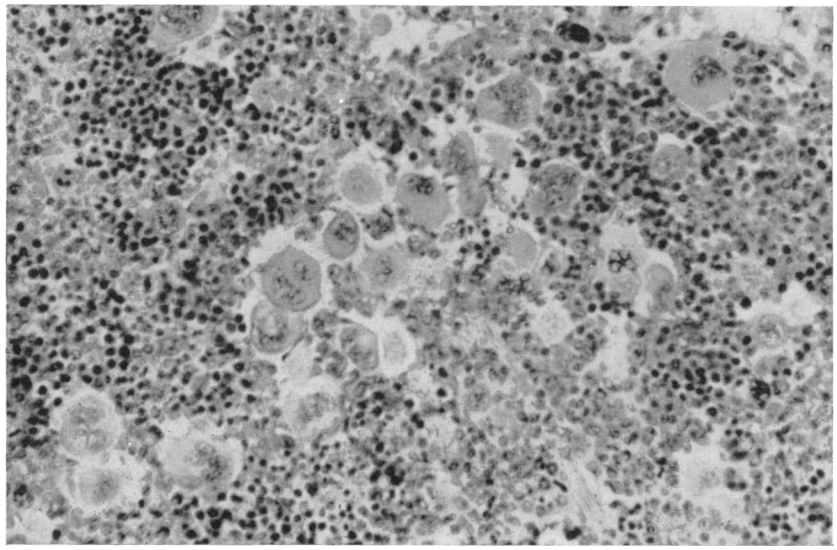


Fig. 8. Mouse spleen 3 months after injection of Sr^{90} showing erythropoiesis and a great number of megakaryocytes (haematoxylin-eosin, × 250).

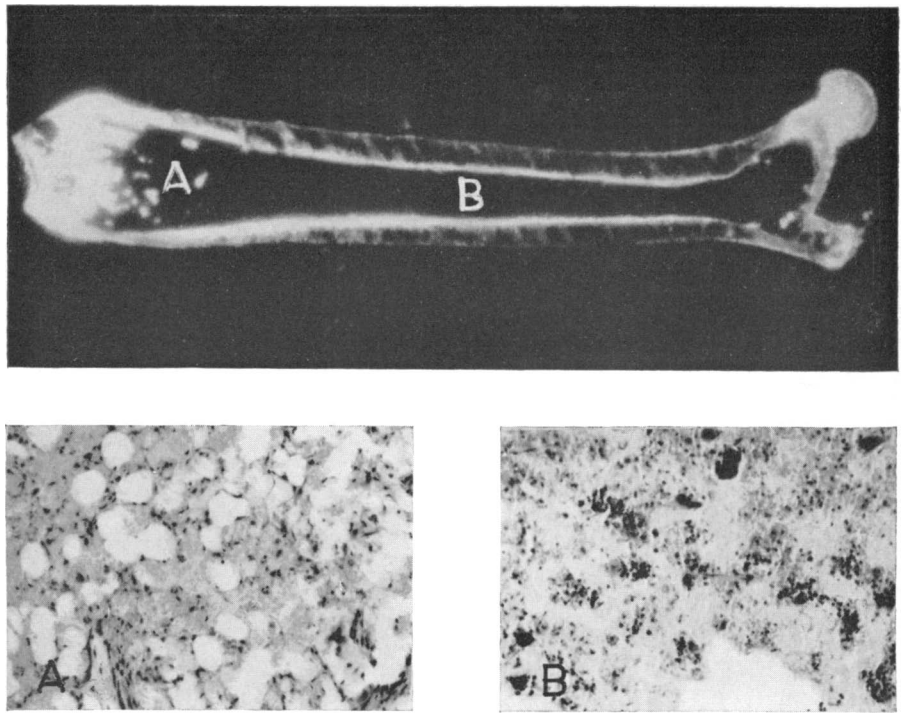


Fig. 9. Autoradiogram of a plastic-embedded femur of a mouse 24 hours after Sr^{90}-injection. High uptake in the distal part of the bone. In addition to the strong accumulation of Sr^{90} in this region, the cellularity of the bone marrow shortly after Sr^{90}-injection is much more reduced in this area (A) than in the diaphysis (B). Increase of fatty tissue at A and islands of active marrow persisting at B. Microphotos A and B, 16 days after isotope administration, (azure-eosinate, × 85). Autoradiogram taken from the series described in reference 19.

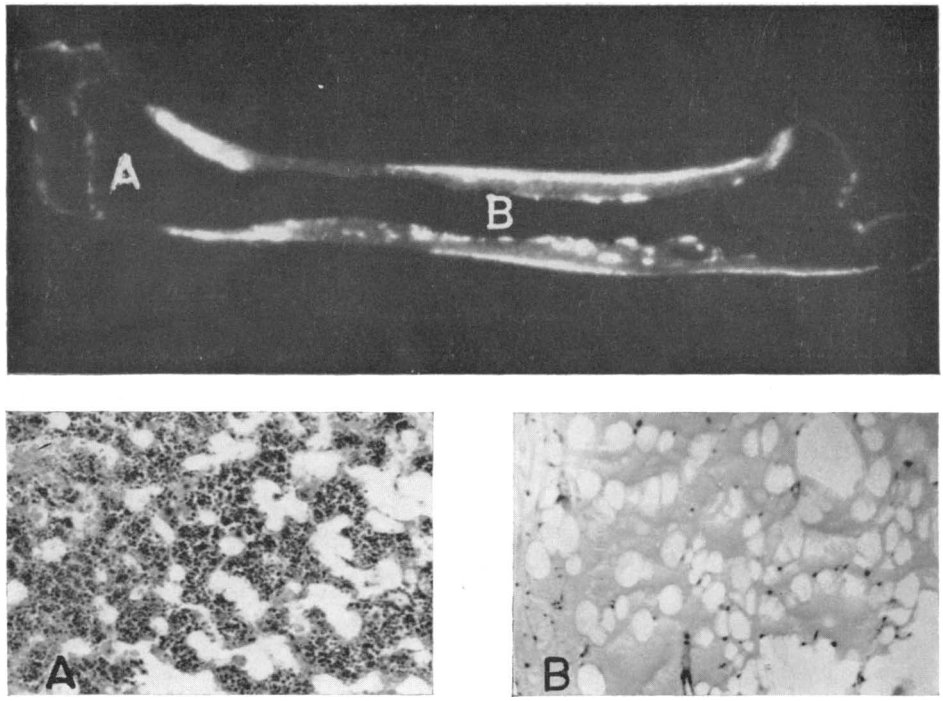


Fig. 10. Autoradiogram of a plastic-embedded femur of a mouse 9 months after Sr^{90}-administration. Disappearance of Sr^{90} from the distal and proximal regions and much stronger retention of isotope in diaphysis. Microphotos A and B illustrate state of bone marrow 9 months after Sr^{90}-injection. Regeneration of marrow and persistence of fatty tissue at A and aplastic, fatty and oedematous marrow at B (azure-eosinate × 85). Autoradiogram taken from the series described in reference 19.