THE DEVELOPMENT OF BONE SARCOMA IN RATS INJECTED WITH STRONTIUM-89 AND THE EFFECT OF STRONTIUM-89 ON THE OFFSPRING OF SUCH RATS

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The study of the effect of radioactive isotopes on living animals and, in particular, on the development of bone tumors, and also the consequences of this effect are problems of great importance at the present time. The literature of recent years dealing with the study of the bone changes and the formation of bone tumors by the action of radioactive isotopes contains only a limited number of reports, and some of these are concerned with the study of strontium-90.

Our investigations are devoted to the study of the action of strontium-89, differing essentially from strontium-90 by its half-life period: Strontium-89 has a short half-life period of 54.5 days, whereas strontium-90 is characterized by a comparatively long half-life period, lasting 19.5 years.

The data in the literature on this subject includes changes in bone tissue under the influence of the "bone" isotopes during both the acute and chronic periods [1-14]. All the authors cited conclude that strontium, when it enters the body, is deposited in the bones, where a process of pathological reorganization of the tissue is initiated, as a result of which, after a certain time, sarcomas of bone develop. The number of sarcomas differed, according to the findings of these authors, and varied between 30 and 70%. These figures, however, relate mainly to strontium-90.

METHOD AND RESULTS

Strontium-89 was injected intraperitioneally into rats in aqueous solution, in a dose of 1 μC/g body weight.

For the experiments 156 rats were used; in 78 of these animals neoplasms of bone were observed, and in 60 of these the tumors were single and in 18 multiple. Altogether 102 tumors developed. Histological investigation showed that these were osteomas and osteogenic sarcomas. In addition we investigated the bones of the offspring of the rats which received injections of strontium-89 (in the first and second generation) for their strontium content.

The largest number of tumors was situated in the femur (42) and tibia (38). Significantly fewer neoplasms developed in other parts of the skeleton. In the humerus, for instance, only eight neoplasms were found; the same number of tumors was present in the bones of the vertebral column. There were five tumors in the bones of the pelvis and only one in the bones of the forearm.

The tumors appeared at the following times: Minimum - 157 days, maximum - 461 days. The highest mortality among the animals was observed in the seventh and eighth months (32 and 47 rats). In the same period (from the sixth to the ninth month), one of the signs of radiation sickness - depilation of the rats - was most pronounced.

A sharp decrease in the number of young in the litters was observed in many females, especially in the first six months after injection of strontium: Instead of 10-12 in each litter, there were only four or five; only later, one year after the injection of strontium, was the number of young rats in the litters once again raised to normal.

To two rats, 52 and 69 days after the beginning of the experiments, eight young rats were born in two litters. Six of these had developmental defects of the bony skeleton: In all of them the tail was hardly developed, and in two, in addition, a severe maldevelopment of the hindlimb was observed (Fig. 1).

Among the rats of the second and third generations, individuals were also encountered in which the tail was shortened, and in one animal in the second generation there was maldevelopment of the hindlimb. This particular rat was born 449 days after the start of the experiment.

Regarding the indications given in the literature concerning the deposition of strontium in the bones, we were interested in the question whether a connection existed between the localization of strontium in the bones and the site at which the tumors developed. In order to answer this question we used the method of autoradiography. Sections of bone from the experimental animals were placed on x-ray film and, after exposure for a period from 15 minutes to five days, the film was developed; as a rule radiation tracks could be seen on it, the intensity of which depended on the amount of strontium in the section. In the course of this work we tried various exposure times. In N. N. Litvinov's work [2, 3, 4] with strontium-90, the

Fig. 1. Rat with maldevelopment of the limb and tail (experimental treatment with Sr^{89}).

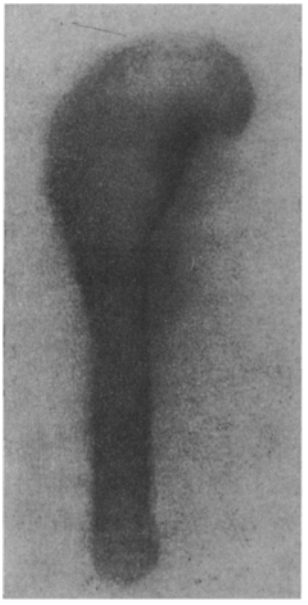

Fig. 2. Autoradiograph of the femur of a rat receiving Sr^{89}. Intensive blackening in the cortical layer of the diaphysis. Exposure 30 minutes.

most intensive impressions were obtained with an exposure lasting several days, and the maximum exposure reached 20 days; in our experiments the optimum time for production of a clear autoradiograph was only 30 minutes. According to our films, the strontium was localized as a rule in the cortical layer of the bones, in the region of the diaphysis, so that darkening of the x-ray film took place more intensively in the area corresponding to the diaphysis than in that corresponding to the metaphysis (Fig. 2). The majority of neoplasms arose in the diaphysis. This suggests that the foci of development of sarcomas coincide with the sites of greatest localization of strontium.

In addition to the bone sections of rats which had received injections of strontium, we also prepared a series of bone sections from young rats of the first generation and, by increasing the exposure times to 1-5 days, we obtained very clear autoradiographs of these sections. This indicated that the bones of the first generation of young rats also contained strontium. Tests with a β-radiation counter confirmed the activity of the young rats' bones and, consequently, the presence of strontium therein. This made it necessary to discover how the strontium passed from the mother to the young. Two ways may be postulated: The first – through the placenta, in the period of intrauterine development, and the second – through the milk. Theoretically, either of these methods of transmission of strontium is permissible, and it is noteworthy that in the literature there are indications of activity of the milk.

We took from the suckling rats receiving strontium-89 a small quantity (about 0.5 ml) of milk and tested its activity. Not one of three samples of milk showed any activity. A few drops of blood were taken from these same rats, and the activity determined. In every case the blood tests showed definite activity, although admittedly only very slight. Moreover, the very fact of the birth of rats with developmental defects of the bony skeleton itself suggests the placental route of transmission of strontium from mother to fetus during the period of intrauterine development. The absence of strontium from the milk in our observations may, to some extent, be accounted for by the considerable length of time elapsing from the start of the experiment to the day of the test (about 200 days). In order, however, to obtain final confirmation of the role of the placental route, we tested the activity of the femur in a newborn rat which had not yet begun to feed on its mother's milk. The result of the test was positive – activity was established, and the autoradiograph gave an obvious impression after an exposure of five days (Fig. 3).

These findings all go to prove that strontium-89 passes from mother to offspring via the placenta.

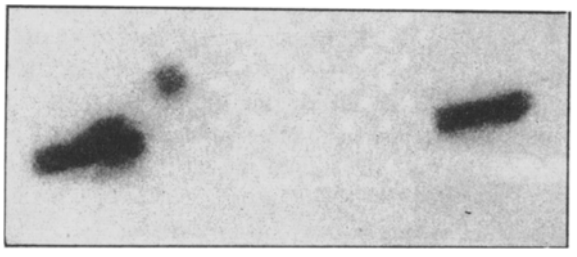

Fig. 3. Autoradiograph of the femora of newborn rats. Exposure 5 days.

During tests of activity, we found radiation in a living rat of the first generation, and also in the bones of the rats of both first and second generations. This fact suggests that the influence of radioactive substances may be spread not only to the animal receiving injections of strontium, but also to the offspring, and even to the second generation.

SUMMARY

Sr^{89} was administered intraperitoneally to rats. In 50% of those which survived for 180 days and longer bone sarcoma appeared (mainly in the femur and tibia), and some of them developed multiple tumors. The tumors were most commonly revealed 8-9 months after the beginning of the experiment. The mortality rate, as well as the loss of hair in the rats which developed no tumors, was greatest at the same periods of time (i.e., during the 8th-9th month). The progeny of rats treated with Sr^{89} (the first and the second generation) was usually born with skeletal anomalies; the autograms of these animals showed the presence of strontium. By checking the activity of milk and blood it was established that Sr^{89} passes from the mother to the progeny through the placenta.

LITERATURE CITED

[1] N. A. Kraevskii, in: The Biological Action of Radiation and Clinical Aspects of Radiation Sickness [in Russian] (Moscow, 1954) p. 170.

[2] N. N. Litvinov, Voprosy Onkol. 2, 3, 285 (1956).

[3] N. N. Litvinov, Arkh. Patol. No. 4, 81 (1956).

[4] N. N. Litvinov, Arkh. Patol. No. 1, 26, (1957).

[5] N. N. Petrov et al., The Appearance and Development of Malignant Growth in Experiments on Monkeys [in Russian] (Moscow, 1951).

[6] A. A. Rubanovskaya and V. F. Ushakova, in: The Toxicology of Radioactive Substances [in Russian] (Moscow, 1957) v. 1, p. 23.

[7] V. N. Strel'tsova and Yu. L. Moskalev, Med. Radiol. 2, 5, 39 (1957).

[8] L. A. Cherkasskii, Voprosy Onkol. 2, 3, 275 (1956).

[9] M. A. Bloom and W. Bloom, Arch. Path. 47, 494 (1949).

[10] F. J. Bryant A. C. Chamberlain, et al., J. Nuclear Energy, 6, 22 (1957).

[11] D. H. Copp, D. J. Axelrod, and J. G. Hamilton, Am. J. Roentgenol. 58, 10 (1947).

[12] J. Jowsey, et al., Brit. J. Exp. Path. 36, 22 (1955).

[13] P. A. Thornton, P. J. Shaible, and L. F. Wolterink, Poultry Sc. 35, 1055 (1956).

[14] M. Tutt, B. Kidman, et al., Brit. J. Exper. Path. 33, 207 (1952).