

## Absorption, Deposition and Excretion of Sr<sup>89</sup> by the Chick in Relation to Route of Administration.\*† (31760)

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In addition to the basic knowledge obtained concerning accretion rates and normal bone kinetics, the application of radiostrontium to the pathology and pathological physiology of the bone has made it extremely desirable to know the time at which the maximum amount of tracer isotope is in the critical tissues so that a minimum amount of isotope may be administered. This is especially true in the clinical localization of bone lesions such as fractures and the search for metastasizing cancer. For clinical purposes, Sr<sup>85</sup> is usually administered intravenously in a single dose and observations are made on blood-disappearance curves, excretion in urine and feces and external counting. However, of the two ways of excreting the isotope the major route will inherently depend upon the mode in which the isotope enters the body. Therefore, to maximize usable information it is necessary to know the influence of administration routes on excretion pattern, not only so that optimum retention or excretion may be accomplished, but also that the most efficient parameter, *e.g.*, urinary or fecal excretion, may be monitored.

Paul *et al*(1) observed that rats showed maximum activity of Sr<sup>89</sup> in the bones 6 hours after orogastric intubation and in the blood in less than 2 hours after administration.

Hansard(2) reported that in ruminants a greater amount of Sr<sup>89</sup> was excreted in the feces rather than in the urine when the isotope was administered either orally or intravenously. Pecher(3,4) found a greater excretion of Sr in feces than in urine of mice, but urinary excretion of Sr exceeded that of feces in rats during the first days following oral ingestion. Retention of Sr<sup>90</sup> in the monkey was found to be the least when administered orally and the greatest retention was

found when the isotope was administered by the intraperitoneal route(5). Durbin *et al*(6) showed that 56% of intravenously administered Sr<sup>90</sup> was excreted *via* the urine in the first 10 days after injected. Data do not, however, appear in the literature on the excretion routes of radiostrontium from the chick due to the difficulty in efficiently separating the urine and feces for metabolic studies.

*Materials and methods.* To determine the time necessary for maximum absorption and deposition of an oral dose of Sr<sup>89</sup>, 100 one-week-old male chicks were fasted for 8 hours and administered 6.12  $\mu$ C Sr<sup>89</sup> (Sr<sup>89</sup>Cl<sub>2</sub>, carrier free) in aqueous solution by orogastric intubation. Four birds were killed at 15-minute intervals from 15 minutes to 3 hours, then at 4, 5, 6, 8, 10, 12, 20, 24, 36, 48, 72, 96, and 144 hours post-administration. From each bird the eyes, left tibiotarsus, and a 5-ml blood sample were taken for Sr<sup>89</sup> analysis by the method of Creger *et al*(7), and the resulting data were calculated by the IBM computer program method of Creger *et al*(8).

To determine the effects of various routes of administration on the excretion pattern of Sr<sup>89</sup>, an artificial anus was surgically prepared in male chicks of approximately 8 weeks of age by the method of Colvin *et al* (9). Following convalescence from surgery the birds were administered 5  $\mu$ C Sr<sup>89</sup> orally, intramuscularly, intravenously, intraperitoneally, and subcutaneously, respectively. The urine and feces were then collected separately at 2, 4, 8, 12, 24, and 48 hours and analyzed for Sr<sup>89</sup> content. At 48 hours the birds were killed and the right tibiotarsus removed for Sr<sup>89</sup> analysis. The remainder of the carcass was also analyzed.

*Results and discussion.* The rate of uptake of Sr<sup>89</sup> by the blood, tibiotarsus, and the eyes is shown in Fig. 1. From Fig 1 it can be seen that the peak concentration (0.05% of

\* Supported in part by the Division of Radiological Health, USPHS Grant RH00354.

† Contribution of Texas Agri. Exp. Station.

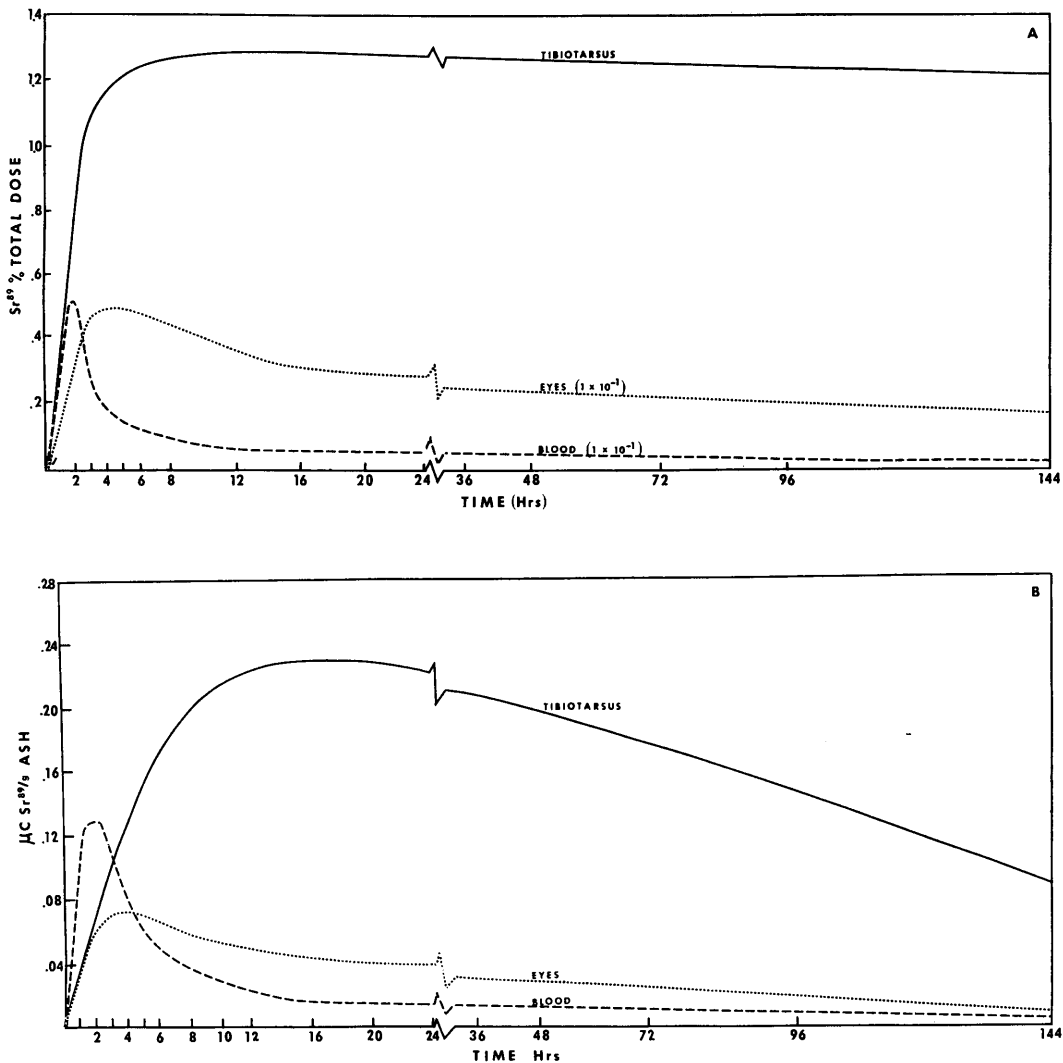


FIG. 1. Rate of deposition of Sr<sup>89</sup>. A. Sr<sup>89</sup>% of dose vs time. B. Specific activity ( $\mu\text{c Sr}^{89}/\text{g ash}$ ) vs time.

the dose) of Sr<sup>89</sup> was reached in the blood 2 hours following oral administration of the isotope and decreased quite rapidly as the radiostrontium was withdrawn from peripheral circulation. By 24 hours after the administration, the Sr<sup>89</sup> content of the blood has reached a negligible level.

It was noted that the rate of uptake of radiostrontium (% of dose) by the tibiotarsus was almost identical to that of the blood for the first 2 hours, implying that the Sr<sup>89</sup> was deposited in the bone as rapidly as it could be transported by the blood from the gut. However, after the maximum activ-

ity had been reached in the blood and the observed activity in the blood began to decrease, that of the tibiotarsus continued to increase until approximately 14 hours following administration of the radiostrontium. This indicates that the Sr<sup>89</sup> was being withdrawn from the blood and deposited to a large degree in the bone. The maximum activity observed in the tibiotarsus was 1.3% of the dose in the 48-hour period.

The eyes of the chick, due to the presence of the sclerotic ossicles, take up radiostrontium, but less rapidly than the tibiotarsus. The peak activity (0.04% of the dose) was

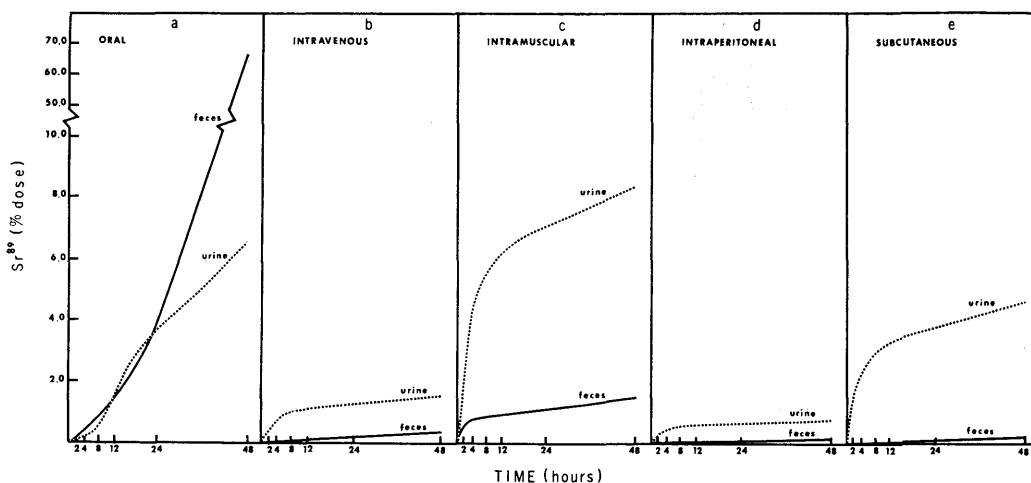


FIG. 2. Influence of administration route on excretion of Sr<sup>89</sup>. a. Oral; b. Intravenous; c. Intramuscular; d. Intraperitoneal; e. Subcutaneous.

reached in the eyes in approximately 4 hours and decreased at about the same rate as blood through 144 hours.

To show the relationship of Sr<sup>89</sup> to the total mineral content of the tissue examined, the specific activity in  $\mu\text{C Sr}^{89}/\text{g ash}$  is shown in Fig 1B. It can be seen that the specific activity of blood at 2 hours is quite high ( $0.1 \mu\text{C Sr}^{89}/\text{g ash}$ ) and that the Sr<sup>89</sup> in bone begins to be diluted by deposition of stable minerals almost as soon as a maximum value is reached.

The effects of various routes of administration on the excretion pattern of Sr<sup>89</sup> are shown in Fig. 2. From Fig. 2a it can be seen that the most rapid and efficient excretion of radiostrontium is accomplished in the chick, as in other species, when the radionuclide is administered orally. In a 48-hour period approximately two-thirds of the total dose of Sr<sup>89</sup> was excreted, and over 90% of the excreted isotope was voided *via* the feces. This observation, however, does not appear surprising considering the discrimination which is exhibited against Sr by the digestive tract of every species which has been studied (10-12).

Once the intestinal mucosa barrier was circumvented, all other routes of administration resulted in a higher recovery of radiostrontium from the urine than from the feces. The most efficient urinary excretion was observed when the isotope was administered

intramuscularly. Intramuscular injection resulted in excretion of 8.4% of the dose *via* the urine in 48 hours (Fig. 2) and only 1.5% by way of the feces. Total excretion following subcutaneous injection of Sr<sup>89</sup> (Fig. 2e) was only slightly more than one-half as efficient as that observed following intramuscular injection, but as in the latter, most of the radionuclide was excreted *via* the urine; 4.6% of the dose appeared in the urine compared to 0.2% of the dose in the feces.

Intravenous (Fig. 2b) and intraperitoneal (Fig. 2d) injections of Sr<sup>89</sup> resulted in excretion patterns similar to intramuscular and subcutaneous injection, but, as observed by Tuttle *et al*(5), the two former methods resulted in much greater retention of the isotope. Body retention of Sr<sup>89</sup> was above 90% as a result of intravenous and intraperitoneal administration; whereas, intramuscular injection resulted in only 77% retention. Maximum skeletal deposition occurred when the isotope was given intraperitoneally in which case there was 7.6% of the dose deposited in the tibiotarsus. Intravenous and intramuscular injection resulted in approximately 5% of the dose being deposited in the tibiotarsus and by subcutaneous administration there was 6% deposited. As expected from the very high excretion of the isotope when given orally and the low retention (29%) there was a smaller amount of Sr<sup>89</sup> deposited in the tibiotarsus (2.3%).

In all groups which were injected with Sr<sup>89</sup> there was a very rapid rate of excretion of the isotope in the first 8 hours after which the excretion rate decreases slightly. On the other hand the rate of excretion following oral administration of Sr<sup>89</sup> continued to increase at a rapid rate through 48 hours.

These data indicate that following the discrimination by the intestinal mucosa, the next great discriminatory barrier which Sr must overcome before skeletal deposition is that exhibited by the kidney.

*Summary.* The maximum concentration of Sr<sup>89</sup> observed in the blood following an oral dose was observed to occur in about 2 hours and decreased to a negligible quantity in 24 hours. The rate of uptake of Sr<sup>89</sup> by the bone was found to parallel the rate of increase in activity of the blood, but the former continued to increase to 14 hours post-injection time. The eyes took up radiostrontium in a similar way, but in much lower concentrations.

It was observed that the most efficient excretion was accomplished when the isotope was ingested orally, and the least efficient elimination was observed when the isotope was administered by intraperitoneal injection. All routes of administration, except the oral route, resulted in higher activity in the urine than in the feces with the highest urinary

excretion occurring as a result of intramuscular injection.

The authors wish to express their appreciation for the technical assistance of Mrs. Jennifer Newton.

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Received October 25, 1966. P.S.E.B.M., 1967, v124.

### Comparative Immunology. Active Immunization of Young Alligators with Hemocyanin.\* (31761)

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Recent immunological studies from a comparative or phylogenetic standpoint have shown this area to be a relatively unexplored region of immunology, and have provided new approaches and different perspectives on old problems as well as a source of fundamentally new information(1,2). The mechanisms of immunity of poikilothermic vertebrates are

of special interest because of the important role of environmental temperature(3). Furthermore, the crocodylian reptiles are believed by Good *et al*(4) to constitute a sequential step in the evolutionary development of the lymphoid system, in which the lymphoepithelial tissue found in the pharyngeal tissue of the alligator appears to be the evolutionary precursor of the tonsils of mammals. Although

\* Supported in part by USPHS Grant HE 06244.