Biochemical Pharmacology, 1964, Vol. 13, pp. 407–412. Pergamon Press Ltd., Printed in Great Britain.

A COMPARISON OF THE DISTRIBUTION OF SOME HALIDE IONS IN THE BODY

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(*Received* 5 *November* 1963; *accepted* 21 *November* 1963)

Abstract—The distribution of the halide ions F^-, Br^-, I^- and tentatively Cl^-, and the pseudo-halide ion SCN^- has been studied in mice and rats. Whole body autoradiography, microautoradiography, double tracer autoradiography, and scintillation counting methods have been used. Similarities and dissimilarities in the distribution pattern of the various ions are described and discussed. Although the chemical and physical properties are similar for the elements studied—with some notable exceptions for fluorine—their distribution patterns in the body show pronounced dissimilarities.

THE present paper constitutes a collection of results issued in a series of research works concerning the distribution of some halide ions in mice and rats. The ions studied have been fluoride,[1] chloride, bromide[2, 3], iodide,[4] and the so-called pseudo-halide thiocyanate.

In earlier papers it has repeatedly been shown that elements from the same group of the periodic system exhibit some similarities of distribution and metabolic behaviour. This is particularly pronounced for elements in group 2: Ca, Sr, Ba, and Ra. Besides the theoretical interest inherent in a comparison of elements with similar physical and chemical properties the elements studied here are also of significance from practical medical points of view.[1–5] Among these the inhibition of dental caries by the fluoride ion may be mentioned. Bromide and thiocyanate are often used as inert indicators for the closely related chloride ion in connection with clinical-physiological kinetic and disappearance investigations, and also in investigations of the extracellular space. Bromide is of pharmacological interest as a sedative and so is thiocyanate by its action on the thyroid metabolism. The dominating roles of iodide are its incorporation into thyroid hormone and, in this connection, its use in radiodiagnostics and radiotherapeutics as well as its significance as a fission product.

MATERIALS AND METHODS

The radioactive nuclides used have been ^{18}F, ^{38}Cl, ^{82}Br, ^{80m}Br, ^{80}Br, ^{131}I, ^{125}I, ^{11}C-CN and ^{35}S-SCN.

The investigations were mainly carried out on normal adult mice and rats. Some of the animals were pregnant; they were generally examined two or three days before expected parturition. The various radioactive nuclide compounds were dissolved in physiological saline solutions. The radioactive solutions were then injected as single

doses i.v. on the mice and i.p. on the rats. Owing to the radiation properties, the half-life of the various radioactive nuclides, and the length of the survival period of the experimental animals, the amount of activity injected varied between 500 and 15 μc, which corresponded to less than 10^{-4} μg of the element in question.

In the autoradiographic studies the animals were sacrificed by immersion in solutions of acetone and solid carbon dioxide (−80°) at intervals after the injection varying from 1 min to several days. In the experiments with ^{18}F, however, the survival periods were not longer than one hour because of the short half-life of this nuclide. Sagittal sections, 10–20 μ thick, were taken through the entire frozen animals at various levels by a sledge microtome. Sectioning and drying of the sections were carried out at about −12° in a freeze-room. Microautoradiographic studies were performed on some special organs and tissues; the gastric mucosa, bones, and teeth. The exposure times varied between 4 hr (^{18}F) and 195 days ($K^{14}CN$). The following autoradiographic emulsions were used: Gevaert Dentus Rapid, Kodak Industrex, and Ilford G5 and Q3. The autoradiographic techniques used have been described in detail previously, especially in the papers[6, 7] and also in the papers.[1–5, 9, 15]

Quantitative measurements were also made of the activity in the various organs in the fluoride, iodide, and bromide studies. The nuclide concentration was generally calculated as the amount of radioactive nuclide per gramme of organ per the amount activity injected per gramme of animal. The organs and tissues to be studied were rapidly excised from the animals after they had been killed by decapitation or by stretching the spine. The wet weight of the organs, tissues, and body fluids was used. Further details about the techniques and their errors have previously been given.[8, 16]

In the autoradiographic investigations the radiation properties of the isotopes used have influenced the possibilities of making detailed observations. Especially favourable conditions have been offered by those isotopes which emit extranuclear electrons (^{125}I, ^{80m}Br).[3, 8, 11]

Some few autoradiographic studies have also been performed with ^{38}Cl. This isotope was produced with thermal neutrons according to the Szilard–Chalmers method. The radioactive solution, as $^{38}NaCl$, was injected i.p. on a mouse. The half-life of ^{38}Cl is only about half an hour, hence only a very short survival time could be used. The autoradiograms obtained further showed a very unsatisfactory resolution owing to the hard β-radiation. The long lived ^{36}Cl has not yet been available in a specific activity high enough to permit autoradiographic investigations.

RESULTS

Blood

Of the ions studied fluoride disappeared most rapidly from the blood after i.v. injection, followed by iodide. The bromide and thiocyanate ions, on the other hand, showed rather high concentration in the blood even a long time after the injection of the radioactive solutions.

Blood vessel walls

Br^-, I^-, and SCN^- were found to accumulate in the blood vessel walls. This was especially evident in *aorta* and *arteria carotis*. The methods used have so far not demonstrated any fluoride accumulation in these tissues.

Central nervous system

All of the ions studied showed a very low concentration in the central nervous system. Of these Br^- showed the highest concentration; 4–24 hr after the injection the bromide concentration in the CNS was 1/3 of the blood level. Considerably more bromide was present in peripheral nerves than in the central nervous system. The foetal brain and spinal cord contained significantly more Br^- than did the corresponding maternal tissues.

Iodide and bromide showed a pronounced accumulation in the choroid plexus, while thiocyanate had only a low concentration in this organ and fluoride did not reach a higher level here than in the other parts of the central nervous system. The order of permeability of the ions into CNS seemed to be $Br^- > I^- > SCN^- > F^-$.

Pituitary gland

Bromide, thiocyanate, and iodide showed a somewhat higher concentration in the pituitary gland than in the central nervous system at corresponding survival periods of the animals. This difference could not be noticed for fluoride.[16]

Eye

^{131}I and ^{82}Br showed a rather high uptake in the various tissues and fluids of the eye. The uptake of thiocyanate was somewhat lower. The concentration of these ions was most pronounced in retina, sclera, and cornea as well as in the peripheral zone of the lens. Fluoride was not visible in the tissues or fluids of the eye.

Bone and cartilage

Fluoride was found to be concentrated in a very high degree in the mineralized hard tissues—bones, teeth and pathologic calcifications. In the unmineralized cartilages, however, no fluoride was noticed. Bromide was to a certain extent concentrated in compact bone. The uptake of ^{82}Br in cartilages, tendons and fasciae was, however, higher. Thiocyanate showed a lower and iodide the lowest tendency to accumulate in compact bone. Tendons, ligaments, and cartilages showed a concentration of thiocyanate, while iodide showed an uptake only in tendons and ligaments and not in cartilages. None of the ions under discussion were accumulated in bone marrow.

Teeth

The highest uptake in teeth was shown by fluoride with the greatest concentration in the pulpal zone of the dentine of developing teeth. Bromide could also be demonstrated in high concentration in teeth with a localization similar to that of fluoride. The uptake of thiocyanate was considerably lower and iodide did not show any tendency to be concentrated in teeth.

Muscles

All of the ions were present in a very low concentration in the muscles. The concentration was somewhat higher in the *myocardium*. This was most pronounced for bromide and thiocyanate.

Thyroid

The thyroid proved to be very specific in its quality to concentrate iodide. No evidence for the statement that the thyroid should be 'halideblind' was found. Beside

iodide, thiocyanate was the only ion which showed some concentration in the thyroid although its concentration there was only slightly higher than that in the blood.

Salivary glands

Iodide, thiocyanate, and bromide were accumulated in the salivary glands. Iodide and thiocyanate showed a higher uptake in the submaxillary gland than in the other salivary glands. Bromide, on the other hand, had a higher concentration in the sublingual gland. The total concentration of bromide in the salivary glands was, however, considerably lower than that of iodide and thiocyanate. Fluoride was not accumulated in the salivary glands.

Gastric mucosa

The investigations showed a strong gastric secretion of thiocyanate, iodide, and bromide ions, although somewhat less pronounced for the last kind. The secretion of fluoride was very low. The order of the secretion of the ions with the gastric juice seemed to be $SCN^- > I^- > Br^- > F^-$.

The highest concentration of the various ions within the gastric mucosa was in the surface epithelium. The activity lined the inner part of the gastric mucosa and could be followed down to the gastric pits. No appreciable concentration of activity was found in the glandular part of the mucosa or in the parietal or peptic cell regions.

Intestinal mucosa

The lumen of the small intestine was practically void of activity. It was found that there was a rapid reabsorption in the duodenum of the various ions secreted by the gastric mucosa.

In the mucosa of the *large intestine* thiocyanate accumulated to a certain extent. Much less was taken up of the other ions studied and fluoride not at all. After long survival periods relatively great iodide activity was visible in the contents of the large intestine. It will seem that this was a consequence of the fact that iodide appears in organic compounds and that iodide albumin is excreted by the intestine.

Liver

This organ did not show any tendency to accumulate any of the ions studied.

Lymptic tissue

This tissue showed a low and even content of all the ions studied. An exception was an accumulation of iodide in the thymus which was found in some pregnant animals and in foetuses. Radioiodide was also accumulated in the marginal sinus of the spleen some days after the injection.

Ovaries

A selective uptake of iodide could be seen in the ovarian follicles. This was not found with the other ions.

Foetus

Regarding the transfer to the foetus there was a marked difference between iodide and the other ions in that iodide was more concentrated in most of the foetal tissues

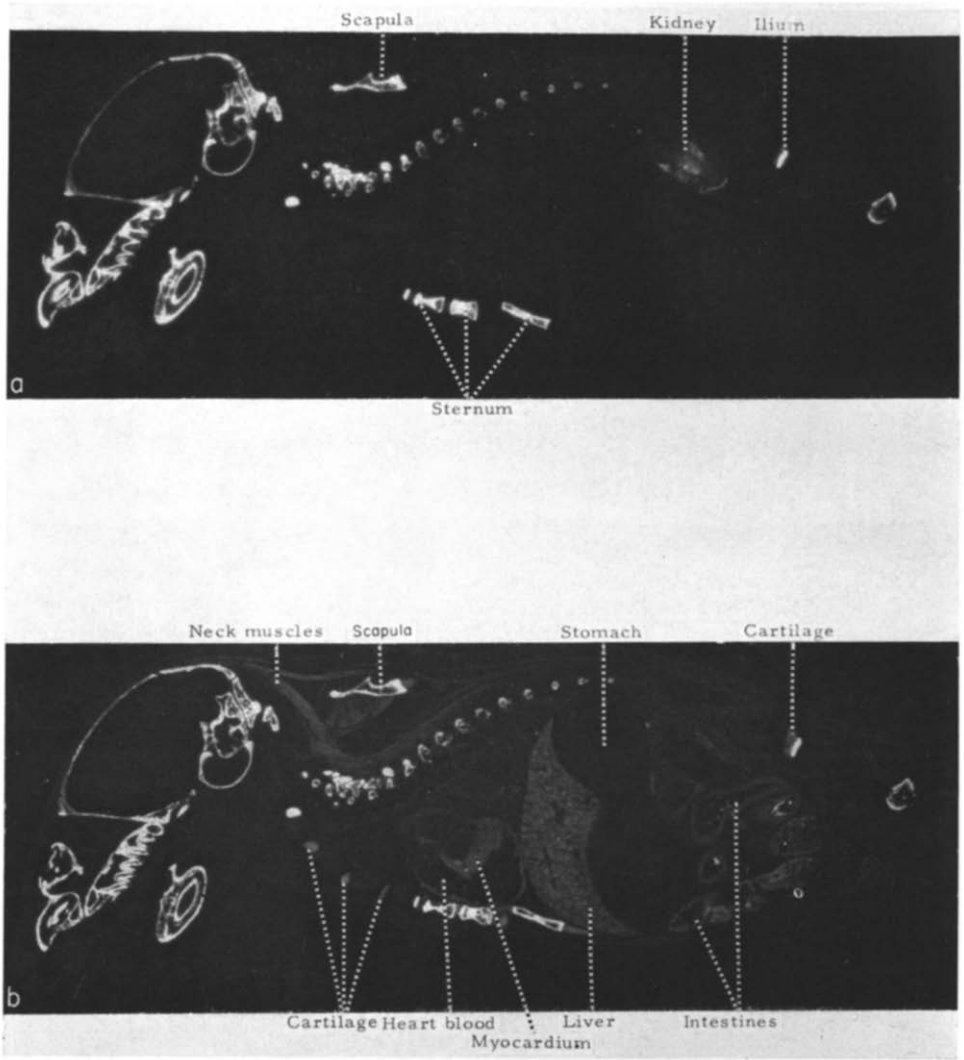

FIG. 1(a) Autoradiogram of ^{18}F in a 15 day-old rat, 30 min after i.v. injection of ^{18}F + ^{45}Ca. Accumulation of ^{18}F can be seen only in the mineralized hard tissues and (due to the excretion) in the kidney.

(b) Autoradiogram of ^{45}Ca from the same section as (a). No ^{45}Ca can be seen in the blood. The highest uptake can be noted in the mineralized hard tissues, but a fairly high concentration can also be seen in cartilage, some muscles, liver and intestinal mucosa and contents.

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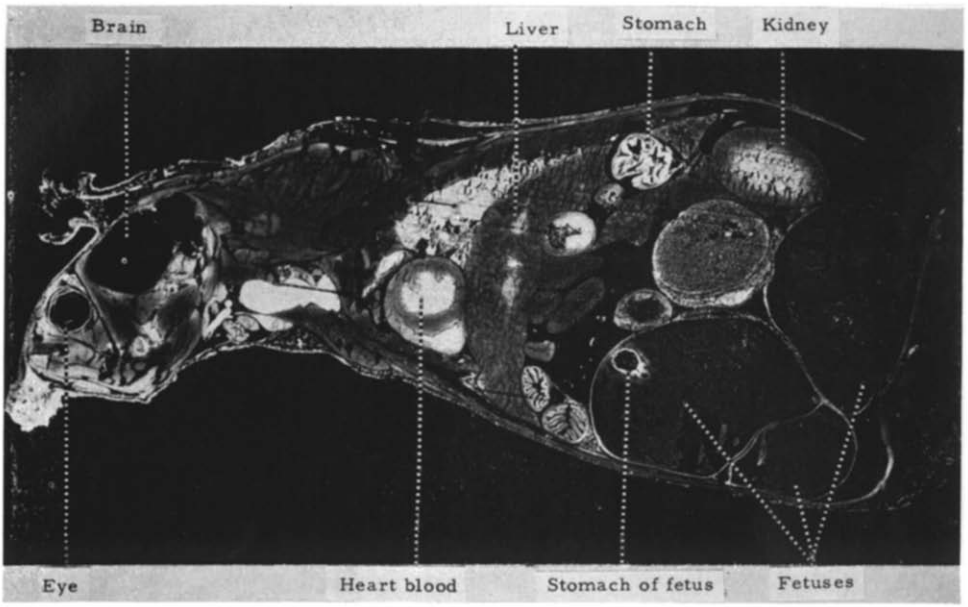

FIG. 2. Autoradiogram showing the distribution of ^{35}S in a pregnant mouse 1 hr after i.v. injection of $K^{35}SCN$. Note the high concentration in the gastric mucosa of both the mother and the foetus.

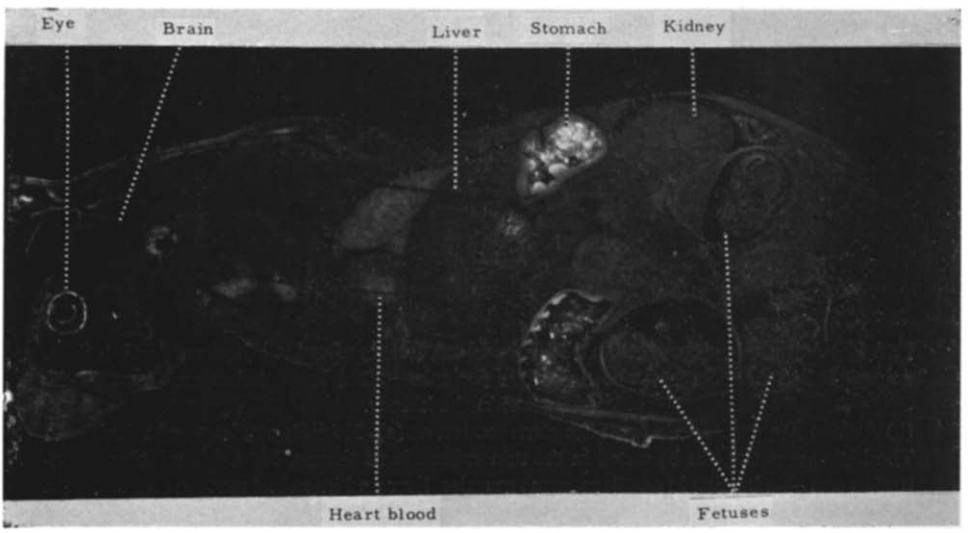

FIG. 3. Autoradiogram of the distribution of ^{82}Br in the body of a pregnant mouse 24 hr after i.v. injection.

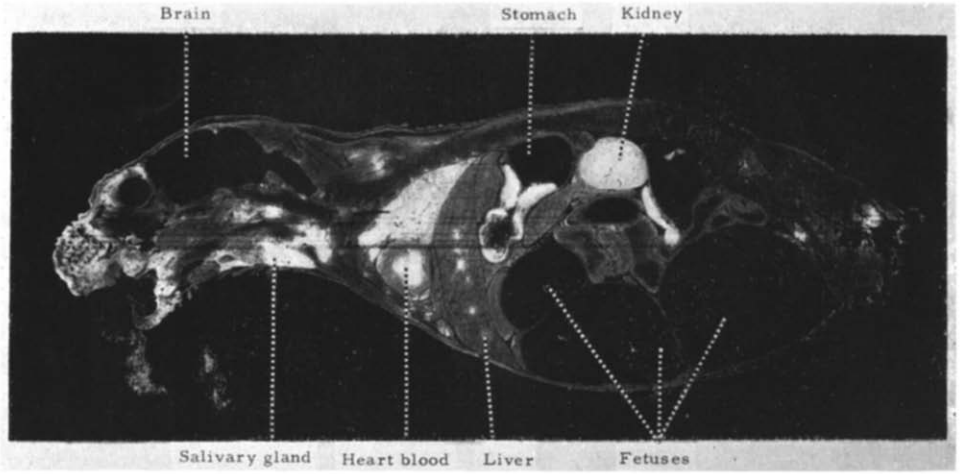

FIG. 4. Autoradiogram showing the distribution of ^{125}I in the body of a pregnant mouse 1 min after i.v. injection. Note the high excretion of iodide via the gastric mucosa.

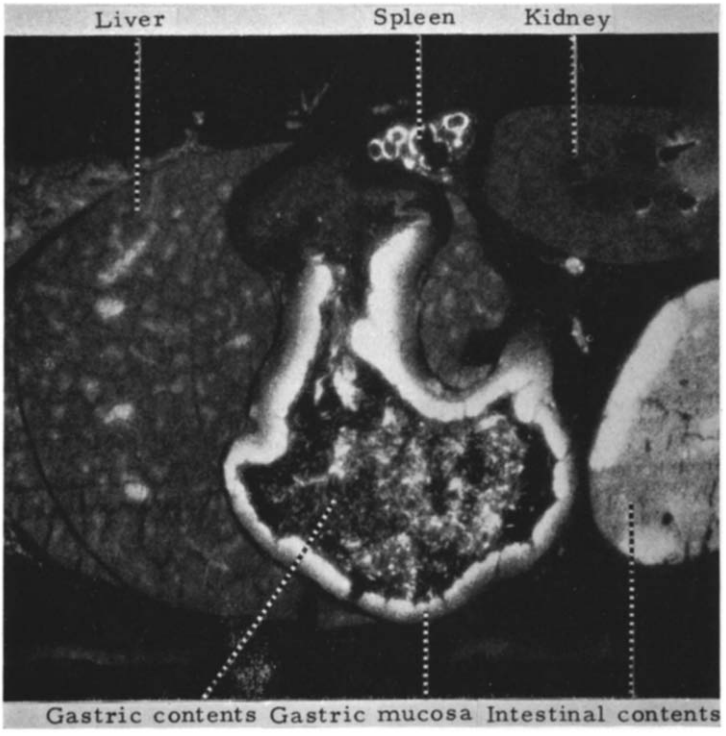

FIG. 5. Autoradiogram of the distribution of ^{131}I in the abdomen of a mouse 5 days after i.v. injection. Note the high concentration of iodide in the gastric mucosa and in the periphery of the follicles of the spleen.

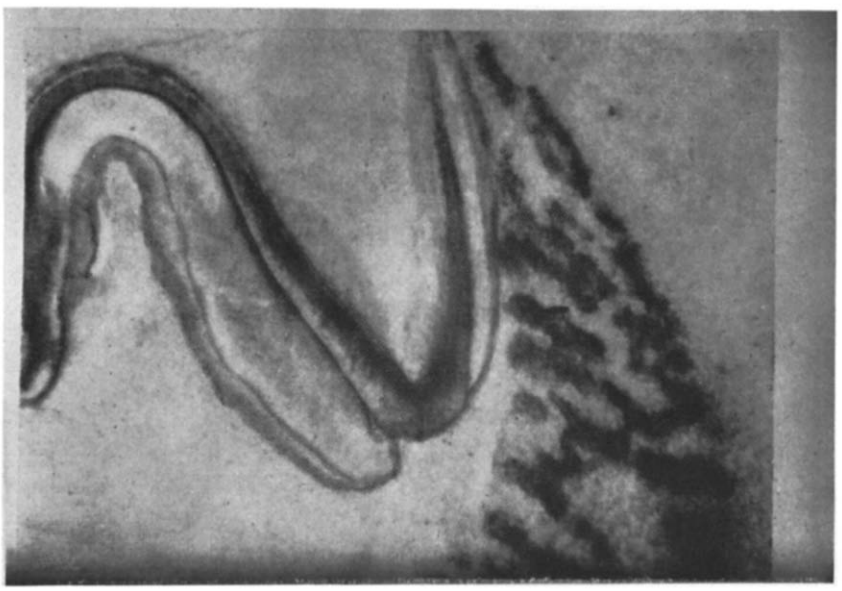

FIG. 6. Micro-autoradiogram of developing molar in 10 days-old rat suckling, 30 min after s.c. injection of ^{18}F. The stained 5μ section is kept in contact with the equally thick emulsion. Black dots and areas show distribution of ^{18}F. Weak diffuse uptake of mineralizing enamel contrasts against strong, concentrated uptake of pulpal dentine and bone trabeculae.

than in the corresponding tissues of the dam, while the other ions did not reach as high a level in the foetus as in the dam. Fluoride was found only in the foetal skeleton, and in much lower concentration there than in the maternal skeleton.

Placenta

Also in the placenta the concentration of iodide was much higher than that of the other ions. In the oldest placentas there was a spotwise concentration of fluoride, which in a special study was found to coincide with the degenerative calcifications that are common at the end of pregnancy.

DISCUSSION

The halide ions show notable differences as regards electronic structure. The fluoride ion has completely closed electronic shells, whereas the other halides only have an outer subshell closed. The ionic radius is considerably smaller for F^- than for other halide ions. It is also well known that fluoride differs markedly from the other halogens in its chemical properties. The solubilities of fluorides are, for example, markedly different from those of other halide compounds. Another important difference is the tendency of fluoride to form strong hydrogen bonds. Therefore it will seem natural that there should also occur biological differences between fluoride and other halides.

Of the ions investigated fluoride has clearly deviated from the others by being an exclusive bone-seeker. The property of fluoride to be easily bound to crystalline calcium salts evidently completely dominates its physiological distribution.

Among the other ions there are both similarities and dissimilarities. However, iodide also shows a certain specificity, partly through the selective uptake in the thyroid and partly because the distribution picture changes more with time than for the others, probably because an increasing portion of the radioiodide occurs in the form of thyroid hormones. Iodide also has other particularities. Thus the placenta forms a partial barrier for the other ions, especially fluoride, while radioiodide occurs in a higher concentration in most foetal tissues than in those of the dam. The tendency to be accumulated in the ovarian follicles is also notable.

The greatest similarities seem to be found in the distribution picture of bromide, chloride, and the pseudo-halide.

Some kinetic similarities of all the halides and pseudo-halide except fluoride, which have been commented upon a good deal in earlier literature, are found in the excretion with gastric juice and saliva. The tendency to be excreted with the gastric juice was most marked for iodide and the pseudo-halide and less for bromide and chloride. Fluoride was only occasionally found in the gastric contents and was probably due to the animals licking their urine. The site of secretion could not be established for chloride due to unsatisfactory autoradiographic resolution with ^{38}Cl, but seems for the rest of the secreted ions to be the surface epithelium of the gastric mucosa. The accumulation of activity in the surface epithelium is probably not due to absorption from the lumen of already excreted activity, since a concentration in the surface layer of the mucosa could be seen as early as one minute after the injection when the activity was not yet autoradiographically observable in the gastric contents. The fact that the halide concentration in the surface epithelium is higher than in the blood seems to indicate that it is that site of an active secretion of the halides.[12–14]

It is notable that the tendency to be concentrated in bone tissues, which is so marked for fluoride, is evident also for bromide and can be noticed for the others but is least pronounced for iodide. An uptake in cartilage is found mainly for bromide while several of the ions, although not fluoride, show an affinity for tendons and ligaments.

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