The Accumulation of $^{Y90}$ from an Equilibrium Mixture of $^{Sr90}-^{Y90}$ by *Artemia salina* (L.)\textsuperscript{1}

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**ABSTRACT**

*Artemia salina* (L.) was chosen as a representative organism in the second trophic level for studying one phase of the process of transfer of radionuclides through a marine food chain. Adult *Artemia* were cultured in filtered sea water which contained $^{Sr90}-^{Y90}$. They were not fed during the experiment which lasted 92 hours. They were removed at intervals, washed, and their radioactivity determined with an end-window counter. At 16 hours the concentration of radioactivity in the *Artemia* was about 40 times that of an equal weight of sea water. The radioactivity decayed at a rate which was similar to the decay rate of $^{Y90}$. It was estimated that only about 5 per cent of the activity remaining at secular equilibrium was due to $^{Sr90}$, the remainder being $^{Y90}$. This biological fractionation of $^{Sr90}-^{Y90}$ may affect the transfer of these hazardous nuclides through food chains leading to human food. The fractionation of these nuclides by laboratory glassware is briefly discussed.

It has become increasingly apparent that in order to determine the biological hazards of radioisotopes from reactor wastes and from fallout we must increase our knowledge of both the rate of accumulation and the maximum concentration in the ecosystems involved. With such information we can more intelligently assess the importance of these isotopes in the biology of the individuals. It has been shown (Boroughs et al. 1956a, 1956b) that strontium and yttrium fed to fish are rapidly excreted. It has also been found that a relatively small amount is accumulated from the water. However, in all instances the amount retained seems to be dependent upon a number of factors, e.g., species, age, food habits, and perhaps sex. It also appears that some organisms actually discriminate against one isotope in preference for another when both are present in the medium (Rice 1956). It is this effect which may reduce the concentration of isotopes such as $^{Sr90}$ at various trophic levels.

Since the planktonic crustaceans form a large percentage of the food at the lower trophic levels in the marine environment, they may serve as a means of transferring isotopes from the phytoplankton to the smaller carnivores. It was the purpose of the present work to determine the amount of $^{Sr90}$ and $^{Y90}$ accumulated by the brine shrimp, *Artemia salina* (Linnaeus), when these isotopes are present in sea water.

**METHODS**

All glassware used was freshly washed with concentrated $\text{H}_2\text{SO}_4$, detergent, and rinsed several times in distilled water. About 61.2 $\mu$C $^{Sr90}-^{Y90}$ from Oak Ridge was diluted to 100 ml with sea water filtered through a glass fiber filter which retained particles of bacterial dimensions (0.1 microns). From this solution five 0.5-ml aliquots were pipetted onto aluminum planchettes and dried. Twenty-five adult *Artemia salina* were added to the sea water containing the $^{Sr90}-^{Y90}$ mixture. Five brine shrimp were immediately removed from the solution, drained, and washed three times with distilled water. Preliminary experiments showed that very little radioactivity was lost by repetitive washing, and that distilled water and sea water washes were equivalent. The *Artemia* were then blotted with filter paper, arranged uniformly on an aluminum planchette, and dried under a heat lamp.

At intervals of 16, 40, 64, and 92 hours
Fig. 1. The radioactive decay of Sr$^{90}$-Y$^{90}$ in Artemia showing the accumulation of Y$^{90}$ rather than of Sr$^{90}$.

later, five Artemia were removed and prepared as above. During this time the brine shrimp were not fed. When each group of brine shrimp was removed, 0.5-ml aliquots of the sea water were also pipetted onto aluminum planchettes and dried.

As each set of samples was prepared it was immediately counted on a commercial scaler using an end-window Geiger-Müller tube. All samples were counted daily in order to follow the radioactive decay, and appropriate corrections were made for coincidence when necessary. Owing to the high cpm, all samples were counted on the third shelf of a lead support, or approximately 4 cm from the Geiger-Müller tube.

Strontium$^{90}$ from Oak Ridge is an equilibrium mixture of Sr$^{90}$ and Y$^{90}$. Strontium$^{90}$ has a half-life of about 28 years, and has a beta radiation with a maximum energy of 0.61 Mev. The daughter product of Sr$^{90}$ decay is Y$^{90}$ which has a half-life of about 2.5 days and has a beta radiation with a maximum energy of 2.18 Mev. At equilibrium the number of counts recorded is due to both the Sr$^{90}$ and its daughter Y$^{90}$. If absorbers are used, or if there is a biological fractionation of the isotopes, the Geiger-Müller tube will count all particles entering it, but because more high energy particles enter the tube per disintegration than low energy particles, it will be more efficient for Y$^{90}$. A lapse of 10 half-lives of Y$^{90}$ or about 25 days re-establishes the secular equilibrium of the isotopes. Thus it was necessary to count the samples at intervals for 25 days in order to be able to tell whether Sr$^{90}$ or Y$^{90}$ had been removed from the solution.

RESULTS AND DISCUSSION

Figure 1 is a semi-log plot of the radioactive decay of Artemia exposed to Sr$^{90}$-Y$^{90}$ sea water for 0, 16, 40, 64, and 92 hours. It can be seen that when the brine shrimp were removed, the cpm/Artemia ranged from 8,200 to 12,000, but 43 days later it had dropped to 35-56 cpm/Artemia. From these curves several conclusions may be drawn. According to Curve A, there was no immediate binding of either isotope by the shrimp, but Curve B indicates that a large amount of radioactivity was taken up by the shrimp during the first 16 hours. The zero-time Artemia had about 55 cpm/Artemia when the samples were counted for the first time. These shrimp averaged 10.3 mg, so that 1 gram of Artemia would take up about 5,335 cpm. The 16-hour Artemia, on
the other hand, had about 10^6 cpm/gram. One gram of sea water had about 25,000 cpm. These numbers show that the Artemia which were merely dipped into the water did not absorb much of the radioactivity present, but that the 16-hour Artemia actually concentrated it about 40 fold (10^6/25,000).

The decay rate indicates that about half the initial radioactivity disappeared in 2.5-3.0 days, and this time is approximately equal to the half-life of Y^90. The high level of radioactivity, therefore, is due to the Y^90 component, and the small residual radioactivity is due to the Sr^90.

The ratio of cpm/g Artemia to cpm/g sea water we call the concentration ratio. Table 1 shows the concentration ratios of the various samples at equilibrium. It can be seen that there is no significant change between the amount of Sr^90 picked up after 16 hours and that picked up after 92 hours in the medium. On a weight basis one can therefore expect adult brine shrimp in a medium containing Sr^90-Y^90 to accumulate about 13 per cent of the total available nuclides. At equilibrium, only half the disintegrations are due to Sr^90, but because of the difference in counting efficiency for the two different energy beta particles, it is thought that only about 5 per cent of the radioactivity at equilibrium is due to the Sr^90.

The concentration ratio at equilibrium is roughly the same regardless of the time the Artemia spent in the radioactive water (up to 92 hours). However, these results might be changed by longer periods of exposure, or by differences in age and sex of the animals.

It is apparent from the work of Chipman et al. (1955, 1956) that age is an important factor in determining the amount of Sr accumulated from solution. These workers exposed Artemia salina nauplii to Sr^90-Y^90 in sea water for 24 hours. Table 2 shows their data calculated on the same basis as in Table 1. These data suggest that young Artemia accumulate more Sr from sea water than do adults. The higher concentration ratios may mean that Artemia which spend their entire lives in sea water containing Sr^90-Y^90 will have more than the five per cent Sr^90 we found with adults.

<table>
<thead>
<tr>
<th>Sample</th>
<th>cpm/g</th>
<th>Concentration ratio</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sea water</td>
<td>36,000</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Artemia—16 hrs</td>
<td>4,600</td>
<td>0.13</td>
<td></td>
</tr>
<tr>
<td>Artemia—40 hrs</td>
<td>3,500</td>
<td>0.10</td>
<td>0.13</td>
</tr>
<tr>
<td>Artemia—64 hrs</td>
<td>4,000</td>
<td>0.14</td>
<td></td>
</tr>
<tr>
<td>Artemia—92 hrs</td>
<td>5,600</td>
<td>0.16</td>
<td></td>
</tr>
</tbody>
</table>

The amount of Sr^90 taken up may also be increased if the Sr^90 is present in phytoplankton which the Artemia eat. Artemia, therefore, discriminate against the long-lived Sr^90 in preference for the short-lived Y^90. It is possible that through such fractionation the more hazardous Sr^90 may be lost by the time the higher trophic levels are reached. On the other hand, some organisms may concentrate strontium almost exclusively from an equilibrium mixture. Thus, Rice (1956) found that 90 or more per cent of the radioactivity in 10 species of algae was due to yttrium, but that Carteria accumulated only strontium. Such organisms as Carteria, therefore, may be a considerable source of contamination which can be transferred up the food chain. It is apparent that the loss of an isotope in one food cycle may promote the enrichment of this isotope in another cycle.

The radioactivity of the sea water samples was also followed from day to day. Figure 2 shows that at zero time the water had an activity of 15 X 10^6 cpm per 0.5 ml. Water samples which were removed 40, 64, and 92 hours later, however, had an activity of 12.5 to 13.5 X 10^6 cpm per 0.5 ml. At equilibrium the radioactivity in all the water samples had increased to 17-18.5 X 10^6 cpm per 0.5 ml. This was an increase in radioactivity of 20-60 per cent above that found when the samples were first counted.
Of particular interest is the fact that the number of counts/minute for all the samples at equilibrium was approximately the same irrespective of the number of counts/minute when the samples were prepared (cf. $0^\text{th}$ dose and $92^\text{th}$ dose). This rise in activity implies that in all the samples some $\text{Y}^{90}$ had been removed from the original equilibrium mixture.

It must be pointed out that using an equilibrium mixture of $\text{Sr}^{90}-\text{Y}^{90}$ for such studies as these presents many problems, because it is difficult actually to assess the role of each isotope in the biological sense. This difficulty is due to the physical and chemical nature of the mother and daughter products. Strontium is very likely ionic in sea water, but yttrium is probably at least partially colloidal. Strontium is bivalent and yttrium trivalent, and there is a significant difference in the size of their ionic radii. These differences mean that the biological binding sites for the two elements may be wholly different, but the sites may also overlap. If the sea water planchettes had been counted only at the time they were made, one would never suspect that there would be a significant change in the amount of radioactivity over a short time interval. Since $\text{Sr}^{90}$ has a half-life of about 28 years, one would expect a constant count rate over a period of 30 days. However, when the isotope from Oak Ridge was pipetted into the sea water, $\text{Y}^{90}$ was immediately lost, most likely by being adsorbed onto the glass. This means that each step in pipetting removed more yttrium from the mixture and upset the equilibrium. To test this hypothesis $\text{Sr}^{90}-\text{Y}^{90}$ was added to flasks con-

![Graph](image_url)
taining sea water or distilled water. Figure 3 shows that more Y\textsuperscript{90} was adsorbed from distilled water than from sea water.

Thus it appears to us that in working with such isotopes which may have a tendency to become adsorbed onto surfaces, much care must be taken in the interpretation of data, since considerable variation may occur depending upon the time intervals concerned. Furthermore, it is apparent that it is very difficult to work with an equilibrium mixture of Sr\textsuperscript{90} and Y\textsuperscript{90} using ordinary glassware.

REFERENCES


