BIOACCUMULATION OF RADIOACTIVE GOLD USED AS A SEDIMENT TRACER IN THE ESTUARINE ENVIRONMENT [1]

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ABSTRACT

The accumulation of radioactive gold by selected members of a marine animal community and sediment material was followed under laboratory and field conditions. In the laboratory an aqueous solution of radioactive gold was placed directly in the gut of blue crabs, toadfish, and croakers. There was little transfer of the isotope to various tissues in these organisms. A group of toadfish which were fed radioactive gold in an aqueous solution retained more of the isotope than did a group which were fed the same amount of radioactivity sorbed onto clay particles. Also, crabs, fish, clams, and samples of bentonite clay were maintained for 25 days in 1,000 liters of cotton-filtered sea water containing radioactive gold. Crabs accumulated the most radioactivity followed, in descending order, by clams, clay, and fish. A field experiment was conducted in cooperation with the U.S. Army Corps of Engineers in the Cape Fear River, N.C. Caged and indigenous free-swimming organisms were exposed to sediment-sorbed radioactive gold used as a sediment tracer by the Corps. Oysters, crabs, and fish maintained in cages in the experimental area were sampled periodically. The maximum level of radioactive gold in the caged organisms (70.9 millimicrocuries per gram wet weight tissue) was detected in oysters 17 hours after the isotope was released. Indigenous organisms collected 41 hours after the radioactivity was released contained no detectable radioactive gold.

The uncontrolled release of radioactivity into estuarine waters could so contaminate marine organisms that they would be unsafe for use as food by man. However, with the use of basic data from laboratory experiments to evaluate the quantity and rates of release of radioactive materials, and with sensitive instruments to measure the resulting levels of radioactivity, radioisotopes can be released into the natural environment without adversely affecting seafood organisms. Also, when released in this manner, radioisotopes can safely be used *in situ* to investigate many ecological problems.

A study was made to determine the bioaccumulation of sediment-sorbed radioactive gold released into the Cape Fear River. This study was carried out with investigators of the U.S. Army Corps of Engineers, who used this isotope to trace sediment movement in the river. Prior to this investigation, radioactive gold had been used successfully as a sediment tracer by the Corps of Engineers in several bays and harbor systems in the United States.

Determinations of gold in marine organisms have been reported by Noddack (1939), Vinogradov (1944), and more recently by Fukai (1962). Equilibrium values for the distribution of gold between sea water and organisms have been calculated by Krone (1959) using the data of both Noddack and Vinogradov. The capacity of marine organisms to accumulate radioactive gold from contaminated sea water or from contaminated food organisms has not been determined.

Since marine organisms could be exposed to radioactive gold used in sediment tracer experiments, an evaluation of the accumulation of this isotope by several marine organisms was made by (1) conducting laboratory experiments to deter-

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mine the behavior of gold in sea water, and the translocation of this element in several animals; (2) observing the transfer of radioactive gold from sea water to a community maintained in a large salt-water tank; and (3) measuring the accumulation of the isotope in animals and sediments in the natural environment.

PRELIMINARY LABORATORY EXPERIMENTS

The behavior of radioactive gold in sea water, its affinity for sediments, and its assimilation by individual organisms were studied prior to following the movement of radioactive gold through a community of organisms. Characteristics of radioactive gold in sea water and the effect of salinity on the sediment-sorption of this isotope were observed. Bentonite clay composed of montmorillonite clay minerals was used in the sediment-sorption experiments because of the natural occurrence of these minerals in marine sediments and because of their reported sorptive properties (Grim, 1953). Experiments on the assimilation of radioactive gold by individual animals were conducted in sea water ranging in salinity from 28 to 32 °/₀₀ and having a temperature range of 27.0° to 30.2° C. These animals included blue crabs, *Callinectes sapidus;* oyster toadfish, *Opsanus tau;* and Atlantic croakers, *Micropogon undulatus;* all collected near Beaufort, N.C.

Gold 199 was selected in preference to gold 198 because of its longer half-life (3.1 days compared with 2.7 days) and its availability in the carrier-free form (2.09×10^5 curies (c.)/gram). The gold 199 in the form of auric chloride was supplied from Oak Ridge, Tenn.

The radioactivity content of sediment, water, and animals in these preliminary experiments was measured with a scintillation detector large enough to contain live animals (4¼ inches diameter by 9 inches long) and a single-channel gamma spectrometer. Measurements were corrected for decay, geometry, and background.

BEHAVIOR OF RADIOACTIVE GOLD IN SEA WATER

The behavior of tracer amounts of radioactive gold in aqueous solutions was investigated by Schweitzer and Bishop (1953). These investigators have shown by filtration and centrifugation that gold (in concentrations less than 10^{-8} M) appears to behave as a radiocolloid in certain aqueous solutions ranging in pH from 2 to 12. Schweitzer and Jackson (1952) also discussed the use of cation exchangers in the identification of radiocolloids. The uptake of a cationic tracer by an exchanger should decrease with an increase in the cationic concentration of the solution. If the tracer is a radiocolloid, however, the uptake increases as the cationic concentration increases. This is attributed to the effects of the cations acting as a coagulant and the exchanger acting as an absorbent.

Several experiments were conducted in this study to determine, qualitatively, the behavior of radioactive gold in sea water. A water sample was prepared by adding 200 microcuries (μc.) of gold 199 (4.9×10^{-11} M) to 1 liter of Millipore-filtered sea water of 30 °/₀₀ salinity and a pH of 8.2. The temperature of the water was maintained at 25°±2° C. during the experiment. Centrifugation of 500 ml. of this sample at 3,000 r.p.m. for 15 minutes forced 58 percent of the radioactivity to the bottom of the tube indicating that gold particles had been formed. When 500 ml. of the initial sample were passed through a Millipore filter of 45 millimicrons (mμ) pore diameter, 100 percent of the radioactivity was removed. Even though some of the gold could have been retained as a result of pores being clogged by large molecules, the gold in sea water appeared to be particulate, rather than ionic.

Further indications of the properties of radioactive gold in sea water were obtained by observing the sorption of gold onto clay. Equal amounts (200 μc.) of gold 199 were placed in four tanks, each containing 1 liter of millipore-filtered water and 1 g. of clay. The first tank contained distilled water; the second, water having a salinity of 8°/₀₀; the third, 24°/₀₀; and the fourth, 34°/₀₀. The waters having salinities of 8°/₀₀ and 24°/₀₀ were prepared by adding appropriate amounts of distilled water to sea water having a salinity of 34°/₀₀. The water in the tanks was agitated for 15 minutes and allowed to stand for 24 hours. The clay slurry was withdrawn from each tank, centrifuged, and the water decanted. The clay was then dried and the amount of radioactive gold measured.

The uptake of gold 199 on clay increased as the salinity increased (fig. 1). There are at least two possible explanations for this sorption phe-

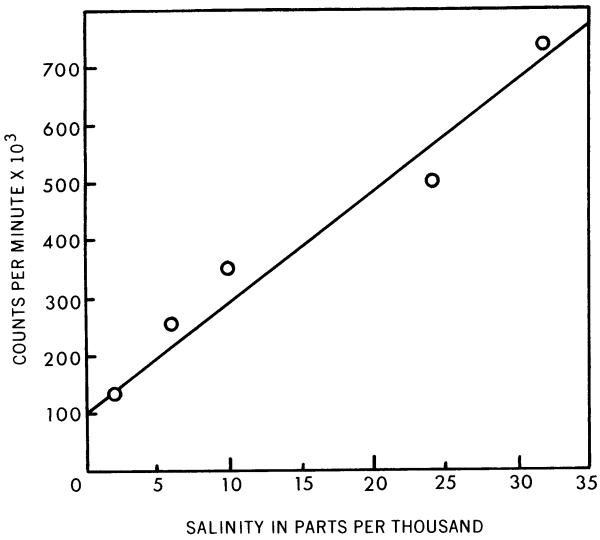

FIGURE 1.—Sorption of gold 199 by montmorillonite clay from sea water with increasing salinity.

nomenon: (1) An increase in salinity caused the clay to clump, and radioactive gold was trapped within the clumps; and (2) The increase in salinity caused the gold particles to coagulate and to sorb more readily onto the clay. If the latter explanation is accepted as correct, it would appear that radioactive gold was colloidal in this sample of sea water.

DISTRIBUTION OF RADIOACTIVE GOLD IN BLUE CRABS

Edible portions of an organism such as a blue crab can be relatively free from radioactivity even though the total radioactivity content of the organism is high. This is possible when the activity is isolated in the stomach or associated with the highly sorptive carapace and other external sites. That this does occur was shown by determining the distribution of ingested radioactive gold in the blue crab. Twelve crabs with an average weight of 152 g. were made radioactive by pipetting 75.7 μc. (50 microliters) of gold 199 directly into the cardiac stomach of each crab. They were then placed in flowing sea water. At intervals of 3, 4, and 5 days after dosing, three crabs were killed, dissected, and measured for contained activity.

Radioactivity content of tissues at 4 days indicated that only a small portion of the gold 199 was assimilated from the stomach (table 1). In view of the short half-life of the isotope, more than tracer amounts of radioactive gold would need to be ingested rapidly in order to build up concentrations in edible tissues.

DISTRIBUTION OF RADIOACTIVE GOLD IN TOADFISH

Bottom feeders, such as the oyster toadfish, could become radioactive by taking in sediments with loosely bound sorbed activity. If the radioactivity were tightly bound to the sediment particles, however, the sediment-sorption phenomenon would reduce the possibility of animal contamination by retaining and confining the radioactivity.

TABLE 1.—*Radioactivity content of crab tissue 4 days after an oral dose of 75.7 μc. of gold 199*

Tissue	Activity	Dose in entire tissue
	μc./g.	*Percent*
Digestive gland	9.3×10^{-1}	12
Stomach—gut	8.4×10^{-1}	3
Gonad	1.6×10^{-1}	.8
Gills	5.3×10^{-2}	.7
Muscle	2.5×10^{-2}	.6
Carapace	3.8×10^{-3}	.08
Blood	1.7×10^{-3}	.04
Total		17

To test the capacity of fish to "strip" radioactive material from sediments, five male toadfish, average weight 325 g., were each force-fed 75.7 μc. of radioactive gold dispersed in 2 ml. of sea water. Five other males of similar size were fed the same amount of radioactivity sorbed onto 25 mg. of montmorillonite clay suspended in 2 ml. of sea water. The sea water used in both instances had a salinity of 30°/oo and a pH of 8.1. After the fish had been fed the isotope, they were placed in flowing sea water. Forty-eight hours later, the fish were killed and dissected, and the amount of radioactive gold in various tissues was measured.

A comparison of the activity of tissues from both groups of fish shows that most of the gold remained sorbed onto the clay as it passed through the digestive system of the fish (table 2). The fish fed the isotope in sea water retained more activity than those fed the isotope sorbed onto clay.

DISTRIBUTION OF RADIOACTIVE GOLD IN CROAKERS

The gastro-intestinal absorption and distribution of gold 199 in the tissues of the croaker were determined after 75.7 μc. of the isotope were

TABLE 2.—*Radioactivity content of toadfish tissues following oral doses of gold 199*

Tissue	Gold administered in solution	Gold administered sorbed onto sediment
	μc./g.	*μc./g.*
Stomach	3.6×10^{-1}	6×10^{-3}
Liver	1.5×10^{-1}	8×10^{-4}
Gut	1.1×10^{-1}	5×10^{-2}
Gill	4.3×10^{-2}	2×10^{-4}
Kidney	4.0×10^{-3}	1×10^{-3}
Blood	4.0×10^{-3}	5×10^{-4}

pipetted into the stomachs of six fish. The fish were then placed in flowing sea water. Three were killed, dissected, and tissues were measured for radioactive gold content after 78 hours and three after 148 hours.

Less than 1 percent of the dose was assimilated by the tissue, about 99 percent having been eliminated within 78 hours (table 3). Of the relatively small amount assimilated, kidney and gills had the highest concentrations of gold 199 per unit weight. After 148 hours the gills and spleen had the highest concentrations. The increase in gold 199 content in some of the tissues during the period from 78 to 148 hours is probably the result of continued assimilation from the digestive tract and translocation of gold 199 from other tissues.

TABLE 3.—*Distribution of radioactive gold in croaker tissue*

Tissue	Dose	
	78 hours	148 hours
	Percent	*Percent*
Kidney	0.038	0.01
Gills	.035	.056
Skin, scales	.017	.009
Liver	.006	.02
Muscle	.003	.03
Heart	.002	.0008
Spleen	.001	.042
Gonad	----------	.001

EXPERIMENTAL ENVIRONMENT STUDY

Since the planned application of radioactive gold in the Cape Fear River would expose the community of indigenous organisms further laboratory studies were conducted with a marine community maintained in a large tank. Data obtained from tank experiments involving relatively large volumes of sea water (1,000 l.) and a community of organisms have advantages over data for individual species held in smaller volumes of water. Increasing the volume of water enlarges the experimental environment, and chronic as well as acute contamination can be observed since observations can be made over longer periods of time because of the improved physiological condition of the organism.

An experimental environment was established to follow the movement of radioactive gold in a marine community. The community, which consisted of clams and clam shells, *Mercenaria mercenaria*; blue crabs; and sheepshead minnow, *Cyprinodon variegatus*; as well as sediment samples of clay, was installed in a large (172 by 117 by 61 cm.) fiberglass tank containing 1,000 l. of cotton-filtered sea water (table 4). Ten clams were placed directly on the bottom of the tank, and 10 in the sediment. Enough gold 199, as $AuCl_3$, was added to the water to give a concentration of 0.0142 μc/ml. After the gold was added, the pH of the water was 8.1. Throughout the experiment the water temperature was maintained at 21°± 3° C. and the salinity at 32±0.10°/₀₀, the latter by adding distilled water when necessary to compensate for evaporation. A plastic impeller pump circulated the water continuously.

Clay sediment introduced into the tank was first placed in glass bowls (3 cm. deep by 12.7 cm. in diameter) after being thoroughly wetted as follows: 6 kg. of clay were suspended in 10 liters of sea water for 48 hours, and the excess water was decanted from the clay, leaving it with a pastelike consistency. In each bowl, 200 g. of the wet clay (125 g. dry weight) were placed forming a smooth substrate that was not disturbed by water circulation.

The radioactivity content of whole animals and sediment was measured periodically, with the organisms afterward being returned to the tank. Oysters, clams, and crabs were prepared for radioactivity analysis by wrapping them in a thin transparent plastic sheet. Fish were placed in dark glass jars containing nonradioactive sea water and counted for 3 minutes. Sediments were removed from the tank with a small diameter (2.5 cm.) core sampler and were not returned to the tank. This procedure of measuring the activity in a whole animal eliminated the need for killing it, and the number of individuals was not reduced with sampling. Also, uptake could be followed on the same individual throughout the experiment.

Appropriate corrections were made for decay, geometry, and background on all radioactivity measurements unless otherwise stated.

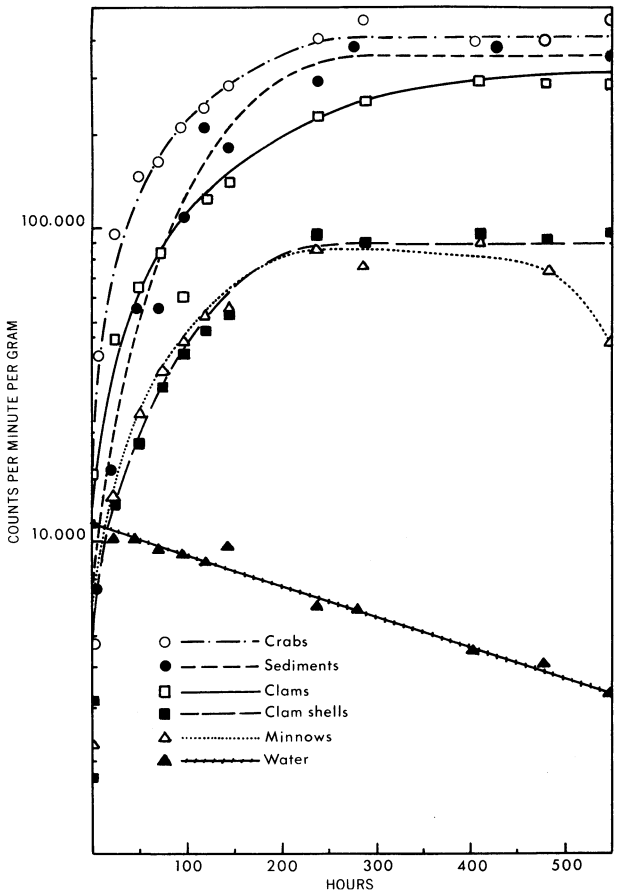

FIGURE 2.—Movement of gold 199 in an experimental marine environment.

Components of the community rapidly accumulated radioactive gold and reached an apparent steady state after 225 hours (fig. 2). There was not a direct relation between the accumulation of the radioactive gold by the components and the loss of the isotope from the water. Even though the radioactive content of the organisms and sediments did not increase after 225 hours, the radioactivity content of the water continued to decrease. This decrease was attributed to sorption of the radioactivity to exposed surfaces in the tank (a total area of 29,784 cm.^{2}) and to accumulation by patches of bacterial film that appeared on the sides of the tank after the experiment had been in progress about 120 hours. Although it was not possible to measure the total amount of film present, 1 g. of this material contained 57 μc. of radioactive gold after 600 hours. The distribution of the radioactive gold in the

TABLE 4.—*Composition of experimental environment showing distribution of gold 199, 600 hours after introduction of the isotope*

Component	Individuals	Total wet weight	Gold 199 content after 600 hours
	Number	*G.*	*μc.*
Water		1.0 x 10^6	4,600
Sediment		1,000	330
Crabs	5	620	340
Clams	20	2,800	1,100
Clam shells	10	130	10
Fish	10	46	230
Total uptake by components			6,610
Loss unaccounted for			7,590

community at the end of the experiment is shown in table 4.

Crabs accumulated more radioactive gold than the other organisms in the experiment. As an estuarine species, blue crabs are a part of a marine community that appears most likely to be exposed to radioactive material in any appreciable concentration. As bottom-dwelling, omnivorous predators and scavengers, blue crabs are, at one time or another, in contact with almost all of the abiotic components of their environment. The carapace and gills offer many surface sites for sorption of materials from the environment. Gill area alone of a blue crab has been estimated to be about 275,000 mm.^{2} (Gray, 1957).

Although sediments often are not included in the food web of marine organisms, many organisms can utilize materials sorbed onto sediments as a source of nourishment. Thus, it is necessary to observe accumulation of gold by sediments as well

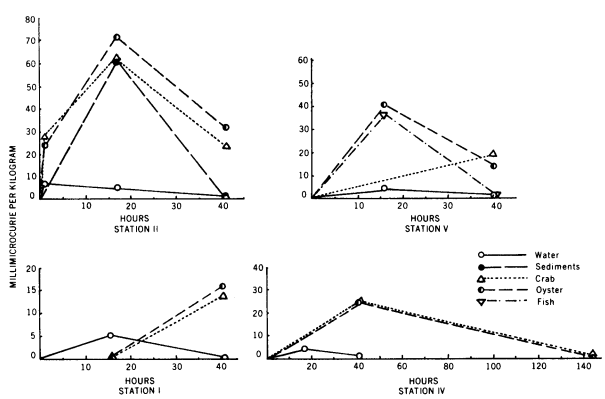

FIGURE 3.—Sorption of gold 199 by sediment in an experimental environment. Curve *A* is activity corrected for decay, curve *B* is observed activity.

as accumulation by the biota. In the present experiment, an increase in sediment activity up to the 250th hour occurred (figs. 2 and 3). From this time until the experiment ended, the activity was maintained with no significant increase or loss; it appears that the sediments were saturated with gold at 250 hours (curve A, fig. 3). The actual activity of the sediments, uncorrected for decay, is shown in curve B of figure 3. The latter values would be of more importance to the health physicists, as they show the actual amount of activity present in the sediments. If the gold were released into sea water in a restricted area where dispersion was minimal, these results indicate that natural decay would exceed uptake after about 150 hours with a resulting decrease in sediment activity.

Live clams accumulated more radioactivity than did the separated clam shells. Since clams feed by filtering suspended matter from large volumes of water passing over their gills, they are susceptible to contamination from particulate radioactive materials released into the estuarine environment. As bottom dwellers, restricted in their movements, they are in contact with sediments and associated radionuclides. A comparison of the mean radioactivity content of 10 clams to 10 separated clam shells on a unit weight basis by a standard t test at the .05 level showed that live clams were significantly more radioactive. The higher concentration of the isotope in the live animals was attributed to the movement of water through the animals. This movement would expose internal tissues as well as shell surfaces to contaminated water.

The clams placed on top of the sediment at the beginning of the experiment accumulated much less radioactive gold than those placed in areas without sediment. As the experiment progressed, the clams burrowed into the sediments, leaving only a portion of their shells exposed. The burrowing clams contained 38 percent less activity than those that remained on the bottom surface of the tank—an indication that a large portion of the gold accumulated by clams was due to sorption of the shell.

Fish accumulate radioactive materials by adsorption, absorption, or by ingestion. In nature, these three modes of uptake can occur simultaneously, singly, or in various combinations, depending upon the physical state of the isotope in the water, the food habits of the fish, and the length of time the fish remains in a polluted area. In this experiment, the sheepshead minnow accumulated the least amount of radioactive gold of any test organisms (fig. 2). The activity of the fish decreased during the latter stages of the experiment. A sloughing off of the epidermal mucous layer containing sorbed activity could account for the loss, or the physiological condition of the fish could have deteriorated and their rate of metabolism changed.

FIELD INVESTIGATIONS

The accumulation of radioactive gold released into the Cape Fear River was observed in both indigenous organisms and in caged organisms collected in Beaufort and maintained in the river. The investigations were coordinated with the release of radioactive gold which had been sorbed onto sediment particles.

The specific problem under investigation by the U.S. Army Corps of Engineers was to determine if sediment being deposited in the channels of the Cape Fear River, N.C., near the Sunny Point Army Piers near Southport, N.C., had been transported by currents from a spoil area on the opposite side of the river (U.S. Army Corps of Engineers, 1964). The answer was sought by tagging a small quantity of the sediment with gold 198, releasing this sediment in the spoil area, and tracing its distribution with an underwater scintillation probe mounted on a sled. The sediment was tagged by first forming a slurry into which 5 c. of gold 198 chloride were thoroughly mixed to allow maximum sorption. This was done in a special conical-bottomed container which served also as the release mechanism for the tagged sediment. The container floated in the water and was towed by boat over a predetermined course while the tagged sediment was being released. The entire procedure has been described in greater detail by Krone (1960).

Gold-tagged sediments were released at two different sites. The first 5 c. "drop" was made on October 24, 1962, at 6 p.m., at high tide, along an east-west line on the southern end of the Spoil area (fig. 4). A second 5 c. drop was made on the northern end of the spoil area on October 25 at 4 p.m., also at high tide. A water sample collected in the drop zone at this time had a

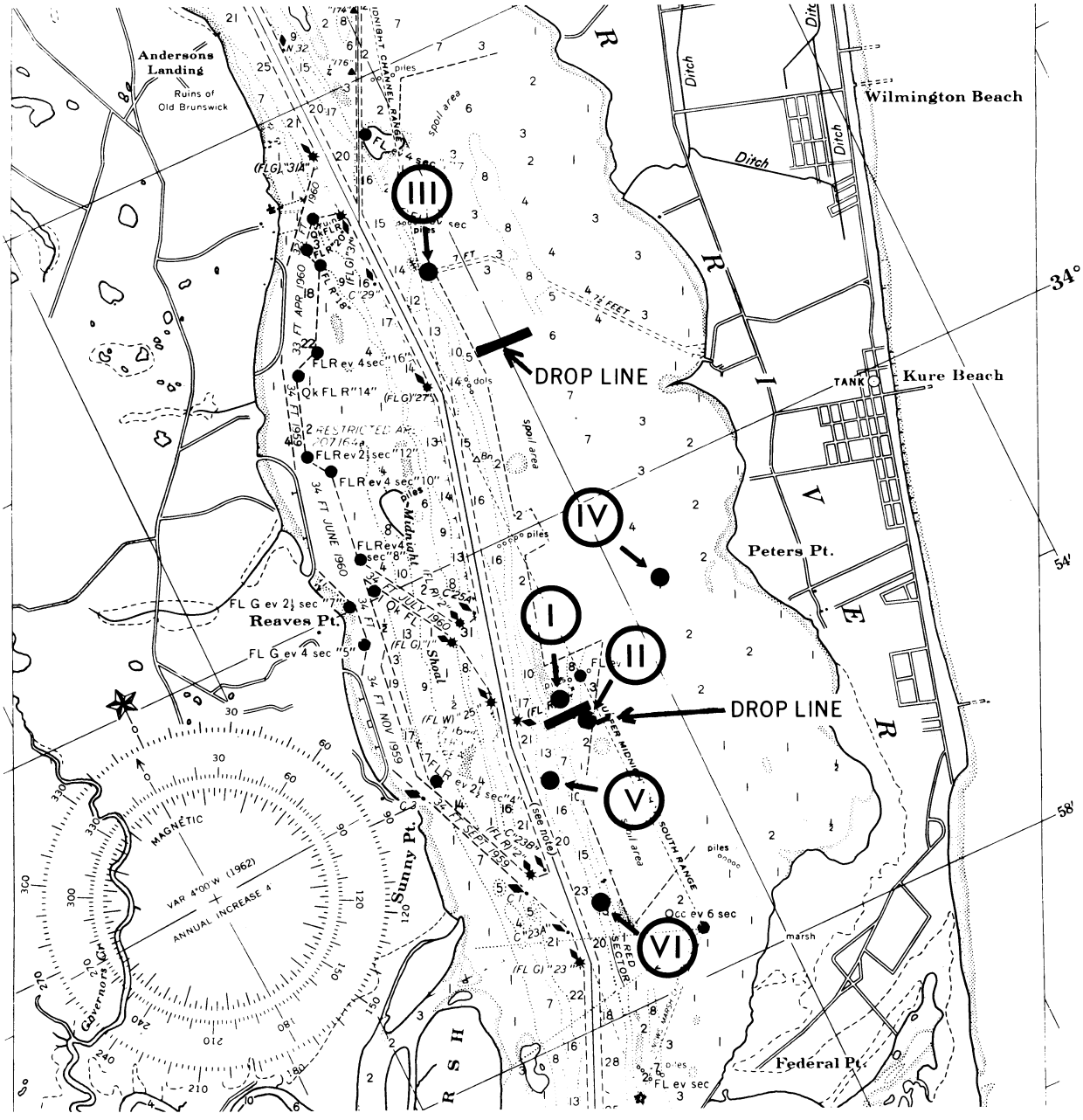

FIGURE 4.—Location of sampling stations and gold tracer drop zones in the Cape Fear River.

salinity of 24 ‰, temperature of 18° C., and pH of 7.9.

Animals and sediment from the drop zone area were collected before and after the release of the labeled sediment. Samples of sediment and sea water, and of blue crabs; Atlantic croaker; star drum, *Stellifer lanceolatus:* flounder, *Paralichthys* sp.; and American menhaden, *Brevoortia tyrannus,* were collected 1 week prior to the first drop in order to measure background activity. Organisms and sediment were collected again on the third day following the release of the labeled sediment. In addition to those species collected before the release of labeled sediment, the white

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shrimp, *Penaeus setiferus*, appeared in samples taken after the release. Sediment samples from the drop zone were composed mostly of sand. Additional information on the sediments is given by the U.S. Army Corps of Engineers (1964).

Cages containing various estuarine organisms were placed at six stations around the proposed drop zone 24 hours before the scheduled release (fig. 4). Test animals at each station included 25 blue crabs; 50 oysters, *Crassostrea virginica;* and 50 mummichogs, *Fundulus heteroclitus*. Ten croakers were included at stations IV, V, and VI. The blue crabs were kept in separate cages to prevent predation on the other test animals. Also, at each station, plastic petri dishes filled with montmorillonite clay were placed in minnow traps to test for adsorption of gold 198. Five to ten animals of each species were removed from the cages for each sample.

The radioactivity content of the field samples was measured at the Bureau of Commercial Fisheries Radiobiological Laboratory, Beaufort, N.C., 110 miles from the sampling area. The detector system consisted of a 3-inch NaI(Tl) crystal coupled to a single-channel gamma spectrometer. Sediment and biological samples were placed in individual plastic bags, packed in an ice chest, and transported to the laboratory as soon as possible after sampling. Water samples were held in screw cap jars. If the water samples were found to be radioactive, the water was Millipore-filtered and counted again, along with the separated material, to determine whether the radioactive gold was associated with suspended material.

The various samples were measured for radioactivity in a manner that permitted the comparison of organisms, sediments, and water. To make these comparisons, it was necessary that the radioactivity in all of the samples be measured under similar conditions of geometry. Organisms containing gold 198 were measured for radioactivity before and after being dissolved in nitric acid and diluted to 900 ml. A factor was thus obtained for converting measurements made on the intact organisms to measurements which would be obtained after dissolving and diluting the organisms to 900 ml. A factor was obtained for the sediments in a similar manner, except that no acid was used and Calgon was added as a wetting agent. No preparation was necessary to measure gold 199 in the 900 ml. samples of water. Since radioactivity measurements in intact organisms and sediments could be converted to measurements based on their being contained in a 900-ml. volume of water, and since all measurements of water were for 900 ml., it was possible to compare activity contained in these three types of samples.

The radioactivity content of water, sediments, and biota varied widely between stations. Biological samples from stations III and VI did not contain measurable amounts of radioactivity at any time (table 5). As one would expect, biological samples from station II, located directly in the drop zone, contained higher concentrations of radioactivity than those of other stations. Lateral dispersion of radiogold was indicated by the increase in activity in the crabs and oysters from stations I and IV after 41 hours elapsed time.

The second application of gold appeared to have little or no effect on the levels of concentration in the samples, except perhaps those in station IV. Oysters and crabs accumulated radioactivity to maximum level 17 hours after the first radiogold

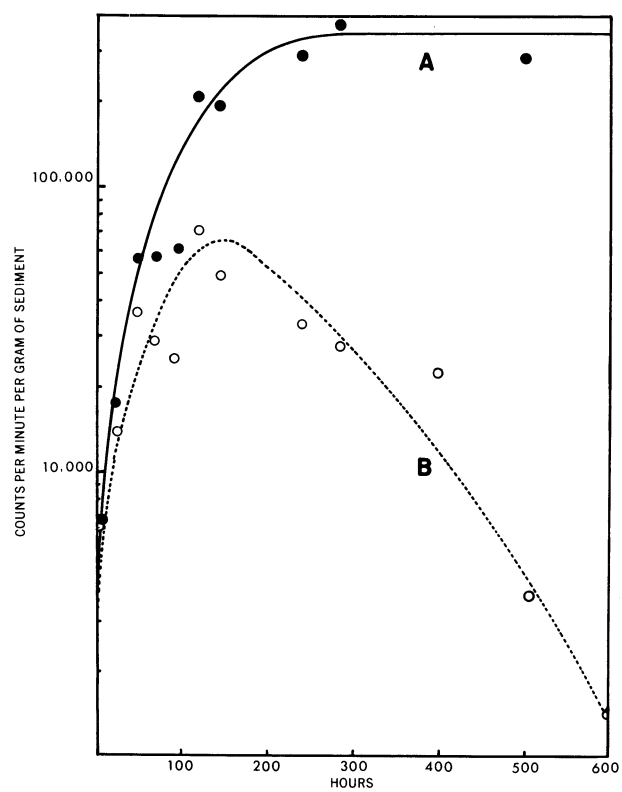

FIGURE 5.—Radioactivity content of biota and sediments from Cape Fear River after release of gold tracer.

application (fig. 5). However, this occurred 5 hours before the second application.

Background samples of biota and water collected in the drop zone area 1 week prior to the release of the labeled sediments did not contain measurable gamma activity in the energy range between 100 and 400 kiloelectron volts (Kev), the setting which includes the photopeak of radioactive gold. However, sediment samples contained gamma activity ranging from 367 to 729 counts per minute per kg. wet weight at this setting. Biota, water, and sediments collected from the same area 41 hours after the release did not show an increase in radioactive content over the background samples.

TABLE 5.—*Radioactive gold content of field samples* [1]

[Activity m.μc/kg.]

Elapsed time in hours	Sample	Station					
		I	II	III	IV	V	VI
1	Water	—	6.8	—	—	—	—
	Sediment	—	NS	—	—	—	—
	Fish	—	NS	—	—	—	—
	Oyster	—	23.8	—	—	—	—
	Clay	—	45.6	—	—	—	—
	Crab	—	27.3	—	—	—	—
17	Water	5.1	4.7	2.96	4.2	4.12	NS
	Sediment	NS	60.3	NS	—	NS	—
	Fish	NS	NS	NS	—	36.90	—
	Oyster	NS	70.9	NS	—	40.22	—
	Clay	NS	—	—	—	—	—
	Crab	NS	61.6	NS	—	—	—
41	Water	NS	NS	NS	NS	NS	NS
	Sediment	NS	NS	NS	NS	NS	NS
	Fish	NS	NS	NS	NS	NS	NS
	Oyster	15.7	31.1	NS	24.5	14.1	NS
	Clay	—	NS	—	—	—	—
	Crab	14.8	22.9	NS	24.9	19.0	—
144	Water	—	—	—	NS	—	NS
	Sediment	—	—	—	NS	—	NS
	Fish	—	—	—	NS	—	NS
	Oyster	—	—	—	NS	—	NS
	Crab	—	—	—	NS	—	NS

[1] NS indicates activity not significantly greater than background; dash indicates no sample was taken.

DISCUSSION

The potential hazards accompanying the release of radioactive material into the marine environment should be investigated so that the most dangerous situation possible is evaluated. The accumulation of radioactive gold from sea water by the animal community contained in the large tank was a simulation of this extreme situation: The closed system offered no opportunity for the animals to escape the contaminated area, and there was neither inflow of nonactive sea water nor outflow of contaminated water. Under these controlled conditions, organisms and sediments accumulated the isotope from sea water, with crabs and filter-feeding organisms accumulating the isotope to the greatest extent.

Surface sorption of radioactive gold, no doubt, is an important factor contributing to rapid accumulation by marine organisms and sediments. This is based on the observation that 50 percent of the isotope added to the water in an experimental environment moved very quickly to organisms and sediments within the tank. It is known that biological accumulation of a radiosotope by sorption occurs rapidly. Further, it has been shown that gold occurs in sea water in a particulate state. Since particles have difficulty passing through biological membranes, surface sorption is most important in their accumulation by marine organisms. The importance of surface sorption in the accumulation of radioactive gold by organisms was demonstrated experimentally when the radioactive content of clams buried in sediment was shown to be less than that of clams resting on the smooth bottom surface of the tank. Although the buried clams had less area exposed, the same average amount of water should have passed through their siphons as through the siphons of those resting on the bottom. If all the accumulation of gold had been a result of metabolism, the gold content of buried and exposed organisms should have been the same.

The literature contains several references pertaining to the surface accumulation of material from sea water. As early as 1937, Harvey demonstrated that particulate matter such as ferric hydroxide was associated with phytoplankton cells by electrostatic attraction to external surfaces. Also, Goldberg (1952) observed that particulate and colloidal iron was utilized by *Asterionella japonica*. Rice and Willis (1959) showed that particulate cerium 144 became attached to *Nitzschia* cells as a result of particles and cells coming in contact with each other and sticking.

The distribution of this isotope within the animals was observed by placing large quantities of radioactive gold directly into the gut of several estuarine animals. Those organs associated with excretion retained more gold 199 than did others, suggesting that the gold was simply being excreted rather than being stored or utilized. The principal route of gold administered as an oral dose to mammals was reported by Spector (1956) to be directly from the alimentary tract to the feces with

little absorption along the way. In the present study, little, if any, radioactive gold was found in the organs of fish that were fed this isotope sorbed onto clay.

In field studies, animals and sediment maintained in cages in the drop zone sorbed little activity. This could have been caused by the tremendous dilution by river water and strong currents, and to the short physical half-life of the gold isotope. Since it was demonstrated experimentally that the animals retained little activity from ingested labeled sediment particles, the initial accumulation of activity was perhaps from unbound gold that was not sorbed onto sediments in the mixing hopper but remained in the water phase of the slurry.

Since gold is not biologically essential and is not concentrated significantly by estuarine animals, the isotope gold 198, with its short physical half-life of 2.7 days, appeared to be a safe and effective tracer for following sediment movement. Even though as much as 5 c. of gold 198 were released in the Cape Fear River at one time, the maximum concentration found in any of the animals tested was 70.9×10^{-5} μc. per g. of oyster at station II (table 5). This is slightly in excess of the maximum permissible concentration (MPC) for gold 198 in water effluent released in an unrestricted area, i.e., 5×10^{-5} μc./ml. (Code of Federal Regulations, 1960). However, station II was purposely located in the drop zone so that the caged animals would be subjected to the most extreme conditions of contamination. There is no evidence now that radiation from the low levels of gold 198 involved in these investigations affected the biota.

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LITERATURE CITED

CODE OF FEDERAL REGULATIONS.
1960. Title 10—Atomic Energy. Part 20—Standards for protection against radiation. Federal Register, November 17, 11 pp.

FUKAI, RINNOSUKE, AND W. W. MEINKE.
1962. Activation analyses of vanadium, arsenic, molybdenum, tungsten, rhenium and gold in marine organisms. Limnology and Oceanography, vol. 7, pp. 186–200.

GOLDBERG, E. D.
1952. Iron assimilation by marine diatoms. Biological Bulletin, vol. 102, pp. 243–248.

GRAY, I. E.
1957. A comparative study of the gill area of crabs. Biological Bulletin, vol. 112, pp. 34–42.

GRIM, R. E.
1953. Clay mineralogy. McGraw-Hill Book Company, Inc., New York, N.Y., 384 pp.

HARVEY, H. W.
1937. The supply of iron to diatoms. Journal of the Marine Biological Association of the United Kingdom, vol. 22, pp. 205–219.

KRONE, R. B.
1959. Silt transport studies utilizing radioisotopes. Second Annual Progress Report. University of California, Hydraulic Engineering Laboratory and Sanitary Engineering Laboratory, 123 pp.

KRONE, R. B.
1960. Methods for tracing estuarial sediment transport processes. Report for San Francisco District Corps of Engineers, U.S. Army. Contract No. DA-04-203 Civeng-59-99, 57 pp.

NODDACK, IDA, AND WALTER NODDACK.
1939. Die Häufigkeiten der Schwermetalle in Meerestieren. Arkiv för Zoologi, band 32 A, N:o 4, pp. 1–35.

RICE, T. R., AND V. M. WILLIS.
1959. Uptake, accumulation, and loss of radioactive cerium 144 by marine planktonic algae. Limnology and Oceanography, vol. 4, pp. 277–290.

SPECTOR, W. S. (Editor).
1956. Handbook of biological data. W. B. Saunders Co., Philadelphia, Pa., 584 pp.

SCHWEITZER, G. K., AND MORRISON JACKSON.
1952. Radiocolloids. Journal of Chemical Education, vol. 29, pp. 513–522.

SCHWEITZER, G. K., AND W. N. BISHOP.
1953. Low concentration chemistry. VI. Some properties of tracer gold in solution. Journal of the American Chemical Society, vol. 75, pp. 6330–6332.

U.S. ARMY CORPS OF ENGINEERS.
1964. Radioactive sediment tracer tests, Cape Fear River, N.C. U.S. Army Engineer Waterways Experiment Station, Miscellaneous Paper No. 2-649, 14 pp.

VINOGRADOV, A. P.
1935–44. The elementary chemical composition of marine organisms (Khimicheskiĭ elementarnyĭ sostav organizmov morya). In three parts. Akademiya Nauk SSSR. Trudy Biogeokhimicheskoĭ Laboratorii. Part I, in vol. 3, 1935, pp. 63–278; Part II, in vol. 4, 1937, pp. 5–225; Part III in vol. 6, 1944, pp. 5–273. (Translation by Julia Efron and Jane K. Setlow, with bibliography edited and newly enlarged by Virginia W. Odum, Sears Foundation for Marine Research (New Haven, Conn.), Memoir Number 2, 1953, 647 pp.).