Levels of $^{65}$Zn in water, sediments, plants, and animals in Alder Slough, a small ecosystem in the Columbia River Estuary, were determined periodically during a one year period. Concentrations of total Zn in organisms were also measured to permit computation of specific activities ($\mu$Ci $^{65}$Zn/g total Zn). Temporal fluctuations in the activities of $^{65}$Zn and the concentrations of total Zn occurred in all components of the ecosystem.

Activities of $^{65}$Zn and concentrations of total Zn varied among individual fish from the study area. Pooled estimates of population standard deviations of these values in all samples of two fish species taken during the year amounted to about 20% of their grand means. Usually, individuals with high $^{65}$Zn activities also proved to have high total Zn concentrations, hence specific activities were less
variable than either $^{65}$Zn or total Zn.

The nuclear reactors at Hanford, Washington, the source of most of the $^{65}$Zn in the Columbia River, were shut down for approximately 45 days during this study. This event presented the opportunity to investigate the response of various components of the ecosystem to reduced $^{65}$Zn input. The rates of $^{65}$Zn specific activity decline in several organisms were measured following reactor shutdown. The time required for specific activity of an organism to be reduced by one-half under the conditions prevailing is termed "ecological half-life". The "ecological half-life" differs from the biological half-life of an organism because:

1. it is defined in terms of specific activity
2. it reflects the continued addition of radioactivity to the organism from its food web and water.

"Ecological half-lives" vary with the trophic levels of the organisms and with changes in prevailing ecological conditions.
Radioecology of $^{65}$Zn in an Arm of the Columbia River Estuary

by

William Charles Renfro

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Radioecology of $^{65}$Zn in an Arm of the Columbia River Estuary

INTRODUCTION

Radioecology is the study of the fate of radioactive substances released into the environment and the manner by which ecological communities modify the distribution of the radioactivity (Odum, 1959). An abbreviated definition by Polikarpov (1966) states that marine radioecology is the science of the interrelation between a radioactive environment and marine organisms. Both authors distinguish two facets of the discipline, namely, radiocontamination of the environment and ecological consequences of ionizing radiations. This thesis deals with the first facet, the distribution and dynamics of a single radionuclide in a small estuarine area.

Sources of Environmental Radioactivity

There are three basic types of radioactive substances: naturally occurring radionuclides, cosmic ray-induced radionuclides, and artificial radionuclides. All elements with atomic numbers greater than 83 occurring in nature are radioactive (Friedlander, Kennedy and Miller, 1964). These naturally occurring radionuclides belong to one of three decay chains that decay successively by emitting alpha and/or beta particles. In each chain the unstable parent radionuclide, $^{238}$U, $^{235}$U, or $^{232}$Th, has such a long half-life that it has not
decayed completely since the origin of the earth some $4.5 \times 10^9$ years ago. Other radioactive elements which occur naturally are $^{40}$K, $^{50}$V, $^{87}$Rb, and a number of heavier elements with half-lives longer than $10^9$ years.

Cosmic rays bombarding the earth are believed to originate in outer space and to be accelerated in interstellar magnetic fields (Friedlander et al., 1964). The nuclear reaction of these high energy cosmic rays with elements in and above the earth's atmosphere produce radionuclides such as $^3$H, $^7$Be, and $^{14}$C which add to the background radiation of the biosphere.

The third source of environmental radioactivity results from man's use of nuclear energy. Prior to World War II the world inventory of artificially produced radionuclides was very small. These radionuclides were mostly produced in minute quantities by nuclear reactions of various elements with particles emitted from naturally occurring radioactive elements or particles accelerated in such devices as cyclotrons and linear accelerators. With the advent of the atomic bomb explosions during World War II the presence of artificial radionuclides in the biosphere emerged as a problem of worldwide concern.

**Artificial Radioactivity in the Biosphere**

Since the first explosion of a nuclear device in New Mexico
on 16 July 1945, there has been an ever-increasing quantity of artificial radionuclides on the earth and in its atmosphere. The number of nuclear explosions conducted by various nations through 1961 totaled 306 (Eisenbud, 1963), although some of these produced comparatively small amounts of radioactivity. One result of these explosions has been to increase the overall radiation levels throughout the biosphere, by the introduction of radioactive fission products and neutron-induced radionuclides.

In the quarter-century since 1942, when the first crude nuclear reactor was successfully operated at the University of Chicago, the use of nuclear energy has steadily increased. Atomic energy is now replacing fossil fuels to generate electricity and to propel ocean vessels. Radioactive wastes can now be contained and the bookkeeping cost of nuclear power has become competitive (Abelson, 1965). The uses to which nuclear energy may be put are innumerable. Besides the production of steam to drive turbines for propulsion of ocean vessels or generation of electricity, it now appears economically feasible in some areas to produce large volumes of freshwater from sea water in large steam-electric plants using nuclear energy. Such dual purpose installations may provide the means to make vast regions of the earth previously unsuitable for habitation fertile. In addition, nuclear power reactors do not pollute the atmosphere with carbon dioxide and other contaminants
associated with the combustion of fossil fuels.

Utilization of nuclear energy is not without problems, however. Consider, for example, the operation for one year of a nuclear power reactor in generating the 20 megawatts of electricity for a typical city of 50,000 population. At an operating efficiency of 20% such a reactor would require a heat output of 100 megawatts and would yield some 14 megacuries of fission product activity after 100 days decay. Assuming these fission products were in the form of liquid wastes at a concentration of one curie per milliliter, the volume of wastes would amount to 14 cubic meters. If this waste could be uniformly mixed throughout all the world ocean (volume about $1.4 \times 10^{18}$ cubic meters), each cubic meter of seawater would have its radioactivity temporarily increased by one picocurie. Clearly, such an answer to the problems of fission product disposal is out of the question and thus present practice is to concentrate and contain these wastes.

Radioactive contaminants other than fission products are also produced in nuclear power reactors. In reactors cooled by water, radioactive products may originate from: a) neutron activation of impurities present in the water, b) activation of hydrogen atoms of the water itself, c) activation of corrosion products of reactor components, and d) escape into the cooling water of fuel and fission products from the fuel elements (Moeller, 1957). In the case of gas-cooled reactors, certain impurities and corrosion products in the
cooling gases may likewise be made radioactive by neutron activation. The design of the reactor will, in most instances, determine the kind and amount of radionuclides released to the environment.

Because nuclear power looms so large in the future of the world, it is crucial that the ecological impact of reactor installations be thoroughly investigated. Radioecological studies in estuaries are particularly important for several reasons. First, most streams with their loads of sediments, domestic wastes, and industrial contaminants (including radionuclides) lead to the sea via estuaries where various processes detain and often concentrate these pollutants. Consequently, estuaries may receive radioactive wastes produced at nuclear installations far upstream. Second, the steadily increasing demands on limited supplies of fresh water make the brackish waters of estuaries attractive for condensing and other industrial uses (Picton, 1960). Finally, radioactive wastes entering estuaries may cycle and recycle until they enter human food supplies in significant quantities (Cronin, 1967).

Thermal Pollution

Thermal pollution of estuaries by nuclear and fossil fuel power plants is another critical ecological problem for environmental scientists. The thermal efficiency of steam turbines is increased by condensing the steam on the exhaust side of the turbines causing a
partial vacuum (Warinner and Brehmer, 1966). This raises the temperature of the cooling waters which are usually taken from a body of water near the plant. To date nuclear powered steam-electric plants have proved less efficient than fossil fuel plants (Mihursky and Kennedy, 1967). This means that the nuclear installation must expend more energy and generate more heat to produce a kilowatt of electricity. As a consequence, the nuclear plant gives up more heat to the condenser waters returning to the environment. The study of ecological effects of nuclear power installations therefore must deal with varying degrees of heat loading as well as the possible presence of radioactive contaminants. A first important step therefore is to study the ways in which radionuclides are acted upon by various components of natural environments not subjected to thermal pollution.

Columbia River Radioactivity

The Columbia River Estuary has a number of characteristics which make it particularly valuable as a site for radioecological research. The area is large and presents many different habitat types and kinds of organisms for study. Within the estuary can be found areas varying from freshwater streams to brackish tidal flats and coastal marine conditions. Because of its great size and relatively small human population, the Columbia River basin has not suffered
so much from domestic and industrial pollution as have many other river basins in the United States. Most important is its continued exposure to trace concentrations of a number of radioactive elements.

For 23 years nuclear reactors at Hanford, Washington have released large quantities of radionuclides in dilute concentrations into the Columbia River. The primary purpose of these production reactors is to provide neutrons to convert fertile material into fissionable material (Glasstone, 1958). A fertile material is an isotope which can be converted into a fissionable isotope. In the Hanford reactors $^{238}\text{U}$, a fertile isotope comprising about 99.3% of naturally occurring uranium, is converted into fissionable $^{239}\text{Pu}$ by the following nuclear reactions:

$$
{^{238}}_{92}\text{U} + {^{1}}_{0}\text{n} \rightarrow {^{239}}_{92}\text{U} \rightarrow {^{239}}_{93}\text{Np} + \beta^{-} \quad (23.5 \text{ mins.})
$$

$$
{^{239}}_{93}\text{Np} \rightarrow {^{239}}_{94}\text{Pu} + \beta^{-} \quad (2.33 \text{ days})
$$

The $^{239}\text{Pu}$, after chemical extraction and processing, serves as fuel for nuclear weapons and nuclear reactors.

The controlled nuclear reactions taking place within the Hanford reactors which provide the intense neutron fluxes necessary to produce the conversion of $^{238}\text{U}$ to $^{239}\text{U}$ also generate large amounts of heat. This heat is dissipated by a "single pass" cooling process.
in which Columbia River water is passed through the reactors and returned to the river. Neutron activation of corrosion products and other impurities in this cooling water produces most of the radio-nuclides present in the river, estuary, and adjacent Pacific Ocean. Lesser amounts of radioactive elements in the river result from fallout of radionuclides from nuclear explosion tests into the drainage system and fission of very small quantities of uranium in the cooling waters. In addition, fuel element jackets occasionally fail releasing small amounts of fission products and activation products to the cooling water (Nelson, 1962). Perkins, Nelson and Haushild (1966) list the following 11 gamma emitting nuclides as being among those occurring in measurable amounts in the river between Hanford and the ocean: \(^{46}\text{Sc}, ^{51}\text{Cr}, ^{58}\text{Co}, ^{59}\text{Fe}, ^{60}\text{Co}, ^{65}\text{Zn}, ^{95}\text{Zr}, ^{95}\text{Nb}, ^{106}\text{Ru}, ^{124}\text{Sb}, \text{and} ^{140}\text{Ba}.\) In the course of their transport down the river the amounts of these radionuclides may be depleted by one or more of the following processes: 1) radioactive decay, 2) accumulation by organisms, and 3) sorption to suspended sediments followed by deposition of the sediments (Osterberg, 1963).

**Objectives**

The purpose of this study is to examine some aspects of the distribution of the neutron-induced radionuclide \(^{65}\text{Zn} \) in the water, sediments, and biota of a small estuarine area. Specific objectives
of the work are threefold:

1. To determine the levels and temporal fluctuations of $^{65}$Zn in several components of the environment.

2. To examine variations in $^{65}$Zn body burdens among comparable individual organisms.

3. To attempt quantitative estimates of turnover rates of $^{65}$Zn by several estuarine organisms.

This approach is influenced by the research of Foster, who after more than 20 years of radioecological studies of the Columbia River, stated:

However, when we are faced with new practical problems of applying concentration factors in order to estimate the level of specific nuclides that may result in fish as a consequence of known amounts in the water, we find that the best available information is rarely the product of controlled laboratory experimentation. Rather, the most usable information stems from field surveys made near the major atomic energy installations or in the Pacific Test area. The conclusion is obvious- we need more experiments designed to simulate closely field conditions and also greater exploitation of the data that can be derived from existing conditions (Foster and McConnon, 1965, p. 216).

Field work began in early April 1966 and continues as a part of radioecological studies by the Oregon State University Department of Oceanography. This thesis deals with research carried out during the period April 1966 through March 1967. Three months after the work began an event unique in the radioecological history of the Columbia River occurred. About 9 July 1966 all the reactors at
Hanford were shut down because of labor-management disagreements. For a period of about 45 days the reactors remained inoperative and during this time the input of $^{65}\text{Zn}$ to the estuary was drastically curtailed. More accurate times cannot be given because the processes involved in shutting down and resuming operations of the six reactors occupied several days. Although this unexpected circumstance forced certain changes in my original objectives and procedures, it presented an unparalleled opportunity to study the response of the Alder Slough system to such an alteration.
STUDY AREA

The Columbia River Estuary is a vast area of extreme physical complexity complicated by such large forces as diurnal tides, varying river discharge, storm waves, and littoral drift (Lockett, 1962). It is an area where the fresh waters with their organisms and elements previously influenced primarily by terrestrial processes meet and mix with oceanic waters having quite different chemical and biological characteristics. Clearly, it is not practical to investigate the radioecology of such a large and complex system in its entirety. Therefore, it seems expedient to select a discrete segment of the estuary within which the biota can be studied and the effects of tides, salinity changes, and other environmental variables can be assessed. Among the prerequisites for such a study area are the following: a) a location accessible both by land and water, b) a body of water with depths shallow enough to permit sampling with a seine, but deep enough to retain water even during lowest tides, c) an area not recently disturbed by man, d) a protected site of relative calm even during storm conditions.

Actually, few sites along the Oregon shore satisfy all these requirements. Most locations in the lower portion of the estuary which are readily accessible by land and water have long been occupied by fish canneries and other commercial establishments.
Further, much of the remaining shoreline has been recently filled or otherwise modified. It is necessary therefore to compromise on a site which could be reached by road but is accessible by boat only during high tides.

Alder Slough, the location chosen, satisfies most of the original requirements and in other respects proves satisfactory. The slough is an "L-shaped" body of water about 1 km south of Tansy Point and 6 km upriver from the mouth of the Columbia River (Figure 1). The base of the "L" parallels a levee extending approximately 3 m above mean sea level (MSL) which is designed to prevent inundation of an adjacent low-lying area during flood stages on the Columbia River or during storm tides in the nearby Pacific Ocean. Such a levee system must provide an egress for streams draining land behind the levee. This is accomplished by installing "tide gates" which are culverts fitted at the estuary end with heavy top-hinged cast iron doors. These doors act as one way valves. During high tides in the estuary the force of water presses the doors shut preventing the intrusion of estuarine water into the stream and causing the water level on the stream side to rise. When the tide falls below the level of the stream the doors are pushed open allowing the accumulated creek water to empty into the estuary.

At the end of Alder Slough near the tip of the "L" (Figure 2) a tidal gate system having five doors was constructed in 1937. This
Figure 1. The Columbia River Estuary showing the location of Alder Slough.
Figure 2. The morphometry of Alder Slough.
system allows the waters of Alder Creek to flow through the levee into the slough.

The shape of the slough changes with the height of the tide. At high tide much of the area is covered so that the sampling site takes on the appearance of an indentation from the adjacent tidal flat. During low tides the pools at the elbow and tip of the "L" are connected by a shallow channel 15 m wide. The base of the "L" is approximately 80 m long and the length of the upper arm is about 100 m. Even during lowest water levels the slough is connected by a shallow channel winding across the tidal flat and running through a sand bar into the Columbia River.

Over most of the slough the sediments are quite soft so that a man walking on them often sinks to his knees. However, in the channel connecting the two pools at the base of the "L" there is often a rapid stream flow at low tide. This flow of water periodically scours the bottom of lighter sediments and consequently the bottom in this channel is comparatively firm facilitating seining operations. For this reason, all fish and shrimp for radioanalysis were collected in this area.

Around the edges of the slough at elevations inundated only during highest tides there is dense rooted vegetation. Most of this vegetation consists of the cattail, *Typha latifolia*, and the sedges, *Scirpus* spp. and *Carex* spp. Except in late winter and early spring,
dense stands of Scirpus and Carex are also present at elevations of -1.0 m MSL and above.

Tides in Alder Slough follow the semi-diurnal rhythm of the Pacific Ocean usually lagging about one-half hour behind the levels experienced at the mouth of the river. Although there is usually good correlation between observed tidal heights and those predicted by the tide tables, strong, sustained winds and changes in flow rates of the Columbia River sometimes upset the predictions. The average daily tidal range is approximately 2.5 m although the range during a 24 hour period varies from 1.86 to 3.51 m during the year (U.S. Coast and Geodetic Survey, 1966).

Water levels in the slough were measured against points on the tide gate of known elevation above (geodetic) mean sea level. Although tidal levels vary with seasonal changes in Columbia River flow, observations during this study suggest that throughout the year the levels oscillate about a plane -0.8 m MSL. This discrepancy results from the fact that local mean sea level differs from geodetic mean sea level.

With each tidal cycle there is an exchange of water in Alder Slough with water of the tidal flat and Columbia River. At high tide (water level +0.5 m MSL) approximately 17,200 m$^3$ of water are contained in the system. Low tide (-2.0 m MSL) volume amounts to about 4,200 m$^3$. Hence, the volume of water entering and leaving
the slough in the interval between consecutive lower low tides and higher high tides approximates $13,000 \text{ m}^3$ or about 75% of the high tide volume. In addition, water from Alder Creek flows into the slough at low tides producing perceptible currents even during seasons of low runoff in the creek.

Despite the large tidal exchange which tends to renew a portion of slough water each day, salinity stratification is evident throughout the year. Figure 3, showing salinities at the sampling station, indicates that water near the bottom is more saline (on the average about 30% higher in salinity) than the surface layer. The presence of this saline water layer near the bottom provides a medium suitable for brackish water fishes which may require at least small concentrations of dissolved salts in their environment. Also, it suggests that these lower layers experience somewhat slower tidal exchange rates than do the upper layers.

Water from Alder Creek plays a dominant role in determining the chemistry and hydrology of the slough. Alder Creek drains a low, marshy area roughly $5.4 \text{ km}^2$. Much of this area consists of peat marsh and alder thickets imparting to the water a dark tan color even during seasons of high creek discharge. Because the surface layer in Alder Slough has this dark coloration at all times, it is concluded that the creek waters exert a strong influence on the hydrology of the slough.
Figure 3. Alder Slough environmental conditions.
Water temperatures measured near the bottom in the channel connecting the pools at the tip and base of the "L" are graphed in Figure 3. During the spring there is a steady increase in bottom temperatures. Values above 15°C occur from June through September followed by a steady decline during the remainder of the study. In general, daytime surface temperatures are higher than bottom temperatures during the spring and summer but are lower during fall and winter.

Seasonal changes in the discharge rate of the Columbia River, shown in Figure 3, influence the water levels and salinities in Alder Slough. Greatest discharge occurs in June when melting of snow accumulated in the upper regions of the drainage basin reaches its peak. During this summer period salinities in the estuary are lowest and the mean tidal level is highest. Following the summer peak, the flow rate of the Columbia River is greatly reduced. This causes the estuary to become a partly mixed system (Burt and McAlister, 1959) resulting in increased salinities in the study area.
METHODS AND MATERIALS

Fishes and invertebrates were collected by seine during low tides from 8 April 1966 to 15 March 1967. All collections employed the same bag seine which was 15 m long and 1 m high. This seine, designed to sink so that its lower edge drags bottom, was constructed of nylon material having a bar mesh size of 6 mm in the wings and 3 mm in the bag. It regularly caught small fish and mysid shrimp as short as 6 mm long. At the start of each seining operation the seine was stretched full length along the bank, then pulled directly across the 15 m channel connecting the two pools at the elbow and tip of the "L". All organisms caught were immediately removed and preserved in 10% formalin solution.

Amphipods (Corophium salmonis) were the only animals collected during this study which were not taken with nets. These small organisms live in cylindrical tubes attached to the sediments or to objects on the bottom. To collect them, small rocks and sticks were picked from the bottom and plunged into a container filled with a dilute formalin solution. After several minutes the amphipods left their tubes and upon dying settled to the bottom of the container. In the laboratory the amphipods were picked out individually with jeweller's forceps until a sufficient number (usually 1,000 or more) for radioanalysis was accumulated.
Samples of algae and rooted vegetation were collected by hand and returned unpreserved to the laboratory. The algae were scraped from the vertical concrete wall of the tide gate. Each sample of algae was taken from the same general area of the wall.

Sediment samples were taken by removing the top centimeter from a small area of the bottom near the tide gate. Before radioanalyzing the sediments, vegetation and other debris were removed with forceps.

To measure the concentration of $^{65}$Zn in the water, the method of co-precipitation described by Osterberg, Cutshall, and Cronin (1965) was employed. Eight liter samples of unfiltered water were brought to pH 7 by addition of NH$_4$OH, then carriers and FeCl$_3$ were added. After mixing for several minutes, NH$_4$OH was again added to bring the pH to about 9 producing a dense flocculent Fe(OH)$_3$ precipitate. The precipitate was removed from the solution by filtering onto a Whatman number 42 filter paper which was then placed in a plastic counting tube for radioanalysis. In this way, the activity of $^{65}$Zn present in the water in both ionic and particulate forms was determined.

All organisms were identified and prepared for radioanalysis as soon after collection as possible. In every case, the sample was washed in tap water to rid it of mud and detritus. Individual fish were dissected and their stomach content removed before radioanalysis.
For most samples wet weight, dry weight, and ash-free dry weight measurements were obtained. Wet weights were obtained by blotting the samples on paper towels then weighing them in preweighed porcelain crucibles. After drying at 60°C in a forced-air oven, the crucible plus dry sample were weighed and returned to the oven. This process was continued until a constant dry weight was obtained. After radioanalysis the dry sample was again transferred to a preweighed crucible, weighed, and ashed in a muffle furnace at 550°C until the ash was white. The crucible plus ash were weighed to obtain ash weight and this value subtracted from dry weight yielded the ash-free dry weight.

All samples were radioanalyzed by gamma ray spectrometry, utilizing a 5 x 5-inch NaI (Tl) well crystal coupled through a photomultiplier to a 512 multichannel analyzer. The sample, contained in a plastic tube, was placed in the well of the detector crystal. Gamma rays emitted from unstable atoms in the sample interact with molecules of the crystal causing these molecules to scintillate. The intensities of these scintillations, which are proportional to the energy of the gamma photons interacting with the crystal, are sensed and converted to pulses of electrical current by the photomultiplier. These current pulses are in turn measured and recorded in the multichannel analyzer. Readout of this stored information is obtained in digital form on punched tape and by typewriter or in analog form on
an X-Y plot. The result is a spectrum relating the energy of the gamma photon with its frequency of occurrence.

Since each gamma emitting radionuclide emits photons of characteristic energy, the radionuclides in the sample can be identified by analyzing the gamma-ray spectrum. Often a sample contains several gamma emitters having characteristic photons of different energies. In this case the spectrum exhibits a series of peaks and valleys, each peak corresponding to a specific energy interaction in the detector crystal. When a photon of a certain energy interacts with the detector crystal, it may impart all or only a part of its energy to the crystal. If all the energy of the photon is imparted to the crystal (photoelectric effect), the event is recorded by the analyzer in the photopeak energy channel characteristic for that radionuclide. However, if only a portion of the total energy of the photon interacts with the crystal (Compton interaction) to produce a scintillation, the event will be registered in a lower energy channel. Compton interactions of higher energy photons may thus be recorded in the photopeak channels of radionuclides emitting photons of lower energy. In this case, it is necessary to make appropriate corrections when determining the activities of the lower energy gamma emitters.

Spectrum analyses to determine the activity of $^{65}$Zn in Alder Slough samples were facilitated by the fact that $^{65}$Zn was usually the only radionuclide having a photopeak with an energy greater than 1.0 million electron volts (Mev). Hence the activity of $^{65}$Zn in such
a sample could be obtained simply by summing the number of counts recorded in the photopeak energy channels and comparing with a similar sum from a standard of known $^{65}$Zn activity. In algae and sediment samples which contained radionuclides with characteristic photopeaks of energies higher than $^{65}$Zn, a computer program developed by Frederick (1967) was employed to calculate $^{65}$Zn activity.

With few exceptions, the samples were radioanalyzed for 100 minutes. The activity per unit weight of $^{65}$Zn in each sample is expressed as picocuries per gram dry weight of sample (pCi/g). In every case, the activity per unit weight of $^{65}$Zn in the sample has been corrected for physical decay occurring after collection. For brevity, activity per unit weight is shortened to "level" or "activity" in the remainder of the text.

Total Zn (radioactive plus stable Zn) in the samples was determined by an atomic absorption spectrophotometer. This device determines the concentration of Zn (or other elements) in a solution by measuring the amount of electromagnetic energy absorbed by the solution as it is atomized into a flame. A lamp with a Zn filament emits light of a characteristic wavelength. A beam of this light is directed through a flame to a photodetector. The sample solution is atomized into the flame and the light is absorbed by atoms of Zn in the flame. The photodetector then measures the change in intensity of the light beam due to absorption which is proportional to the
concentration of Zn in the solution. Concentrations of total Zn
expressed in micrograms Zn per gram dry weight (μg/g) are deter-
mined by comparing absorbances of the sample to those of standards
of accurately known Zn concentration.
RESULTS AND DISCUSSION

The Alder Slough Ecosystem

An ecosystem may be defined as "any area of nature that includes living organisms and nonliving substances interacting to produce an exchange of materials between the living and nonliving parts..." (Odum, 1959). This concept can be used as a model in the broadest sense to visualize the relationships between the various biotic and abiotic components of an area in nature. It may be applied to systems as small as temporary ponds or as large as oceans. Although the physical size of an ecosystem is not a measure of its complexity, it is often advantageous to study an ecosystem of small size, if for no other reason than ease of sampling.

With a total area approximating one city block, Alder Slough is small enough to be sampled and studied effectively. The dominant macroscopic plants and animals are relatively few in number, so a general description of the ecosystem can be made. Broadly speaking, there are three major components: water, sediments, and biota. Between each of these components there is a constant flux of $^{65}$Zn, the material of interest in this study. Table 1 lists these components, including a partial catalog of the more prominent plants and animals. No attempt was made to collect or identify all the biota.
<table>
<thead>
<tr>
<th>Table 1. Major Alder Slough ecosystem components.</th>
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<tr>
<td><strong>Water</strong></td>
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<tr>
<td><strong>Sediments</strong></td>
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<td><strong>Plants</strong></td>
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<tr>
<td><strong>Chlorophyta</strong></td>
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<tr>
<td><em>Enteromorpha intestinalis</em></td>
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<td><strong>Tracheophyta</strong></td>
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<tr>
<td><em>Typha latifolia</em></td>
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<tr>
<td><em>Carex spp.</em></td>
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<tr>
<td><em>Scirpus spp.</em></td>
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<tr>
<td><strong>Animals</strong></td>
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<tr>
<td><strong>Arthropoda</strong></td>
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<tr>
<td><em>Neomysis mercedis</em></td>
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<tr>
<td><em>Corophium salmonis</em></td>
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<tr>
<td><em>Anisogammarus conferviclus</em></td>
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<td><em>Crangon franciscorum</em></td>
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<td><em>Pacifastacus trowbridgii</em></td>
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<tr>
<td><em>Gnorimosphaeroma oregonensis</em></td>
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<td><strong>Chordata</strong></td>
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<tr>
<td><em>Oncorhynchus tshawytscha</em></td>
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<tr>
<td><em>Cyprinus carpio</em></td>
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<td><em>Mylocheilus caurinus</em></td>
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<tr>
<td><em>Microgadus proximus</em></td>
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<td><em>Gasterosteus aculeatus</em></td>
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<tr>
<td><em>Micropterus salmoides</em></td>
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<tr>
<td><em>Perca flavescens</em></td>
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<td><em>Cymatogaster aggregata</em></td>
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<td><em>Cottus asper</em></td>
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<td><em>Leptocottus armatus</em></td>
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<td><em>Platichthys stellatus</em></td>
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For example, a large benthic infauna undoubtedly present was not sampled. Similarly, zooplankton and phytoplankton populations were not studied although they probably greatly influenced the distribution of $^{65}\text{Zn}$ in the ecosystem.

The sections which follow attempt to describe the role of the major ecosystem components in distributing $^{65}\text{Zn}$ in Alder Slough. From the biota several of the more important organisms are selected for detailed examination. Particular attention is given to the response of these components to reduced input of $^{65}\text{Zn}$ to the ecosystem after shutdown of the Hanford reactors.

**Water**

Water is the primary agent for the transport of $^{65}\text{Zn}$ from the Hanford reactors to the Columbia River Estuary. Because Zn has a marked tendency to become associated with particles, the percentage of $^{65}\text{Zn}$ atoms in the particulate form steadily increases during its passage downriver. According to Perkins, Nelson and Haushild (1966), $^{65}\text{Zn}$ in reactor effluent is mostly cationic (98% in February 1965). During 1964 the annual average proportion of $^{65}\text{Zn}$ in the particulate phase at Pasco, Washington, 70 km downstream from the reactors, was about 30% and at Vancouver, Washington, 420 km downstream, the proportion had increased to about 80% (Perkins et al., 1966). These authors also showed that the $^{65}\text{Zn}$ activity per liter of river water underwent a tenfold depletion between Pasco and Vancouver during most of 1964. Some of the loss of $^{65}\text{Zn}$ activity
from the river water can be ascribed to biological activity, but deposition of particles associated with $^{65}$Zn must also be an important loss mechanism. However, during periods of peak river discharge in May and June there is a large increase in activity at Vancouver due to scouring of riverbed sediments containing $^{65}$Zn. Presumably, these same processes are operating in Alder Slough to continually reapportion the activity of $^{65}$Zn between water and sediments.

In Figure 4 are shown activities of $^{65}$Zn in the water of Alder Slough. As previously stated, the Hanford reactors were shut down about 9 July and were not put back into operation until about 25 August. Columbia River water was allowed to pass through the inoperative reactors during this period. This flow of water continued to act on surfaces of the reactors causing the transport of neutron-activated corrosion products downriver and accounting for at least a portion of the $^{65}$Zn activity measured in the water after shutdown.

The first water sample from Alder Slough to be radioanalyzed was collected 22 July, about 13 days after reactor shutdown. According to Nelson, Perkins and Haushild (1966), about 10 to 12 days would be required for a parcel of water to travel from the reactors to Astoria under the prevailing river discharge rate (intermediate flow—about 8,000 m$^3$/sec.). Hanson (1967), in a study of the Columbia River Estuary concurrent with this work, estimated the
Figure 4. Activities of $^{65}$Zn in water and sediment. Standard deviations are indicated for sediment activities.
flow time to be 12 days. Assuming that $^{65}$Zn would be dispersed longitudinally in the course of its travel downriver and assuming also that at least several days were required for tidal exchange to reduce the level of $^{65}$Zn in the slough, it is probable that the $^{65}$Zn activity of the 22 July water sample had not been greatly influenced by reactor shutdown. However, water samples taken 4 August showed a reduction in $^{65}$Zn activity followed by marked decreases later in August. On this basis, it appears that the Alder Slough ecosystem first experienced the effects of reactor shutdown about late July.

The reactors resumed operations about 25 August during a period of low Columbia River flow (4,000 m$^3$/sec.). Under these conditions, the first newly activated $^{65}$Zn would require more than 18 days to reach Astoria (Nelson et al., 1966). Hanson (1967) estimated this time to be 19 days. In Alder Slough the $^{65}$Zn activity was low on 12 September (19 days after reactor start-up), but was slightly higher than the 30 August value. Based on these considerations, Alder Slough experienced diminished input of $^{65}$Zn from about 1 August through mid-September. After mid-September the $^{65}$Zn levels increased and remained at about 4 pCi/L. There was a further increase in $^{65}$Zn activity in December and January followed by a low $^{65}$Zn value in February caused by increased discharge of Alder Creek water into the slough.
Specific activities of $^{65}$Zn in the water samples are not available because total Zn concentrations were not determined.

**Sediments**

Waldichuk (1961) described sedimentation as a concentrating process in which dissolved and particulate material are abstracted from the water and deposited on the bottom. He lists the following processes associated with sedimentation: physico-chemical action of sorption, flocculation, ion exchange, precipitation, co-precipitation, and biological activity. Because sediments concentrate many trace elements, the rates of exchange of these elements between the sediments and water may affect their availability to the biota (Duke, Willis and Price, 1966).

The mechanisms by which $^{65}$Zn is held on the sediments determine rates of exchange. Johnson (1966) considered ion exchange, formation of insoluble precipitates, formation of complexes with organic matter, and specific sorption (including lattice penetration, isomorphous substitution, chemisorption, and formation of coordination complexes) as possible mechanisms for retention of $^{65}$Zn. In a series of experiments in which he eluted Columbia River sediments with seawater and dilute metal solutions, Johnson (1966) showed that some mechanism stronger than ion exchange is responsible for retaining $^{65}$Zn on the sediments in the presence of seawater.
Sediments in Alder Slough are predominantly dark silty sands. A small area of the bottom near the tide gate at an elevation of -2.0 m MSL was the site from which all sediment samples were obtained. Sediment from the top cm, analyzed by the pipette method of sediment size analysis (Krumbein and Pettijohn, 1938), consisted of 50% sand, 46% silt, and 4% clay. Below the first cm the sediments in the slough are black and reduced. When disturbed they release large amounts of hydrogen sulfide. A core sample was taken to examine the distribution of $^{65}$Zn at various depths below the surface. The $^{65}$Zn activity was $26.7 \pm 2.1$ pCi/g in the top cm, $0.4 \pm 0.4$ pCi/g at 10 cm, and below detectable limits at 20 cm. Because $^{65}$Zn was virtually absent in the dark, reduced sediments, only the the surface sediments were radioanalyzed thereafter.

The activities of $^{65}$Zn in sediment samples are graphed in Figure 4. Discounting some abrupt fluctuations, the levels of $^{65}$Zn are low from late July to mid-August followed by a sharp rise to high levels in late August. From these high levels there is a decline during September to intermediate levels in October and November. A second decline to a low activity in mid-January is followed by high levels in February and March. When smooth curves are drawn through the data points of both water and sediment activities, the curves are seen to be roughly mirror images. Why the $^{65}$Zn activity in the sediments increases as the activity of the water decreases
and vice versa is not apparent. It could be speculated that conditions favoring sedimentation of $^{65}$Zn from the water result in an increase in sediment activity and that conditions favorable to the release of $^{65}$Zn from the sediments cause the levels of $^{65}$Zn in the water to rise. However, an answer to the relationship must await a much more detailed study.

To date, techniques for the determination of total Zn concentrations in sediments have not been perfected. Therefore specific activities of $^{65}$Zn in sediments could not be obtained. As will be discussed in a later section, these values would have been very helpful in understanding $^{65}$Zn exchange between the sediments and animals that regularly ingest significant quantities of mud.

**Plants**

**Green Algae**

The green alga, *Enteromorpha intestinalis*, is an important component of the Alder Slough ecosystem. It is especially prominent on the concrete walls of the tide gate, hanging like a green curtain from the higher high tide mark to the subtidal zone. During periods of rapid growth, its thalli (fronds) appear as convoluted, soft tubes shading from bright to dark green.

In the mature plant there are four general regions (1) an
attaching holdfast, (2) a short stalk, (3) a terminal swarmer producing region, and (4) a long growing region between (2) and (3) (Ramanathan, 1939). These regions are not distinct, but grade into one another. The actively growing part of the plant is the region immediately above the stalk. In the terminal fertile region every cell forms swarmers that escape through an opening in the cell wall. After the swarmers escape, the empty thallus persists as a collapsed transparent ribbon but eventually breaks away.

There are three kinds of plants in this alga, male, female, and asexual. All grow together and are macroscopically indistinguishable from one another. Biflagellate gametes from a male and a female plant fuse to produce a zygote which immediately germinates to grow into a new plant. This new plant produces only zoospores that swarm for a while, settle on suitable substrate, and grow into new plants. The new plant derived from the zoospore produces only male or female gametes, and the alternation of generations is completed (Ramanathan, 1939).

The _E. intestinalis_ growths in Alder Slough appear to fluctuate seasonally. As the photoperiod increases during spring and summer, the biomass of the alga also increases. The alga is most luxuriant during June, July, and August when solar insolation is greatest. In late September the alga thalli begin to be lost and biomass decreases so that much of the plant growth appears old and dull with short thalli.
It is in this condition that the alga passes through the fall and winter months.

Figure 5 shows activities of $^{65}$Zn concentrations of Zn in samples of *E. intestinalis* analyzed during this study. The $^{65}$Zn activities increased gradually during spring and summer until early August, when the levels of $^{65}$Zn in the water of the slough declined. Between the 4 August and the 11 August collections the alga apparently responded to the decrease in $^{65}$Zn input by falling to about one-half its previous $^{65}$Zn activity. Thereafter the levels of $^{65}$Zn in the plant continued to gradually decrease through September.

By late September some of the algal growth, having shed its thalli, was present as small tufts or knobs representing the holdfast and stalk. Samples of this "knobby" alga had much higher $^{65}$Zn activities probably because the base of the plant does not have as fast a Zn turnover rate as does the rapidly growing thallus.

Unfortunately, the total Zn concentrations of many of the alga samples were lost, some because of a laboratory accident in which the ash samples were spilled and some because of improper sample preparation. Therefore, values for the concentrations of Zn are available only for August, September, and October. All but one of these values fall between 95 and 120µg/g.

Specific activities of *E. intestinalis* decreased sharply between 4 August and 11 August. Thereafter, they declined more slowly
Figure 5. Zinc-65, total Zn, and specific activities of green algae. Standard deviations around $^{65}\text{Zn}$ activities are shown.
through mid-September then increased in late September. The initial rapid decrease in specific activity indicates a very fast Zn turnover rate (i.e., short biological half-life) for this alga. In contrast, the "knooby" alga maintained a relatively high specific activity despite lowered $^{65}$Zn activity in the water suggesting that the non-growing basal portion of the plant turns Zn over more slowly.

Short biological half-lives for $^{65}$Zn have been observed in other marine algae. Chipman, Rice and Price (1958) found that cultures of the marine phytoplankton, Nitzschia, rapidly absorbed almost all $^{65}$Zn introduced into their medium. Gutknecht (1963) determined biological half-lives for $^{65}$Zn in the marine benthic algae Ulva and Porphyra to be four and seven days respectively in continuous light. Under conditions of darkness these values increased to six and 12 days. The results of a number of $^{65}$Zn uptake experiments in varying pH, temperature, light, and inhibitors using living as well as killed algae led Gutknecht to conclude that $^{65}$Zn uptake and loss can largely be attributed to non-metabolic absorption-exchange processes. Such non-metabolic mechanisms for the exchange of $^{65}$Zn undoubtedly operate in all parts of the Alder Slough ecosystem also.

Rooted Vegetation

The biomass of rooted vegetation increased to a maximum in the summer then began to diminish as the photoperiod shortened in
the fall. By November, most of these plants had died and decayed, with a large amount of the material being transported from the area by tidal exchange. In general, the levels of $^{65}\text{Zn}$ in the rooted plants were low. The cattail Typha latifolia which grows mainly along the shores and is bathed by water only during the higher high tides, had lowest levels of $^{65}\text{Zn}$. Radioanalyses of cattail samples taken on 23 and 30 August 1966 showed their stems contained 1.4 pCi/g on both dates (Table 2). The specific activities of these cattail samples amounted to 0.01 $\mu$Ci $^{65}\text{Zn}$/g total Zn, which is very low when compared to the specific activities of other organisms in the study area. These low $^{65}\text{Zn}$ specific activities suggest that cattails derive very little Zn from the water. Probably, they obtain most of their nutrients and minerals from $^{65}\text{Zn}$-poor sediments below the surface as is the case of the salt marsh plant, Spartina (Pomeroy et al., 1966).

The sedges Scirpus spp. and Carex spp. are intertidal, growing in dense stands at elevations low enough to be partially covered by slough water much of the time. Scirpus is a slender plant with a network of roots near the surface of the substrate. Its $^{65}\text{Zn}$ activity, like that of the cattails, was low. Carex is a tall plant with a thick stem and a tough, tuberous root system deeply imbedded in the sediments. The $^{65}\text{Zn}$ activity in Carex was much greater than that of other rooted plants but less than that of green alga. The $^{65}\text{Zn}$ specific activities of Scirpus and Carex were many times
higher than those of *Typha* which grows in much less intimate contact with Columbia River water.

Table 2. Zinc-65, total Zn, and specific activities of rooted vegetation in Alder Slough.

<table>
<thead>
<tr>
<th>Plant</th>
<th>Collection date</th>
<th>$^{65}$Zn pCi/g</th>
<th>Total Zn µg/g</th>
<th>Specific activity $\mu$Ci $^{65}$Zn/g total Zn</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Typha</em></td>
<td>23 Aug.</td>
<td>1.4</td>
<td>113.6</td>
<td>0.01</td>
</tr>
<tr>
<td><em>Typha</em></td>
<td>30 Aug.</td>
<td>1.4</td>
<td>98.7</td>
<td>0.01</td>
</tr>
<tr>
<td><em>Scirpus</em></td>
<td>11 Aug.</td>
<td>6.0</td>
<td>7.8</td>
<td>0.77</td>
</tr>
<tr>
<td><em>Scirpus</em></td>
<td>30 Aug.</td>
<td>3.5</td>
<td>17.2</td>
<td>0.20</td>
</tr>
<tr>
<td><em>Carex</em></td>
<td>26 Jun.</td>
<td>18.2</td>
<td>18.1</td>
<td>1.01</td>
</tr>
<tr>
<td><em>Carex</em></td>
<td>11 Aug.</td>
<td>28.3</td>
<td>27.7</td>
<td>1.02</td>
</tr>
</tbody>
</table>

**Animals**

Alder Slough is primarily the habitat of small or young animals. Among the most abundant invertebrates are the amphipods, *Anisogammarus confervicolus* and *Corophium salmonis*, and the isopod, *Gnorimosphaeroma oregonensis*. The latter two seldom exceed 10 mm total length (rostrum to tip of telson) and *A. confervicolus* is rarely longer than 15 mm. One of the dominant invertebrates in terms of biomass in the slough is the sand shrimp, *Crangon franciscorum*. On the average, the sand shrimp found in the slough are smaller than those present in the estuary proper.
An exception to these observations is the crayfish, *Pacifiasticus trowbridgii*. Several adult crayfish measuring four to six inches were caught, although this species probably is normally an inhabitant of Alder Creek rather than the slough.

The fishes of Alder Slough are also predominately young animals. Of all the fishes caught and observed in the study area, only the threespine stickleback, *Gasterosteus aculeatus*, and the staghorn sculpin, *Leptocottus armatus*, are regularly present as adults in the area. Adult carp, *Cyprinus carpio*; peamouths, *Mylocheilus caurinum*, and shiner perch; *Cymatogaster aggregata*, are occasionally present, but are probably only transients in the slough.

**Amphipods**

*Corophium salmonis* is a small gammarid amphipod which probably outnumbers most of the other macroscopic animal species in Alder Slough. It lives in tubes constructed of sediment particles cemented together with glandular secretions (Barnes, 1963). The tubes, attached to various objects in the water and to bottom sediments, are very flexible and open at both ends except for those tubes found on mud which appear to stand upright from one end. On firm objects, such as stones and sticks lying on the bottom, there may be as many as ten tubes per square centimeter of surface area. Generally, there is a single occupant for each tube.
Very little information concerning the biology of *C. salmonis* is available. Haertel and Osterberg (1967) found this species to be abundant during winter and spring in the freshwater plankton of the Columbia River Estuary. It was also abundant year-round in the benthos and was the amphipod most consumed by fish in fresh waters.

Observations made during the present study indicate that reproduction occurs during spring and summer. Females bearing eggs and developing young were most abundant in May, June, and July.

The food of these interesting little animals is not known. Because they seldom exceed 7 or 8 mm from tip of the head to the end of the telson, dissection of their digestive tract is difficult. Several attempts to examine stomach contents did not yield meaningful information. However, a study of their mouth parts gives some insight into their feeding habits. In many crustaceans the major feeding appendages are the antennae, mandibles, and maxillae. Among the various species each of these appendages may differ both in form and function. In *C. salmonis* the antennae are strong appendages with several stout spines and many setae. These appendages appear well adapted both for grasping small prey and for stirring the sediments. The mandibles do not have the sharp cutting edges possessed by many raptorial crustaceans and therefore *C. salmonis* do not seem capable of capturing and masticating comparatively
large prey. The first and second maxillae are provided with very long, closely spaced setae which, in turn, bear hundreds of microscopic setules giving each seta the appearance of a feather. All the setae point anteriorly and slightly downward to form an extremely fine-meshed branchial filtering basket.

Barnes (1963) suggests that Corophium may dig up the mud and strain fine detritus through the filter setae. It may also use its appendages to create a current through the tube in order to filter food. Enequist (1949) observed this habit among many soft-bottom amphipods of the Skagerak. From the above considerations it is likely that C. salmonis is an omnivorous filter feeder subsisting on benthic algae, detritus, and microscopic animals.

The activities of $^{65}$Zn in amphipods are presented in the upper graph of Figure 6. When the input of $^{65}$Zn to Alder Slough diminished after shutdown of the Hanford reactors the levels of $^{65}$Zn in C. salmonis declined rapidly through August. Following this rapid decrease there occurred a more gradual decrease through mid-December, then an increase. The continued gradual decline in $^{65}$Zn activities, even after the reactors resumed operations and input of $^{65}$Zn into the slough increased, is probably a result of the changing physiology of these animals. As water temperatures begin to diminish in the fall the metabolic rate and food intake of the amphipods also diminish. Hence, their intake rate of $^{65}$Zn falls and also
Figure 6. Zinc-65, total Zn, and specific activities of amphipods. Samples are composed of 600 to 2,000 individual animals. Suspect total Zn concentrations are indicated by question marks.
the activity of $^{65}$Zn in their bodies is reduced by physical decay. The increase in $^{65}$Zn levels occurring in January, February, and March is difficult to explain and cannot be reconciled by this explanation.

Four of the total Zn concentrations shown in Figure 6 appear unreasonably large when compared to the majority of values which were all near 125$\mu$g/g. These anomalous values, denoted by question marks, are disregarded and in their place the values picked from the proper points on the dashed line connecting reliable concentrations are used to calculate specific activity. When the anomalous values are discounted, the concentrations of total Zn appear fairly constant. There is a slight decline during September followed by a very gradual increase throughout the remainder of the sampling period.

After reactor shutdown the $^{65}$Zn specific activities of C. salmonis quickly fell to a level about one-half its former concentration. During the period September-December specific activities showed a gradual decline followed by a rapid increase during January, February, and March.

The response of the amphipod specific activities to reduced $^{65}$Zn levels in the slough water was similar to that of the alga, Enteromorpha intestinalis. In both cases there was a rapid readjustment to a lower level indicating that algae and amphipods have
very rapid turnover rates for Zn. There is also an increase during January and February in $^{65}$Zn activities of amphipods which corresponds to a similarly-timed rise in the alga.

The rapid decrease in specific activity of *C. salmonis* may be due, in large part, to a quick decline in the specific activity of its food. If indeed its food consists of microscopic algae, animals, and detritus as suggested by the structure of its feeding appendages, then the turnover rate of its food is very fast. Chipman, Rice and Price (1958) have shown that phytoplankton cultures quickly accumulate large amounts of $^{65}$Zn from their medium. By reproducing rapidly in a medium reduced in $^{65}$Zn, the phytoplankton can also disperse $^{65}$Zn quickly (i.e., by biological dilution). Detritus has been defined by Darnell (1967, p. 376) as "all types of biogenic material in various stages of microbial decomposition which represent potential energy sources for consumer species." He pictured a particle of detritus as consisting of the particle of organic substrate plus a large microflora (bacteria, fungi, etc.), microfauna (protozoans, etc.), and adsorbed organic materials. The rapid division rates of these organisms must insure fast turnover of $^{65}$Zn at least on the surface of the detritus particle.

**Sand Shrimp**

The sand shrimp, *Crangon franciscorum* grows to a maximum
length around 75 mm and is fairly abundant along sand beaches and in estuaries from southern Alaska to southern California (MacGinitie and MacGinitie, 1949). At one time this species and a close relative, C. nigricauda supported a flourishing fishery in San Francisco Bay. These bay shrimp were originally exploited commercially in 1869 (Bonnot, 1932). In one peak year, 1930, the total catch amounted to 2.7 million pounds, worth more than $100,000 at dockside. In recent years the industry has apparently become insignificant. For example, the 1963 catch from San Francisco Bay totaled only 1,225 pounds and sold for $368 (California Dept. of Fish and Game, 1965).

Adult females carrying as many as 8,000 eggs attached to their ventral abdominal surface and pleopods usually are found in deeper waters of the estuaries. Females with eggs in the eyed stage can be found from December to June although some egg-bearing individuals may be found in any month (Israel, 1936). The eggs hatch in high salinity waters and the larval stages are planktonic. The earliest post-larval shrimp are usually found in brackish or nearly fresh water on shallow tidal flats or sloughs. The young move into deeper waters as they grow and generally mature at lengths above 40 mm. According to some investigators, these shrimp live for only one year (MacGinitie and MacGinitie, 1949).

Sand shrimp inhabiting Alder Slough averaged near 30 mm total length throughout the year. The majority measured 20 to
40 mm in all collections, with small individuals 10 to 20 mm occurring from June through October. Very few individuals remained in the slough after growing to lengths above 40 mm. Apparently, these larger shrimp began to move from the shallow areas as they approached maturity. No egg-bearing females were taken in the slough.

Numerous stomachs were opened and their contents examined to determine the role of sand shrimp in the trophic structure of the slough. To examine stomach contents, an incision was made in the carapace to allow the esophagus, stomach, and anterior intestine to be extracted with small forceps. After estimating its fullness, the stomach was opened and its contents studied. The percentage volumes of the items in the stomach were estimated after identifying them. The stomach lies in the anterior dorsal portion of the cephalothorax immediately above the mouth parts and behind the eyes. Unlike many shrimp species, the stomach of C. franciscorum does not appear to be an efficient triturating organ. It lacks the muscle attachments and efficient gastric mill necessary to grind food items into small, digestible pieces. However, the mandibles of sand shrimp are sturdy appendages with four sharp cusps on each median face which are capable of grinding food into manageable sizes prior to its entry into the stomach.

Most sand shrimp stomachs were empty. In those which contained foot items, the identifiable remains were of animal origin.
Animals occurring most often were the isopod, *Gnorimosphaeroma*, the amphipods, *Corophium* and *Anisogammarus*, and polychaete worms. Small mysid shrimp were also occasionally encountered. In addition to these animal remains most stomachs were found to contain from 10 to 50% mud and sand grains. These results suggest that *C. franciscorum* is a carnivore which ingests some mud in swallowing its prey. Price (1961) states that the sand shrimp, *Crangon septemspinosa*, of Delaware Bay is a scavenger and predator. Its stomach contained planktonic crustacea, worms, invertebrate eggs, and small mollusks. It also ate dead animals.

Sand shrimp for radioanalysis and total Zn measurements were divided into cephalothorax and abdomen samples. Figure 7 shows the levels of $^{65}\text{Zn}$ and total Zn in the cephalothorax samples. The $^{65}\text{Zn}$ activities of the abdomen samples averaged 64% of those in the cephalothoraxes. In all cases, the trends in $^{65}\text{Zn}$ levels in the abdomen and cephalothorax samples were similar and for this reason the abdomen values are not presented.

The $^{65}\text{Zn}$ levels in sand shrimp were less in late June than in mid-April. From 26 June to 22 July there was a rise, then a decline in $^{65}\text{Zn}$ levels after the Hanford reactors were shut down. These values continued to decline very gradually even after the reactors again became operative and caused the levels of $^{65}\text{Zn}$ in the water to rise slightly after mid-September (Figure 7). Between February
Figure 7. Zinc-65, total Zn, and specific activities of sand shrimp. Samples consist of cephalothoraxes from 16 to 98 individuals.
and March a faster rise in $^{65}$Zn activities was observed.

The concentrations of total Zn in sand shrimp cephalothoraxes are shown in the middle graph of Figure 7. Total Zn concentration obtained for the 22 July sample appears unreasonably large and consequently is excluded in the calculation of specific activity. With the exception of the first three values, total Zn levels in sand shrimp cephalothoraxes remained near 85μg/g for the entire sampling period.

Specific activity is a measure of the proportion of radioactive to total atoms of an element in an organism, organ, or other component of an ecosystem. In the case of a fish, specific activity may be affected by a) requirements of the species for the element, b) specific activities of the fish's food and water, c) physico-chemical forms of the radionuclide, d) turnover rates of the element in the various compartments of the fish's body, e) age, sex, and physiological condition of the fish, and f) numerous other interrelated environmental and physiological factors (Lowman, 1960). A change in the specific activity of the fish indicates that one or more of these factors has been altered. Thus, when $^{65}$Zn input to Alder Slough decreased, the specific activities of sand shrimp and all other components of the ecosystem began to diminish.

By determining the rate of change of specific activity during an interval of reduced $^{65}$Zn input, an estimate of Zn turnover time
under one set of natural conditions was obtained. As Odum and Golley (1963) emphasize, turnover times are not constants, but extremely interesting variables. The time required to reduce the activity of an organism by one-half is termed biological half-life ($T_{b/2}$). If the effect of physical decay of the radionuclide is also included in the half-life determination, the resulting time is often called effective half-life ($T_{e/2}$). In most instances in which biological or effective half-lives have been determined, the organism has been rendered radioactive then placed in a non-radioactive medium and fed non-radioactive food. The specific activity in sand shrimp from Alder Slough was affected not only by physical decay and biological turnover but also by the continued uptake of $^{65}$Zn and stable Zn from water and prey which were also declining in $^{65}$Zn specific activity. Specific activities declined continuously from late July until late November then began to rise through March. The value for 22 July was calculated using an estimated total Zn concentration of 93$\mu$g/g.

When plotted on a linear graph, the trend of sand shrimp specific activities from 1 August to mid-September appeared to curve exponentially (Figure 7). These points plotted on a semi-log graph could be closely fitted with a straight line (Figure 8) suggesting that the loss rate proceeded as a first-order reaction. The equation for the line of regression through the specific activity values was:
\[ \log Y = 0.17455 - 0.00394 X \]

of the form, \( \log Y = a + b X \)

where: \( Y \) = specific activity

\( a \) = \( Y \)-intercept

\( b \) = regression coefficient

\( X \) = time in days.

To calculate the time required to reduce specific activity by one-half (\( T_{\text{eco}/2} \)), the following equation was derived:

\[ \log Y = a - b X_0 \quad (A) \]

\[ \log \frac{Y}{2} = a - b X_{\frac{1}{2}} \quad (B) \]

where: \( X_0 \) = time zero when \( Y = 1 \)

\( X_{\frac{1}{2}} \) = time at which \( Y = \frac{1}{2} \)

subtracting (A) - (B):

\[ \log Y - \log \frac{Y}{2} = -b X_0 + b X_{\frac{1}{2}} \]

\[ \log \frac{Y}{Y/2} = -b 0 + b X_{\frac{1}{2}} \]

\( \log 2 = b X_{\frac{1}{2}} \)

hence, \( X_{\frac{1}{2}} = T_{\text{eco}/2} = \frac{\log 2}{b} \).

Apparently no term is presently available to describe the time required to reduce specific activity by one-half under such conditions. Therefore, the term "ecological half-life" (\( T_{\text{eco}/2} \)) is proposed and is to be distinguished from effective half-life (\( T_{e/2} \)) because the latter is not affected by continual addition of radioactivity to the
organism from its food and water. The ecological half-life of $^{65}$Zn in sand shrimp under the conditions prevailing in Alder Slough during this study is determined with the following equation:

$$T_{\text{eco}/2} = \frac{\log 2}{b} = \frac{0.30103}{0.00394} = 76.4 \text{ days.}$$

To obtain some effective half-lives to compare with the ecological half-life derived from field data, laboratory experiments were performed using sand shrimp. Three sand shrimp of nearly equal size were taken from Alder Slough and held in the laboratory in individual plastic containers filled with 300 ml of slough water which was changed every second day. Each shrimp was fed one adult brine shrimp (Artemia) which had been kept in a sea water solution of $^{65}$Zn for three days then bathed in non-radioactive sea water for one day. Each shrimp was periodically placed in a plastic counting tube filled with slough water and radioanalyzed in a well type NaI(Tl) crystal. In each case, the experiment was terminated by death of the experimental shrimp, usually during a molt.

The loss of $^{65}$Zn from each experimental shrimp, measured as counts per minute in $^{65}$Zn photopeak channels (CPM), is shown in the lower graph of Figure 8. There is an initial rapid loss of $^{65}$Zn during the first eight to ten days after ingestion of the radioactive brine shrimp followed by a more gradual rate of loss for the remainder of the experiments. Of particular interest is the fact
Figure 8. Loss of $^{65}$Zn from sand shrimp observed in the field and in laboratory experiments.
that the $^{65}\text{Zn}$ level in each experimental animal decreased rapidly in the initial loss phase to nearly the same level (about 33 CPM) and thereafter declined at a slower rate. This suggests that the initial fast rate of activity loss is from loosely bound $^{65}\text{Zn}$ after which the shrimp is left with $^{65}\text{Zn}$ which has been assimilated. Lines of regression fitted to the slow portions of the loss curves allowed estimation of the $^{65}\text{Zn}$ loss rate during this period. Specimens A and C which lived for four weeks after entering the slow loss phase had effective half-lives of 26 and 30 days respectively.

The much slower rates of $^{65}\text{Zn}$ loss in field animals may be due to their continued intake of radioactivity from their food and water. In particular, the continued intake of $^{65}\text{Zn}$ from the mud in the stomachs of sand shrimp may partially account for this slower loss rate. It will be recalled that the levels of $^{65}\text{Zn}$ from the sediments of Alder Slough were actually increasing during this period when $^{65}\text{Zn}$ activities of other components of the ecosystem were declining. For this reason, it is to be expected that those animals which may take in a significant portion of their $^{65}\text{Zn}$ from the sediments may be expected to have a relatively slow loss rate.

**Shiner Perch**

The shiner perch, *Cymatogaster aggregata*, is abundant along the coast from southern California to Alaska. It belongs to the
family Embiotocidae, a group of primarily marine species which bear living young. Growing to lengths up to 200 mm, these fish often provide sport and bait for fishermen along the shore and in estuaries. The breeding season extends from April to July with 8 to 36 young being released 10 to 12 months later (Clemens and Wilby, 1961).

Shiner perch were first caught in Alder Slough in mid-July. Most of these were pregnant females 80 to 110 mm in standard length and containing five to ten young. The young perch, measuring 30 to 40 mm, were born in late-July and were abundant in the slough throughout the late summer and fall. Mean standard lengths of the young fish born in July steadily increased in the following three months at a rate of approximately 9 mm per month. It is this population of juvenile fish which is dealt with in this section. No shiner perch were observed in collections after 26 October.

Young shiner perch 30 to 45 mm long were very selective feeders. Stomachs of these fish examined during July and August were almost exclusively filled with small planktonic animals. Usually 60 to 90% of the stomach contents of the very young shiner perch consisted of copepods with the remainder being branchiopods, amphipods, and annelid worms. In ensuing months these small fish grew rapidly, reaching an average standard length above 50 mm by September and exceeding 60 mm by October. To support their increased
bulk, shiner perch turned to larger prey but continued to include copepods in their diet. These older juveniles ate amphipods (mainly Corophium), isopods, and annelid worms as well as copepods. Apparently, the fish were able to catch the tube-dwelling amphipods when the small crustaceans were out of their tubes or else the fish could somehow suck the amphipods out of the tubes because only a few mud tubes were observed in the perch stomachs. Based on the stomach contents of C. aggregata from Alder Slough, this species in its early stages is a carnivore that primarily eats filter feeding animals such as copepods and cladocerans.

The fishes were analyzed for $^{65}\text{Zn}$ and total Zn individually after removing their gut contents. From each collection five juvenile shiner perch, comparable in length, were chosen for individual analysis. In this way it was hoped that a better understanding of the nature of the variability between individuals in the slough could be attained. In Figure 9 the $^{65}\text{Zn}$ activities, total Zn concentrations, and specific activities of the fish are shown. Although a value in each of the three graphs relates to an individual fish, no attempt has been made to distinguish corresponding values for each individual. In all cases, the trend lines connect median values to minimize the influence of extreme values. Although the median is a less efficient estimator of the central value than the mean if the population is normally distributed, in small samples it is not influenced
Figure 9. Zinc-65, total Zn, and specific activities of shiner perch.
markedly by extraneous values (Dean and Dixon, 1951).

The $^{65}$Zn activities of juvenile shiner perch increased during late July then decreased rapidly following reduced input of $^{65}$Zn to Alder Slough after reactor shutdown. As in the case of the sand shrimp, $^{65}$Zn activities continued to fall even after $^{65}$Zn levels in the slough increased again in mid-September (Figure 9). This continued decline may also be at least partially explained by the decreasing temperatures during this period, which act to reduce food intake and metabolic turnover of Zn by the fishes' bodies.

Considerable variation among otherwise comparable individuals is apparent in Figure 9. The ranges in the various samples of five fish varied from 18 to 91 pCi $^{65}$Zn/g. For each $^{65}$Zn activity value shown, a standard deviation was calculated based on a) weighing errors, b) counting errors, c) variations in sample geometries, and d) uncertainty in the activity of the $^{65}$Zn standard used to establish counting efficiency of the analyzer. The standard deviations of all but the first group amounted to 5 to 9% of the nominal $^{65}$Zn activities and thus were small when compared with the differences between all individuals in a sample. The standard deviations of $^{65}$Zn and total Zn values for shiner perch in the first collection were larger (12-13%) than others because of the greater error involved in weighing the small new-born fish. Calculation of the pooled estimate of the population standard deviation:
yielded a value of 22.4 pCi $^{65}$Zn/g.

Median values for total Zn concentrations in juvenile shiner perch varied from 100 to 150 µg/g and remained near 125 µg/g in most samples. Ranges of total Zn values were from 21 to 50 µg/g. Calculation of the pooled estimate of the population standard deviation produced a value of 14.9 µg/g.

Specific activities of the small fish increased somewhat during late July then began to fall rapidly with the advent of reduced $^{65}$Zn levels in the slough. The rate of decline was much more precipitous than that of sand shrimp. Figure 10 shows the specific activities of the juvenile shiner perch collected during the period of reduced input of $^{65}$Zn into the ecosystem. The data are plotted on semi-log paper and fitted with a "least squares line" to determine the rate of decline. The time required for specific activity to be reduced by one half under these conditions was estimated in the following calculation:

$$T_{eco/2} = \frac{\log 2}{b} = \frac{.30103}{.00803} = 37.4 \text{ days.}$$

The rapid rate of decrease of shiner perch specific activities after $^{65}$Zn input to the ecosystem was reduced, probably reflects
Figure 10. Loss of $^{65}$Zn from shiner perch.
the food habits of these fish. The basic food of young shiner perch, most of which measured 40-55 mm during the interval of decreased activity, was filter-feeding copepods and cladocerans. Assuming that a large portion of the food ingested by copepods and cladocerans consisted of phytoplankton and detritus, the specific activities of these planktonic animals as well as that of the phytoplankton, should have fallen quickly after reactor shutdown. Hence the young shiner perch, feeding near the bottom of the food web, reacted quickly to changes in the specific activity of the water.

This same pattern has previously been described by Foster (1958), who showed that algae and net plankton in the Columbia River exchange phosphorus very rapidly so that the specific activities of $^{32}$P in these organisms are virtually no different from that of the water. He also showed that aquatic animals in higher trophic levels had lower $^{32}$P specific activities due to physical decay of the $^{32}$P as it was passed up the food chain.

**Starry Flounder**

*Platichthys stellatus*, the starry flounder, occurs in the coastal waters of the north Pacific Ocean from Japan to southern California. It is also known from the Bering Sea and the Arctic Ocean (Orcutt, 1950). Small fish are usually found near shore in protected areas and often are found many miles up rivers in fresh water (Gunter,
Large adults attain lengths greater than 50 cm and may exceed 3 kg in weight; exceptional flounders may reach 9 kg and 100 cm. In general, young flounders remain in shallow areas while large ones move to the sea and may occur at depths of 300 m or more. Although this species does not support a large fishery by itself, it comprises a significant part of trawler catches along the coast from northern California to British Columbia.

Spawning occurs once a year during November to February, reaching its height about December and January in shallow waters near river mouths. The fertilized eggs being slightly buoyant are pelagic. The larvae which hatch about three to five days after fertilization rapidly increase in length from 2 to 12 mm before metamorphosing into the characteristic flounder shape. After metamorphosis, the juvenile flounders grow at a rate of about 8 mm per month so that one year old fish are 80 to 100 mm long (Orcutt, 1950).

Like the shiner perch, starry flounder change their food habits as they increase in size. The very young larvae depend on minute planktonic organisms for food. At about 100 mm in length their jaws and teeth are powerful enough to crush small mollusks and at even larger sizes the flounder eat fish and hard-shelled invertebrates. Orcutt (1950) examined the stomach contents of starry flounders 40 to 150 mm long and found that their primary food was the amphipods *Gammarus* and *Corophium*. Incidental items included ostracods,
phylllopods, cladocerans, isopods, and polychaetes. Analysis of the stomach contents of starry flounders caught by otter trawl in the Columbia River Estuary by Haertel and Osterberg (1967) showed that zero year class fish mainly ate copepods while zero to two year class fish primarily ate amphipods.

Study of the stomach contents of starry flounders taken from Alder Slough also show that amphipods constitute the bulk of the diets of juveniles. Other organisms often eaten were polychaetes, isopods, mysids, and copepods. Flounder sometimes ate large quantities of copepods to the exclusion of other prey species when the copepod populations became very dense. This phenomenon was also observed by Haertel and Osterberg (1967). Another item consistently found in flounder stomachs is mud. The flounder, like sand shrimp, live on and in the substrates and capture many of their prey items from the mud. As a result, they ingest significant amounts of mud. The foregoing evidence indicates that juvenile starry flounders should be considered as secondary consumers which eat herbivores (copepods, mysids, etc.) as well as omnivores (amphipods, isopods, polychaetes).

The juvenile starry flounders selected for analysis of $^{65}$Zn and total Zn ranged in standard length from 47 to 98 mm with most being 50 to 70 mm long. Median $^{65}$Zn activities increased gradually (though with greater fluctuations than those of shiner perch) during spring and
summer months then declined after July when reactor shutdown caused $^{65}$Zn input to the slough to be reduced (Figure 11). Like the other organisms studied, this decline continued even when $^{65}$Zn levels in the water again rose, probably as a result of lowering temperatures and decreased metabolic turnover of Zn.

Variation among individual $^{65}$Zn activities can be seen in Figure 11. Standard deviations were calculated for each $^{65}$Zn activity based on a) counting errors, b) weighing errors, c) variations in sample geometry, and d) uncertainty in the activity of the $^{65}$Zn standard used to determine the counting efficiency of the analyzer. These standard deviations ranged from 5 to 7% of the nominal $^{65}$Zn activities and are thus fairly small compared to differences between many individuals. Hence variation among individuals overshadows analysis errors. The largest range, 186 pCi $^{65}$Zn/g, was observed in the first sample in which the flounder with highest body burden had a $^{65}$Zn activity two to almost three times higher than others. This fish was radioanalyzed a second time and its high activity confirmed. Ranges of the other samples were from 26 to 161 pCi $^{65}$Zn/g with most being 40 to 90 pCi $^{65}$Zn/g. Calculation of the pooled estimate of the population standard deviation yielded a value of 37.1 pCi $^{65}$Zn/g.

The concentrations of total Zn in starry flounders, shown in Figure 11, tended to follow $^{65}$Zn activities. The fish with high $^{65}$Zn
Figure 11. Zinc-65, total Zn, and specific activities of starry flounders.
activity in the first sample also had a high total Zn concentration. Similarly, other individuals high in $^{65}$Zn were usually found to have high levels of total Zn. Thus, specific activities were less variable than total or $^{65}$Zn concentrations. Except for the first, all samples had ranges from 14 to 82 µg Zn/g with most ranges falling between 20 and 65 µg Zn/g. The pooled estimate of the population standard deviation was 32.4 µg Zn/g. Total Zn concentrations appeared to increase slightly during the summer and fall. After October there were greater fluctuations in median values and more variation among individuals than in earlier months.

Spring and early summer months saw definite increases in specific activity. In early August, about four weeks after reactor shutdown, specific activities began to decrease, and continued to diminish even after the reactors resumed operations. The upper graph in Figure 12 is a semi-log plot of specific activities of flounders during the period of decreased $^{65}$Zn input into the ecosystem. Calculation of the ecological half-life based on the slope of the regression line fitted to the data follows:

$$T_{eco/2} = \frac{\log 2}{b} = \frac{.30103}{.00216} = 139.4 \text{ days.}$$

Loss rate experiments were performed in the laboratory to provide effective half-lives for comparison with the ecological half-life obtained from the field data. Three juvenile flounders from
Figure 12. Loss of $^{65}$Zn from starry flounders observed in the field and in laboratory experiments.
Alder Slough and similar in size to those taken during the summer were used in the experiments. These fish were individually maintained at a constant temperature (15° - 16° C) in gallon jars half filled with water from Alder Slough. Each fish was fed one or two adult brine shrimp which had been held for three days in sea water containing 25 μCi 65Zn/L then rinsed for five hours in non-radioactive sea water. Water in the jars was discarded and replaced daily during the first four days. However, the supply of water from the slough was soon depleted so that the test fish were held in the same batch of slough water after the fourth day. It was constantly aerated and was filtered each day. Radioanalysis of this water before and after the experiments revealed a fourfold increase in 65Zn activity. This was not believed to have a significant effect on the loss rate from the flounders because 65Zn activities per unit wet weight of the test fish were at least 10^5 times greater than that in the water.

The loss rate of 65Zn from each fish was measured by periodically placing it in a water-filled plastic container and counting atop a solid 5 × 5 inch NaI(Tl) crystal. The counting efficiency for this configuration was not determined, and therefore the 65Zn values are expressed as counts per minute per gram dry weight (CPM/g) in the 65Zn photopeak channels. Although the test fish were fed one to three non-radioactive brine shrimp daily, they were generally inactive and became emaciated. Each experiment was terminated
by death of the test fish.

The lower graphs in Figure 12 present the results of loss rate experiments conducted in the laboratory. In each curve the initial $^{65}\text{Zn}$ values were high because the flounder retained some or all of the radioactive brine shrimp in its gut at the time of radioanalysis. After the first few days of the experiment, during which the water was changed daily, the test fish retained about one-half of the $^{65}\text{Zn}$ consumed. Regression lines fitted to the slow component of each loss curve were extrapolated back to the ordinates (time zero). Values of these intercepts were then assumed to represent the $^{65}\text{Zn}$ each fish assimilated. The times required for the test fish to lose one-half the $^{65}\text{Zn}$ initially assimilated are given, with related measurements, in Table 3.

<table>
<thead>
<tr>
<th>Flounder</th>
<th>Standard length mm</th>
<th>Dry wt. g</th>
<th>$^{65}\text{Zn}$ initially assimilated CPM/g</th>
<th>Effective half-life days</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>57</td>
<td>0.30</td>
<td>300</td>
<td>56</td>
</tr>
<tr>
<td>B</td>
<td>67</td>
<td>0.52</td>
<td>110</td>
<td>91</td>
</tr>
<tr>
<td>C</td>
<td>57</td>
<td>0.29</td>
<td>95</td>
<td>162</td>
</tr>
</tbody>
</table>

The effective half-lives obtained in these laboratory experiments appeared to have an inverse relationship to the initial
concentration of $^{65}$Zn. Flounders A and C were almost identical in length and weight, but A assimilated three times the initial amount assimilated by C. However, the half value time for C was three times longer than that of A, suggesting that the turnover rate is proportional to initial concentration. Flounder B, although larger than A and C, initially assimilated an intermediate activity and lost $^{65}$Zn at an intermediate rate.

No good explanation comes to mind for the apparent dependence of effective half-life on initial concentration of $^{65}$Zn assimilated. Such a relationship would suggest that the more $^{65}$Zn initially assimilated by the flounder, the more rapid would be its turnover of $^{65}$Zn at some later time. The results of the sand shrimp experiments (Figure 8) appear more reasonable. In these experiments, the excess, loosely bound $^{65}$Zn was quickly lost until all three shrimp reached a more or less common $^{65}$Zn level. This common level might be thought of as a measure of the number of Zn atoms essential for normal physiological functioning and hence these $^{65}$Zn atoms are held tightly and turned over slowly. As mentioned previously, the three test flounders became emaciated and finally died during the experiments. It is possible that the poor physiological conditions of the test fish affected the results. For this reason, no attempt to compare field and laboratory half-lives is made.

The ecological half-life of 139 days for starry flounders living
under the conditions prevailing in Alder Slough is the longest of all organisms considered in this study. Part of the reason for this long half-life lies with flounder food habits. Starry flounder subsist on a variety of omnivores such as amphipods whose specific activities decreased rapidly after reactor shutdown. Thus, the specific activities of the flounders might also be expected to decline rapidly. The fact that flounder specific activities decreased more slowly than expected probably results from their ingestion of mud. Since the mud entering the flounder gut was increasing in $^{65}$Zn during the period when $^{65}$Zn input was reduced in all other components of the ecosystem, this continual intake of relatively high activity retarded the rate of decrease of flounder specific activities.
GENERAL DISCUSSION

Temporal Fluctuations

Definite seasonal patterns in the levels of $^{65}$Zn in Columbia River water may, to some extent, affect $^{65}$Zn activities in Alder Slough organisms. The activity of $^{65}$Zn in Columbia River water varies because the proportion of the total amount of river water which is used to cool the reactors changes as the rate of river flow past Hanford changes. Thus, at low river flow a larger fraction of the total number of Zn atoms in the river passes through the reactors and is made radioactive than at high flow periods, tending to increase $^{65}$Zn activity during low river flow. The magnitudes of these seasonal fluctuations in $^{65}$Zn activity are damped because the concentrations of Zn in the water follow the river flow rates closely, attaining maximum levels during high river flow (Silker, 1964). Perkins, Nelson and Haushild (1966) observed that $^{65}$Zn activities in Columbia River water at Vancouver, Washington reached peak levels in May and June 1965 (due to scouring of sediments containing $^{65}$Zn) then decreased to low levels from August through November 1965.

Concentrations of total (radioactive plus stable) Zn in organisms in the ecosystem generally had a tendency to increase slightly from a low level during the summer, however, this was not always the case.
Some of the seasonal changes in total Zn concentrations in the animals may be due to changes in their physiological requirements for Zn with increasing age or to seasonal changes in input of Zn into the ecosystem as a result of Columbia River Zn concentration fluctuations.

The fact that temporal changes in total Zn concentrations in organisms do occur is a strong recommendation for the determination of specific activities rather than $^{65}$Zn activities alone. With specific activities, the fraction of radioactive atoms in the Zn pool of an organism is known and can be used to determine trends which might not otherwise be apparent. For instance, $^{65}$Zn activities in starry flounder during August and September showed only an irregular tendency to decline (Figure 11). In contrast, determination of specific activities revealed that the proportions of radioactive to total Zn atoms in the flounders were declining markedly.

Specific activities in some of the poikilothermic animals in Alder Slough increased during spring as water temperature increased. This was thought to be due to increased turnover of Zn brought about by rising food intake and metabolic rates. Had the input of $^{65}$Zn into the ecosystem not been reduced beginning about late July, the specific activities in these animals would have been expected to increase further during summer and fall (Foster and McConnon, 1962). Even when $^{65}$Zn input again rose there was no corresponding
rise in specific activities in the animals probably because falling water temperatures slowed metabolic rates and Zn turnover. A second explanation for this failure of the animals to respond to renewed $^{65}$Zn input into the ecosystem may be that the $^{65}$Zn was no longer available to them in large quantity. At the base of all food chains are plants and in most aquatic ecosystems the phytoplankton cells are probably always in near equilibrium with the $^{65}$Zn specific activity of the water. Thus, the phytoplankton provide a source of $^{65}$Zn directly or indirectly to animals in all trophic levels. However, in the fall and winter months solar insolation is greatly reduced due to greater cloud cover and increasing southern declination of the sun. Under these conditions, phytoplankton standing crops and production rates are low so that this link between the $^{65}$Zn in the water and the animals is restricted.

**Individual Variation**

Juvenile starry flounder and shiner perch taken in Alder Slough in the same seine haul and having similar lengths were analyzed individually for $^{65}$Zn activities and total Zn concentrations. Pooled estimates of the population standard deviations for these concentrations approximated 20% of their grand mean values. This estimate is in general agreement with Seymour (1966) who indicated that a 25% error in the $^{65}$Zn activity in oysters (Crassostrea gigas) in
Willapa Bay, Washington would not be uncommon. However, both these estimates stand in sharp contrast to the $^{65}$Zn variance among individual whitefish (Prosopium williamsoni) taken from a 20-mile reach of the Columbia River between Hanford Ferry, and Richland, Washington by Foster and McConnon (1965). These authors found differences between the least and the most radioactive muscle tissues ranging from about 2- to 60-fold during 1961 and 1962.

**Turnover Rates**

The $^{65}$Zn levels of all components in the Alder Slough ecosystem except the sediments decreased markedly following shutdown of the Hanford reactors. Because water is the primary means by which $^{65}$Zn is transported from the reactors to the estuary, the drastic reduction in input of $^{65}$Zn was first reflected in the activity of the water. Hanson's (1966) study of $^{65}$Zn activity in the ionic and particulate phases of river water at Astoria showed that the total $^{65}$Zn activity per liter fell rapidly during the latter half of July. In Alder Slough the activity of $^{65}$Zn declined from late July through August reaching lowest levels in early September.

The levels of $^{65}$Zn in the sediments of Alder Slough behaved
quite differently from those of the water (Figure 4). Following reactor shutdown and through the summer and fall $^{65}$Zn activities in the water remained at low levels while levels in the sediments showed exactly the opposite trends. Although no direct evidence is at hand, it was speculated that conditions favorable for sedimentation of $^{65}$Zn from the water resulted in an increase in $^{65}$Zn activities in the sediments.

The rates at which specific activities of the organisms declined are of interest for at least two reasons. First, the rate of decline in a particular species should be closely related to the food habits of the species and hence to its position in the trophic structure of the ecosystem. Secondly, the rate of decrease has direct application to public health aspects of environmental radioactivity.

Foster (1959) showed that $^{32}$P specific activities of Columbia River algae and net plankton were nearly the same as that of the water because the turnover of P by these organisms is very rapid. He also showed that $^{32}$P specific activities in animals were lower than those of algae and net plankton reflecting differences in P turnover rates among organisms and radioactive decay of $^{32}$P as it is transferred up food chains.

For a radionuclide such as $^{32}$P, which has a short physical half-life (14.3 days), the effects of radioactive decay along the food chain are more apparent than in the case of a radionuclide such as
$^{65}$Zn with a long half-life (245 days). As a result of the long half-life of $^{65}$Zn and because of the mid-summer reduction of $^{65}$Zn levels, the inverse relationship between trophic level and specific activity of the organisms was not evident during this study. Although no clear-cut relationship between specific activity and trophic level was evident, a definite relationship between food habits of organisms and the rates of specific activity decrease was observed.

The specific activity of the green alga, Enteromorpha intestinalis, was reduced by one half within a one-week period. Such a fast decrease indicates that this primary producer had a very rapid rate of Zn turnover under the conditions prevailing in Alder Slough in August.

The amphipod, Corophium salmonis, is thought to be a filter feeder which subsists on microscopic plants and animals and detritus. Following reactor shutdown, amphipod specific activities dropped sharply (Figure 6) so that by late August they were approximately one-half the corresponding value for late July. Such a rapid rate of specific activity decrease indicates that C. salmonis not only turned Zn over quickly but also relied on food that had decreased sharply in specific activity.

Juvenile shiner perch, Cymatogaster aggregata, experienced a very rapid rate of specific activity loss (Figure 9). These small fish primarily ate herbivorous copepods. Because they were growing
rapidly and feeding at a low trophic level, the young perch showed a quick response to reduction in $^{65}$Zn input.

Starry flounders, *Platichthys stellatus*, and sand shrimp, *Crangon franciscorum*, are bottom dwellers which eat other animals. Thus, they are comparatively high in the food webs of the Alder Slough ecosystem. Furthermore, both species ingest significant amounts of bottom sediments which were shown to increase in $^{65}$Zn activity during reactor shutdown (Figure 4). As a result of their feeding habits, both the flounder and shrimp continued to assimilate $^{65}$Zn and thus their specific activities decreased at relatively slow rates (Figures 7 and 11).

Polikarpov (1966) stated that marine radioecologists must not reduce their studies to the estimation of the degree of danger to humans but must also consider the effects of radionuclides on communities of marine organisms. Although this is wise counsel, we can benefit from the establishment of some practical guides regarding maximum amounts of radioactive levels allowable in the hydrosphere. One such "housekeeping rule", termed the "specific activity approach", was advanced by a working committee on oceanography (National Research Council, 1962, p. 22). It states:

If the specific activities of the elements of the sea in the region of growth, development, and habitation of marine food organisms can be maintained below the allowable specific activities of these elements in man and his seafood, the allowable radiation for any individual cannot
be exceeded as a result of the consumption of marine products.

The specific activity approach could be effectively utilized in the event of acute radiocontamination of an ecosystem. For example, consider the accidental release of a high specific activity $^{65}\text{Zn}$ solution into an estuary. In the immediate vicinity of the release the concentration of Zn (both radioactive and stable) might prove toxic to many organisms. However, with time the Zn solution would be diluted by mixing and dispersed by tidal flushing. At the same time, other processes, such as sedimentation and bioaccumulation at all trophic levels may concentrate the Zn. To safely eat organisms from the contaminated estuary, their $^{65}\text{Zn}$ specific activities must be less than the allowable specific activity for man. If the food organisms exceed this level it will be necessary to refrain from eating them until their $^{65}\text{Zn}$ specific activities fall to below the allowable level. The waiting period will be a function of the ecological half-life of $^{65}\text{Zn}$ in the organisms under the prevailing conditions. As an alternative to waiting for Zn turnover and radioactive decay to reduce specific activities, stable Zn in proper physico-chemical form and in amounts below harmful levels could be added to the ecosystem. High levels of stable Zn in the environment would tend to increase the exchange rate between stable Zn and $^{65}\text{Zn}$ atoms and also possibly increase stable Zn concentrations in the organisms. In this way, $^{65}\text{Zn}$ specific activities could be reduced.
SUMMARY

The artificial radionuclide $^{65}$Zn is produced by neutron activation of impurities in Columbia River waters used to cool the nuclear reactors at Hanford, Washington. During its transport down the river $^{65}$Zn is depleted by radioactive decay, bioaccumulation, and sedimentation. Zinc-65 is found in all the components of the Columbia River Estuary: water, sediments, plants, and animals.

To study temporal fluctuations, variations among individual animals, and rates of turnover of $^{65}$Zn, samples of water, sediments, and biota collected periodically from a small estuarine ecosystem were analyzed for $^{65}$Zn by gamma-ray spectrometry and for total Zn by atomic absorption spectrophotometry. During the one-year period of this study the Hanford reactors were shut down for approximately 45 days, drastically reducing $^{65}$Zn input into the ecosystem.

The activity of $^{65}$Zn in Columbia River water varies with the flow rate of the river. In most years highest levels occur in June and low levels from August through November. Following reactor shutdown in July 1966, $^{65}$Zn activities in the water of the study area fell quickly, reaching lowest levels in the first half of September and rising slightly after mid-September. In contrast, the $^{65}$Zn activity in the sediments behaved in a manner almost exactly opposite that
of the water. During the spring and summer prior to reactor shutdown, the $^{65}$Zn levels in green algae and most animals gradually increased in response to increasing solar insolation and rising water temperatures.

Variations among individual fishes in the activities of $^{65}$Zn and total Zn concentrations were examined. The pooled estimates of the population standard deviations for both quantities in starry flounders and shiner perch approximated 20% of their grand mean values.

After $^{65}$Zn input was reduced following reactor shutdown, all organisms in the ecosystem decreased in $^{65}$Zn specific activity ($\mu$Ci $^{65}$Zn/g total Zn). The rates of specific activity decline were dependent on food habits of the organisms as well as many other prevailing ecological conditions. The time required for specific activity in a species to be reduced by one-half under prevailing conditions was termed ecological half-life. Ecological half-lives would be important considerations in the event of contamination of an ecosystem with radioactivity.


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