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THE OCCURRENCE AND DISTRIBUTION OF RADIOACTIVE NON-FISSION PRODUCTS IN PLANTS AND ANIMALS OF THE PACIFIC PROVING GROUND

By
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June 12, 1957

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Operated by the University of Washington under Contract No. AT(45-1)540 with the United States Atomic Energy Commission

Abstract

Radiochemical separations were made on selected samples from the Pacific Proving Ground by means of ion-exchange resin column and precipitation techniques. The isotopes in the separated fractions were identified by their half lives, maximum beta energies, and gamma energies. In addition to radioactive fission products, the non-fission radioisotopes, Mn^{54}, Fe^{55}, Fe^{59}, Co^{57}, Co^{58}, Co^{60}, and Zn^{65}, also were identified in some samples. The latter isotopes accounted for almost all of the radioactivity in the zoological samples of marine origin.

Contents

THE OCCURRENCE AND DISTRIBUTION OF RADIOACTIVE NON-FISSION PRODUCTS IN PLANTS AND ANIMALS OF THE PACIFIC PROVING GROUND

Introduction

After the detonation of the Castle series of nuclear devices in 1954, fission product chemical separations were made on several organs and tissues of yellow snapper (Lutianus vaigiensis) and mullet (Neomyxis Chaptalli and Chelon vaigiensis) collected at Edna Island, Eniwetok Atoll, on July 3, 1954. In addition, the levels of Ca^{45} in the samples were determined. The total activities contributed by non-fission products were as follows:

	Per cent of activity contributed by non-fission products				
Organism	Bone	Muscle	Skin	Liver	Gut
Yellow snapper	99.0	98.0	86.2	95.8	50.5
Mullet	75.3	95.1	83.8	91.4	0

These results, in conjunction with β-absorption and radioactive decay data, indicated the presence of large amounts of non-fission isotopes in the fish samples. In addition, the levels of non-fission products were higher in each instance in the carnivorous fish (yellow snapper) than in the detritus feeder (mullet), suggesting that selection in favor of the non-fission isotopes was occurring in these animals.

Kawabata (1954) reported the presence of the non-fission product Zn^{65} in several organs and tissues of the albacore and big-eyed tuna, thus confirming the earlier postulate of Okano (personal communication to Kawabata), based on γ spectra, that Zn^{65} might be present. Subsequently this isotope was identified in several species of fish (Rinehart et al., 1955; UWFL-43 1955; Yamada et al., 1955, 1956; Saeki, Okano, and Mori 1955; Mori and Saiki 1956; Yoshii 1956; Nagasawa et al., 1956; Yamatera et al., 1956) and in plankton (Seymour et al., 1957).

In 1956 two additional non-fission product isotopes were reported in biological samples: Co^{60} by Weiss and Shipman (1956) and Fe^{59} by Amano et al., (1956). Amano and his co-workers also postulated the presence of Fe^{55} on the basis of decay and absorption characteristics of the iron fractions. Yoshii (1956) also suggested the presence of Fe^{55} in fish samples and Kawabata (personal communication, paper in press) subsequently confirmed the presence of Fe^{55} in contaminated fish.

In γ spectra studies done by the present authors during the latter part of 1956 the presence of Zn^{65} and Co^{60} in many organisms was established; also the presence of Co^{57}, Co^{58}, and Mn^{54} in several samples was suggested. In the present study all of the above isotopes were identified. In addition Fe^{55} and Fe^{59} were found.

Materials and Methods

The procedures employed in the present investigation.

(1) Gamma spectra were made on dried or ashed samples to determine if gamma peaks other than those from expected fission products were present.

(2) Selected samples were wet or dry ashed, dissolved in acid solution, and separated by a cation exchange technique on Dowex 50.

(3) The various fractions from the resin columns were examined by means of γ spectrometry, β mass absorption, and radioactive decay determinations.

(4) Those fractions containing several isotopes (e.g., rare earths or heavy metals) were separated further by subsequent cationic exchange or by chloride complexing techniques on anionic exchange resin.

(5) The fractions from the second ion exchange treatment (4) were examined by the methods set forth in (3).

(6) Chemical separations by precipitation techniques were done on selected fractions from (3) or (4) to further establish the identity of the isotope in question and to rule out the presence of isotopes of other elements with similar γ characteristics.

Control experiments with radioactive spikes were conducted to determine the reliability of the results obtained with the techniques used.

The radiochemical technique for the separation of the nuclides in the samples by means of ion-exchange columns was adapted from the procedure developed by Swartout et al. (1944) and Tompkins et al. (1947) of the Manhattan Project. This procedure employs the selective adsorption on, and elution from, an ion-exchange column with the aid of certain organic acids which serve as specific complexing agents for individual ionic species. This technique was used by Swartout, Tompkins and others at the Clinton Laboratories, Oak Ridge, to separate and purify fission product species in curie and trace amounts, and again by Kimura et al. (1956), Takanobu et al. (1956), and Ishibashi et al. (1956) to separate and identify the individual fission products in various biological and non-biological samples collected in the vicinity of Bikini Atoll and in Japan.

1. Preparation of the samples

Samples of ash weighing from 108 to 2,568 mg were wet ashed under the conditions given in Table 1, evaporated to dryness and redissolved in 0.1 N or 0.2 N HCl. The sample was filtered and an aliquot was removed for radioassay. The filtrate was then passed through an ion-exchange column which was prepared in the following way.

2. Preparation of the resin column

Dowex 50*, a sulfonated polystyrene resin of the cationic type, supplied in the H-form was washed three times with a greater volume of 1-6N HCl, rinsed twice with distilled water and twice with dilute HCl. The resin suspension was added to a glass column containing a glass wool plug in a constriction at the base of the

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* Dow Chemical Co., Midland, Michigan

tube. Three-column volumes of 0.1 N HCl were run through the resin until the eluate was clear and the desired flow rate was obtained. The quantity and mesh size of the resin bed for the experiments differed slightly as indicated in Table 1.

3. Elution procedure

The sample was allowed to pass through the column at a rate of 0.2-0.5 ml per minute and then the column was washed with 20-60 ml of dilute acid of the same normality as that in which the sample was dissolved. The flow rate was controlled by adjusting the height of a separatory funnel containing the appropriate eluting agent. The above fractions contained those ions not adsorbed to the resin and were designated as the anion fractions. The cations adsorbed to the resin bed were removed progressively by passing through the column the required volumes of 0.5 per cent oxalic acid, and of 5 per cent citric acid adjusted to pH of 3.5, 4.1, 4.6, 5.1, 6.6 and 6.1 with NH_4OH. In some of the experiments a mixture of 0.15N NH_4Cl and 0.1N HCl was used to remove Cs^{137}. Subsequent experiments with spikes have shown the NH_4Cl-0.1N HCl step to be ineffective and its use has been discontinued.

A summary of the elution schedule for each experiment is given in Appendix Table 1. The eluates were collected in calibrated test tubes in fractions of 2-20 ml, either by hand or by fraction collector, and aliquots of these were used for radioassay. The resin bed was removed from the column at the termination of the elution and was ignited for radioassay.

In some of the experiments certain fractions were passed through a second resin column for further separation of the nuclides.

Table 1. The methods used in the ion-exchange experiments

Experiment	Weight wet	Weight ash	Digesting procedure	Column size	Column resin mesh size	Flow rate ml/minute
		(gms)		(mm)		
I Clam kidney Belle Is., Eniwetok Atoll 9-22-56	1.10	0.142	0.2 N HCl	8x100	50-100	0.5
II Fish* livers Bikini Atoll 9-1956	7.29	0.213	Aqua regia	6x50	50-100	0.5
III Bonito livers Fox Is., Bikini Atoll 9-23-56	8.90	0.202	Perchloric and concentrated HNO_3	6x100	100-200	0.5
IV Fish livers** Belle Is., Eniwetok Atoll 5-22-54 and 6-6-54	19.10	0.491	Perchloric and concentrated HNO_3	6x100	100-200	0.5
V Top inch, island soil Belle Is., Eniwetok Atoll 9-19-56	0.906***	0.888	Concentrated HCl	6x100	100-200	0.5
Spikes Ce^{144}-Pr^{144}, Sr^{90},Co^{60} Mn^{54},Cs^{137},Ru^{106}-Rh^{106},Zr^{95}- Cb^{95},Zn^{65},Fe^{59}	0	0.994****	Concentrated HCl	6x100	100-200	0.2--0.5

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* Mullet, jack, grouper, surgeon, and big-eye

** Miscellaneous reef fish

*** Dry weight

**** Island soil from Kusaie Atoll collected 7-5-56 used as carrier

In selected experiments, the rare earth fractions (ammonium citrate, pH 3.5 a-d) were combined, evaporated to dryness, ashed and redissolved in 0.2N HCl for separation of the individual rare earth elements by secondary elution according to the method of Kimura et al. (1956). Ten ml of a 1:1 suspension of Dowex 50 in the H-form (0.2N HCl) were added to the rare earth fraction in a beaker and stirred several times. A "mother column" of Dowex 50 resin was prepared as described above and converted to the NH_4^+-form by passing NH_4 citrate, pH 2.5, through the column, until the eluate was at pH 2.5. The rare earth resin suspension was added to the column and ammonium citrate solutions of pH 2.8, 2.9 and 3.1 were passed through until the rare earth fraction started moving down the column as determined by a survey meter. Ammonium citrate solutions of pH 3.1 (120 ml), 3.3 (360 ml), and 3.4 (300 ml) were added to elute the individual rare earth elements.

In other selected experiments pH 3.5 and/or 4.1 fractions were reeluted from ion exchange columns containing Dowex 1*, an anionic type resin (supplied in the chloride form), to separate Mn, Co, and Zn from each other according to the method of Kraus and Moore (1953). These fractions were dried, ashed, and redissolved in 3-5 ml of concentrated HCl and added to a column of 6 mm diameter and 260 mm length. The resin of 100-200 mesh was prepared in the following manner.

The resin was twice soaked in distilled water in a beaker and the solution containing the small particles was decanted.

collected 7-5-56 used as carrier

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* Dow Chemical Co., Midland, Michigan

The resin was then added to the column and 30 ml of 12 M HCl were allowed to pass through under pressure at a flow rate of 0.1-0.2 ml per minute to remove the iron impurities in the resin. The sample was added to the column and the eluates were collected as described above at a flow rate of 0.5 ml per minute. Next, 8-13 ml of the following molarities of HCl were added successively: 12, 6, 4, 2.5, 0.5, 0.005 and 0, and the fractions were collected separately. The resin was removed from the column and ignited for assay.

4. Identification of the nuclides present in the fractions

One-ml aliquots of each oxalate or citrate fraction were placed on stainless steel planchets 1½ inches in diameter, dried under heat lamps, and counted in a methane gas-flow chamber or an end window counter containing a 2-inch diameter Anton Geiger tube. The HCl fractions, however, were first taken to dryness, treated with a few drops of concentrated nitric acid, dried, and rediluted to the original volume with water. One-ml aliquots of these fractions were then placed on stainless steel planchets as above and dried.

The remainders of the fractions containing significant amounts of radioactivity were dried and analyzed in a single-channel, 50-position, automatic-advance, gamma spectrometer using a 2-inch, well-type sodium iodide crystal. Radioactive decays and β mass absorption curves were obtained for some of these fractions.

Correction factors for converting β and γ counts to disintegrations per minute were calculated by the use of standards and spikes. Self-absorption corrections for β counting were made for 5 per cent citrate dried on $1\frac{1}{2}$-inch plates and in some instances for 0.5 per cent oxalate plated and flamed on the same size planchets. These correction factors are as follows:

I. Oxalate samples

Isotope	Correction factor	
	Anton	Methane gas-flow
Fe^{59}*	8.3	1.54
Zr^{95}-Nb^{95}	15.4	1.54
Ru^{106}-Rh^{106}	5.6	1.54

*Fe^{55} was counted only in the methane gas-flow chamber. A total correction factor of 4 was used (based on 50 per cent geometry and an assumed counting efficiency of 50 per cent).

II. Citrate samples

Isotope	Correction factor	
	Anton	Methane gas-flow
Fe^{59}	11.9	3.2
Co^{57,58,60}	13.1	3.1
Co^{60}	11.6	2.8
Sr^{90}-Y^{90}	4.7	1.9
Y^{91}	5.4	2.0
Zr^{95}-Cb^{95}	18.8	3.2
Ru^{106}-Rh^{106}	6.0	2.2
Cs^{137}	6.2	2.2
Ce^{144}-Pr^{144}	6.5	2.2

The disintegration rates of Mn^{54} and Zn^{65} were determined entirely by γ counting. Also, in those instances where more than one γ -emitting isotope was present in a fraction, the disintegration rate for each isotope was calculated from the γ spectrum, back-calculated to c/m β, totaled, and compared with the observed β -count rate. The difference between the calculated and observed β count was rarely greater than 10 per cent and was usually 5 per cent or less.

The radiochemical separations for the fission products and for Fe^{55} and Fe^{59} were made according to the methods outlined by Kleinberg (1954) of the Los Alamos Scientific Laboratory. The methods for Mn^{54}, Zn^{65}, and vanadium were taken from the procedures developed at the University of California Radiation Laboratory (Meinke, 1949). The determinations for cobalt were made according to two different methods: one was taken from Kleinberg and the other from Hillebrand and Lundell (1953).

Results

The identification of the isotopes in the present samples was confirmed for both the chemical and ion-exchange resin column methods by the use of radioactive spikes. In the latter technique the spikes were used to determine accurately the elution pattern under the conditions of the present work. Regardless of the number of isotopes used in an experiment, the elution pattern remained essentially the same; the isotopes were always eluted in the same fraction as shown in Table 2.

Table 2. Summary of the isotopes eluted from Dowex 50 cation-exchange resin

Fraction		Isotopes present
Anions		Ru^{106}-Rh^{106} Zr^{95}-Nb^{95}
0.5% Oxalic Acid		Zr^{95}-Nb^{95} Fe^{59}
0.15 N NH_4Cl+ 0.1 N HCl		Zr^{95}-Nb^{95} Cs^{137}
5% Ammonium Citrate		
pH 3.5	a-c	Cs^{137} Ce^{144}-Pr^{144} Zn^{65}
	d-e	Zn^{65} Co^{60}
	f-j	Mn^{54}
pH 4.1	a-e	Mn^{54}
	f-h	Sr^{90}
pH 4.6		Sr^{90}

Also, spike experiments were done at this Laboratory, in cooperation with Kawabata*, using radioactive Mn, Co, Fe, Zn, and stable Cu and Ni according to the method of Kraus and Moore (1953). The divalent transition elements were adsorbed onto and eluted from a chloride-treated column of Dowex 1. The elution pattern for these elements is given in Table 3.

Table 3. Elution pattern of divalent transition elements from Dowex 1 by complexing with HCl solutions

Fraction	Milliliters of elutriant	Element present
12 M HCl	10	Ni
6 M HCl	10	Mn
4 M HCl	10	Co
2.5 M HCl	10	Cu
0.5 M HCl	10	Fe
0.005 M HCl	20	Zn
Distilled water	10	--
Ignited resin	--	Zn

In addition to the above described spike runs, ion-exchange separations were made on five samples from Bikini and Eniwetok Atolls. These included four biological samples and one soil sample as follows:

I Clam kidney, Belle Island, Eniwetok Atoll, collected September 22, 1956.

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* Dr. Toshiharu Kawabata, on leave from the National Institute of Health, Tokyo, Japan

II Bonito liver, Fox Island, Bikini Atoll, collected September 22, 1956.

III Fish liver homogenate, How Island, Bikini Atoll, collected September 22, 1956.

IV Fish liver homogenate, Belle Island, Eniwetok Atoll, collected May-June, 1954.

V Soil, Belle Island, Eniwetok Atoll, collected September 19, 1956.

The results for the first two separations -- clam kidney and bonito liver -- are described in detail. The summarized results of these and the other three samples are shown in Table 4.

Sample I

Clam kidney sample I (Belle Island, Sept. 22, 1956) was passed through a Dowex 50 cationic resin column (Fig. 1). The anions were not adsorbed onto the resin bed and were washed out of the column with a .2 N HCl. This fraction consisted of Ru^{106}-Rh^{106} and Zr^{95}-Nb^{95} in the anion form. The disintegration rates of the Ru^{106}-Rh^{106} (13,000 d/m) and Zr^{95}-Nb^{95} (2,430 d/m) in the fraction were determined from the γ spectrum curve (Fig. 2). On the basis of the disintegration rate calculated from the γ spectrum the β count for the fraction should have been 2,190 c/m. The observed β count was 2,300 c/m.

Zr^{95}-Nb^{95}, in the cation form, and Fe^{59} were present in the oxalate sample. Based on calculations from the γ spectrum curve, cationic Zr^{95}-Nb^{95} accounted for 280 d/m and Fe^{59}, 2,680 d/m (Fig. 2). Fe^{55} was detected at a very low efficiency in the fraction because an end window geiger tube was used for the measurement.

Table 4. Summary of the levels of radioisotopes in five selected samples

Isotope	I Clam kidney Belle Is., Eniwetok Atoll 9-22-56		II Fish liver homogenate How Is., Bikini Atoll 9-22-56		III Bonito live Fox Is., Bikini Atol 9-23-56	
	d/m	%	d/m	%	d/m	%
Unknown anions	---	---	---	---	4,980	1.1
Ru^{106}-Rh^{106}	13,000	0.7	---	---	---	---
Zr^{95} Nb^{95}	2,710	0.2	---	---	100	---
Y^{91}	46,400	2.6	---	---	---	---
Total RE mainly Ce^{144} Pr^{144}	---	---	---	---	---	---
Sr^{89} Sr^{90}	---	---	---	---	---	---
Cs^{137}	---	---	---	---	---	---
Mn^{54}	38,500	2.2	8,600	6.4	8,200	1.8
Fe^{55}	1,300,000*	73.5*	20,600	15.2	250,000	55.5
Fe^{59}	2,680	0.2	---	---	488	0.1
Co^{57}	170,000	9.6	11,000	8.1	12,900	2.9
Co^{58}	162,000	9.2	4,800	3.5	4,130	0.9
Co^{60}	32,200	1.8	9,800	7.2	13,400	3.0
Zn^{65}	---	---	78,700	58.1	156,000	34.7

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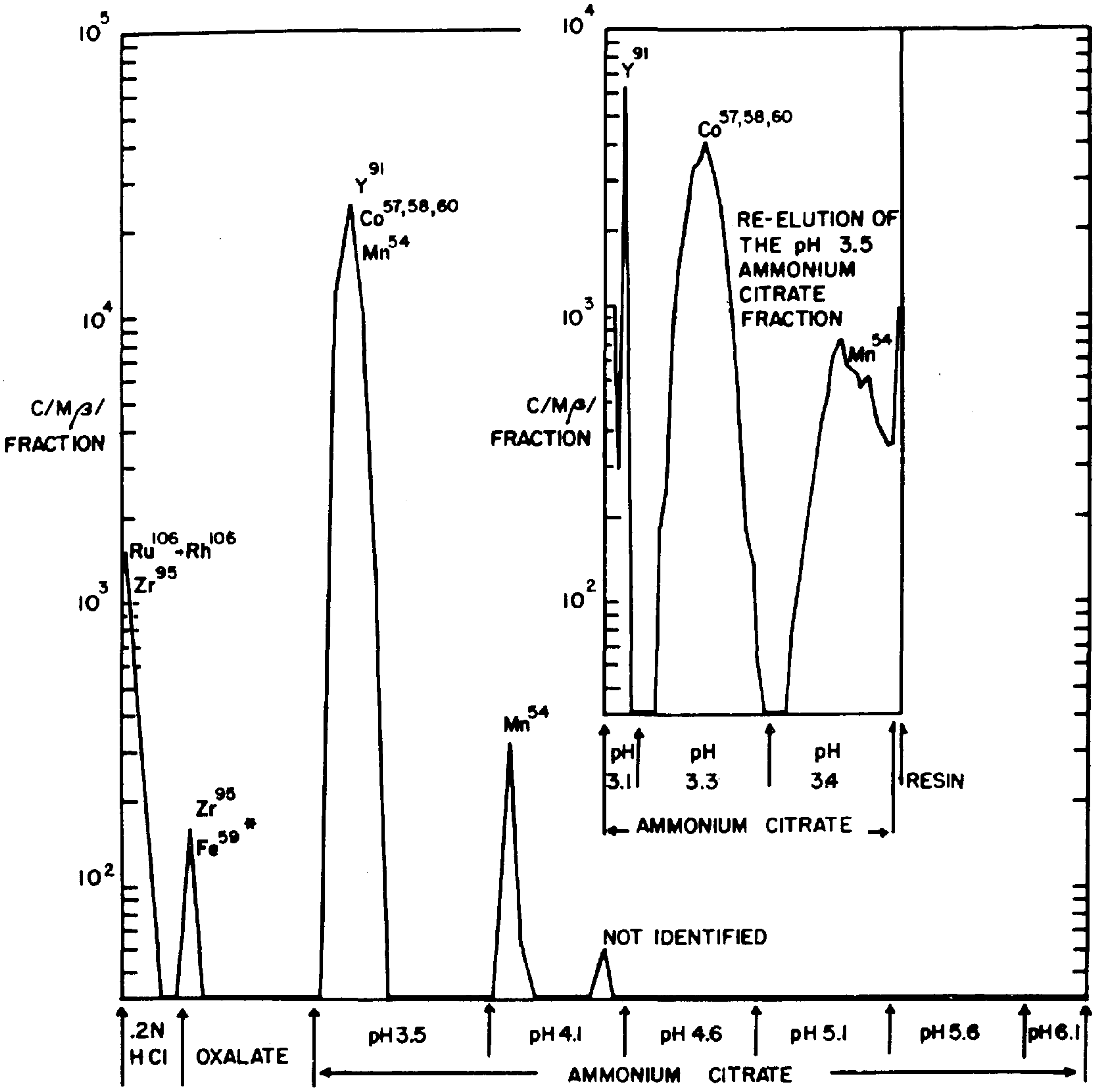
* Based on the ratio of Fe^{55} to Fe^{59} observed in other organisms collected at the same time

Table 4. (continued)

Isotope	IV Fish liver homogenate Belle Is., Eniwetok Atoll May, June, 1954 d/m	%	V Soil Belle Is., Eniwetok Atoll 9-19-56 d/m	%
Unknown anions	2,480	0.3	---	---
Ru^{106}-Rh^{106}	---	---	36,000	46.8
Zr^{95} Nb^{95}	---	---	6,060	7.9
Y^{91}	---	---	---	---
Total RE mainly Ce^{144} Pr^{144}	---	---	33,200**	43.6
Sr^{89} Sr^{90}	---	---	1,290	1.7 (1.5% Sr^{90})*** (.2% Sr^{89})
Cs^{137}	---	---	320	0.4
Mn^{54}	4,280	0.5	---	---
Fe^{55}	888,000	95.3	---	---
Fe^{59}	---	---	---	---
Co^{57}	1,400	0.2	---	1-2
Co^{58}	---	---	---	
Co^{60}	7,300	0.8	---	
Zn^{65}	29,000	3.1	---	---

** Includes a small amount of Y^{91} not separated from Ce^{144}-Pr^{144} by a second elution

*** Based on chemical separation



* Fe^{55} not detected in elution curve due to air and window absorption.

Fig. 1 Elution curves for clam kidney sample I, Belle Island, Eniwetok Atoll, September 22, 1956, from Dowex 50 cationic resin columns.

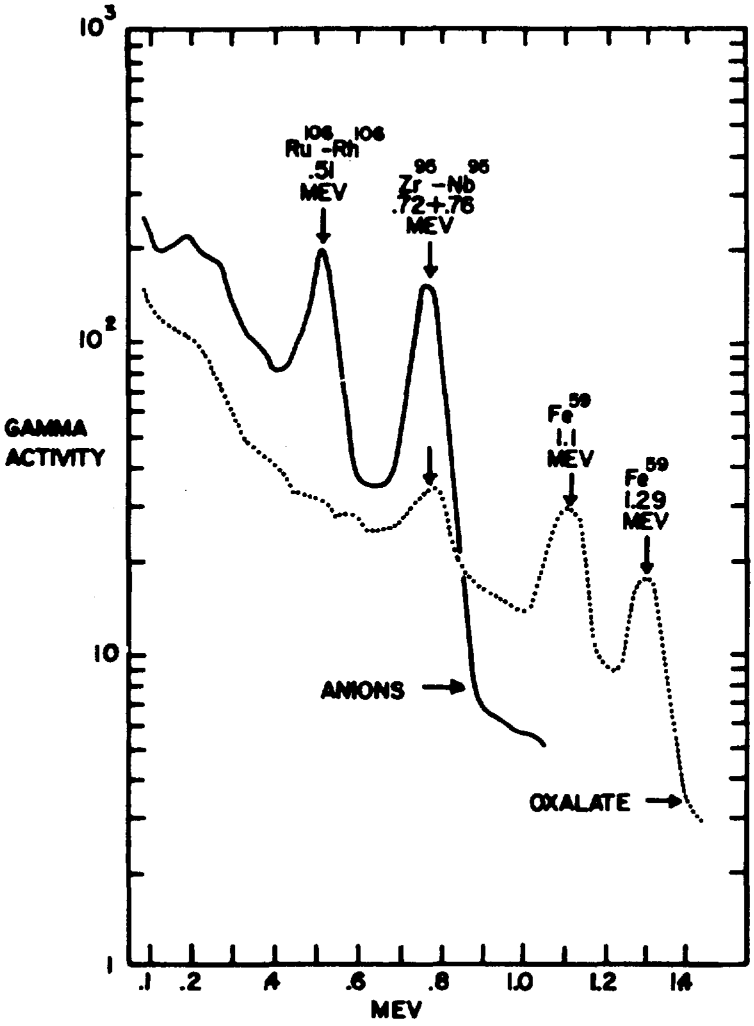


Fig. 2 Gamma spectrum curves of the anion and oxalate fractions from clam kidney sample I eluted on Dowex 50.

However, if Fe^{55} were present in this sample in the same ratio to Fe^{59} as that observed in other samples collected at the same time, the activity would have been approximately 1,300,000 d/m of Fe^{55}.

The ammonium citrate fraction at pH 3.5 exhibited high levels of β radiation in samples 3.5 b-c-d-e (Fig. 1). Because the γ spectrum contained several peaks which could not be identified a further ion exchange separation was made (Fig. 1, inset) on this fraction according to the method of Kimura et al. (1956).

The first peak of the second elution with ammonium citrate at pH 3.1 was sharply separated from the next peak at pH 3.3 (Fig. 1, inset). Although the first peak had a high β disintegration rate the γ activity was very low. The isotope in this peak was identified as Y^{91} since yttrium was known to be eluted at this position and in addition the β radiation had a maximum energy equal to that of Y^{91} (1.54 MEV, 600 mg/cm^2 range in aluminum). The observed half life for this fraction was identical with the 58-day half life of Y^{91}. The total Y^{91} content of the sample, based on the β count, was 46,400 d/m.

The second peak of the second elution occurred at pH 3.3, fractions c through r (Fig. 1, inset). Examination of the fractions comprising this peak by γ spectrometry revealed γ peaks of the following energies (Fig. 3).

0.08 MEV (weak)
0.12 MEV
0.51 MEV
0.81 MEV
1.0 MEV
1.17 MEV
1.33 MEV

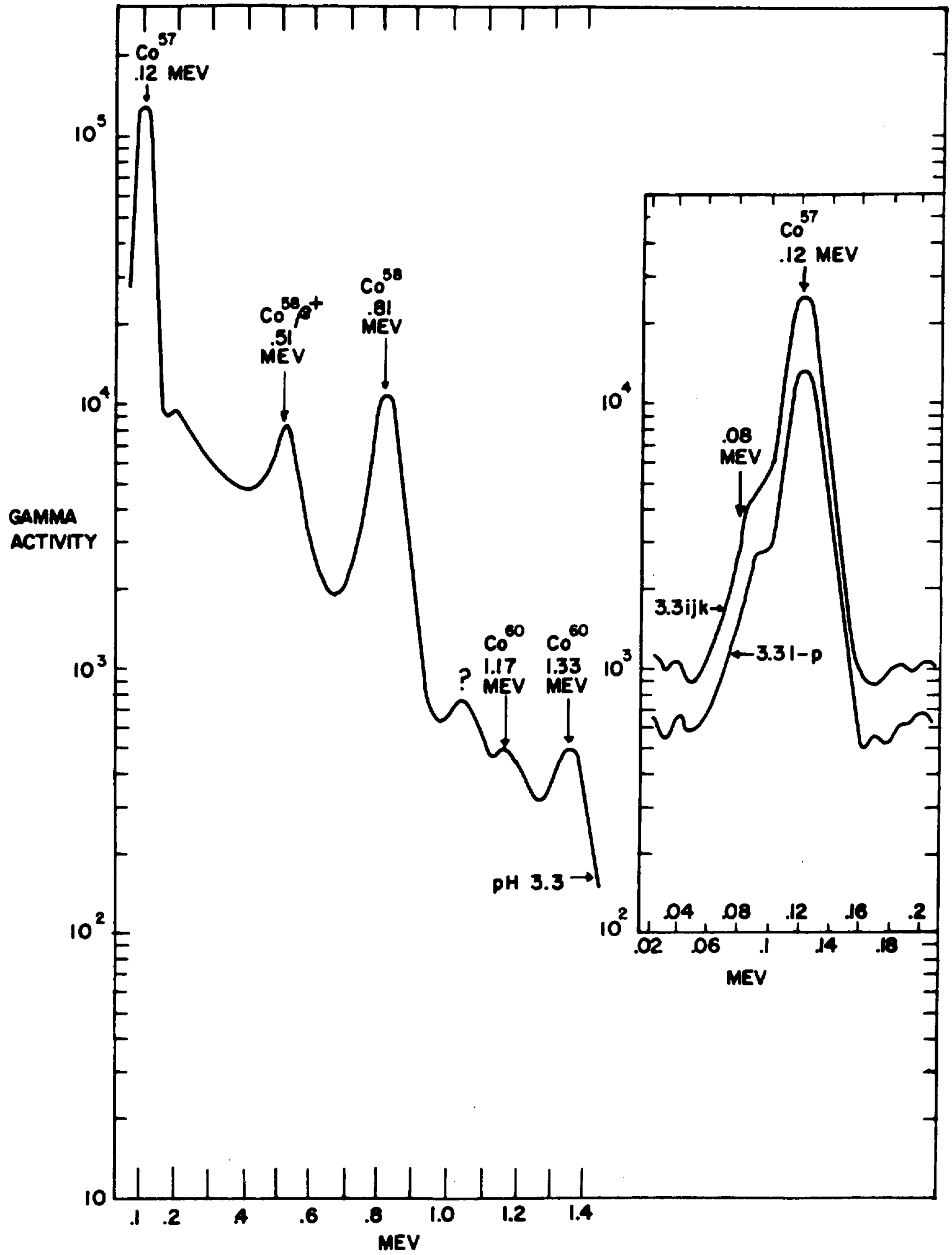


Fig. 3 Gamma spectrum curves of the pH 3.3 ammonium citrate fraction from clam kidney sample I eluted on Dowex 50.

The 1.17 and 1.33 MEV peaks are identical with those of Co^{60} and the fraction was tentatively identified as containing several cobalt isotopes (Co^{57}, .12 MEV; Co^{58}, .51, .81 MEV; Co^{60}; and an unknown with a γ peak of 1.0 MEV). The β emission from the fraction had a maximum energy of .35 MEV and approximates the maximum β energies of $Co^{57,58,60}$ (Co^{57} .26 MEV β^{+} 100%; Co^{58} .48 MEV β^{+} 15%; Co^{60} .31 MEV β^{-} 100%). Co^{56}, which might be confused with Co^{58} with regard to half life and its .84 γ emission, was not present. The 1.5 MEV β^{+} (100%) of Co^{56} and its higher energy γ peaks were not detected.

Further tests were run on the so-called cobalt fraction to determine if the γ peaks identified as Co^{57}, Co^{58}, and Co^{60} could be separated by chemical or further resin column treatment. The cobalt fraction was divided into two samples -- pH 3.3 c-h and pH 3.3 l-p -- so that both the leading edge and the trailing edge of the peak could be examined independently.

Confirmatory chemical separations for cobalt were made on the pH 3.3 c-h fraction by two different techniques in which duplicate samples were used for each method. A total count of 5,330 c/m β (counted in the methane gas-flow chamber) was present in each sample.

(1) Separation of cobalt by the α-nitroso-β-naphthol precipitate technique (Hillebrand and Lundell, 1953). From an original 5,330 c/m β in the samples, totals of 1,390 and 1,510 c/m β were counted in the cobalt separations. Gamma spectrum curves made from the separations were essentially the same as those of the original sample except the peaks were steeper and

more sharply defined in the separations. The relative heights of the γ peaks attributed to Co^{57}, Co^{58}, and Co^{60} were unchanged (Fig. 4).

The low yield of cobalt by the α-nitroso-β-naphthol precipitation technique is due partly to loss of cobalt on the iron scavenge utilized in the technique and to high self-absorption in the sample*.

(2) Separation of cobalt by the Los Alamos method (Kleinberg, 1954). From an original 5,330 c/m β in each of the duplicate samples, totals of 4,490 and 4,730 c/m β were recovered in the cobalt separations made by the Los Alamos method. The γ spectrum curves were essentially the same as those of the original sample and as those from the α-nitroso-β-naphthol cobalt separations.

After the cobalt separations were made by the Los Alamos method, vanadium separations (Meinke, 1949) were done. V^{49} has a .12 and .08 MEV γ and an approximately one-year half life and in both γ energy and half life resembles Co^{57}. In order to determine that the observed .12 MEV γ peak in the cobalt separations was from Co^{57} and not from V^{49}, the vanadium separations were made. It was found that the activity in the vanadium separations was not significantly greater than background when counted either for β or γ. Therefore, the .12

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* Potassium cobaltinitrite precipitation and extraction of cobalt thiocyanate complex into amyl alcohol ether, with appropriate scavenging steps. (as described in Collected Radiochemical Procedures, LA-1721)

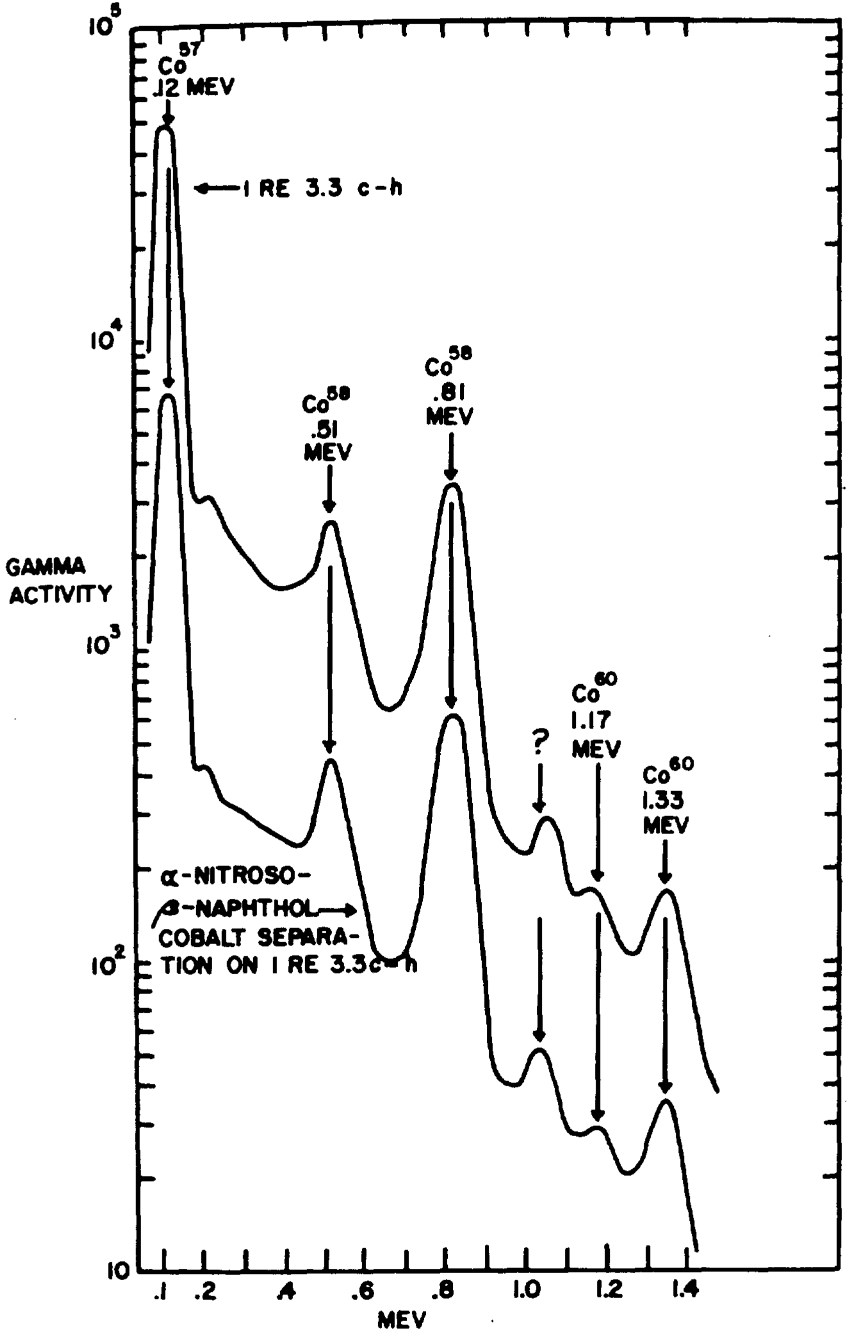


Fig. 4 Gamma spectrum curves of the pH 3.3 c-h ammonium citrate fraction from clam kidney sample I eluted on Dowex 50 and of an α-nitroso-β naphthol cobalt separation done on the pH 3.3 c-h fraction.

MEV γ peak was considered to be from Co^{57}.

The two cobalt separation methods and the subsequent vanadium separations were made on the pH 3.3 l-p fraction. The results were similar to those for the pH 3.3 c-h samples.

The above results indicate that Co^{57} (170,000 d/m), Co^{58} (162,000 d/m), and Co^{60} (32,200 d/m) comprise this fraction. All subsequent studies on cobalt in samples from the Pacific Proving Ground by means of cationic and anionic exchange resin techniques, chemical and γ spectrometric methods, and by observations of decay rates support this conclusion and will be discussed later in this paper.

The third peak in the second elution curve occurred at pH 3.4 (Fig. 1, inset). The β activity in the fraction was low with respect to the γ activity and had a maximum energy of approximately 1.0 MEV (range 410 mg/cm^2 aluminum). The γ spectrum had a pronounced peak at .84 MEV (Fig. 5). Both the β and γ characteristics of the fraction are identical with those of Mn^{54}. Cobalt separations on the fraction done by both the α-nitroso-β-naphthol and the Los Alamos methods did not have β or γ activity significantly above background. Chemical separation for Mn^{54} (Meinke, 1949) was not done on the fraction containing Mn^{54}. However, subsequent chemical separations on other samples containing the .84 MEV γ peak have given positive Mn^{54} separations and will be discussed later.

The ashed resin from the second elution experiment on clam kidney sample I also contained only Mn^{54} (Fig. 5).

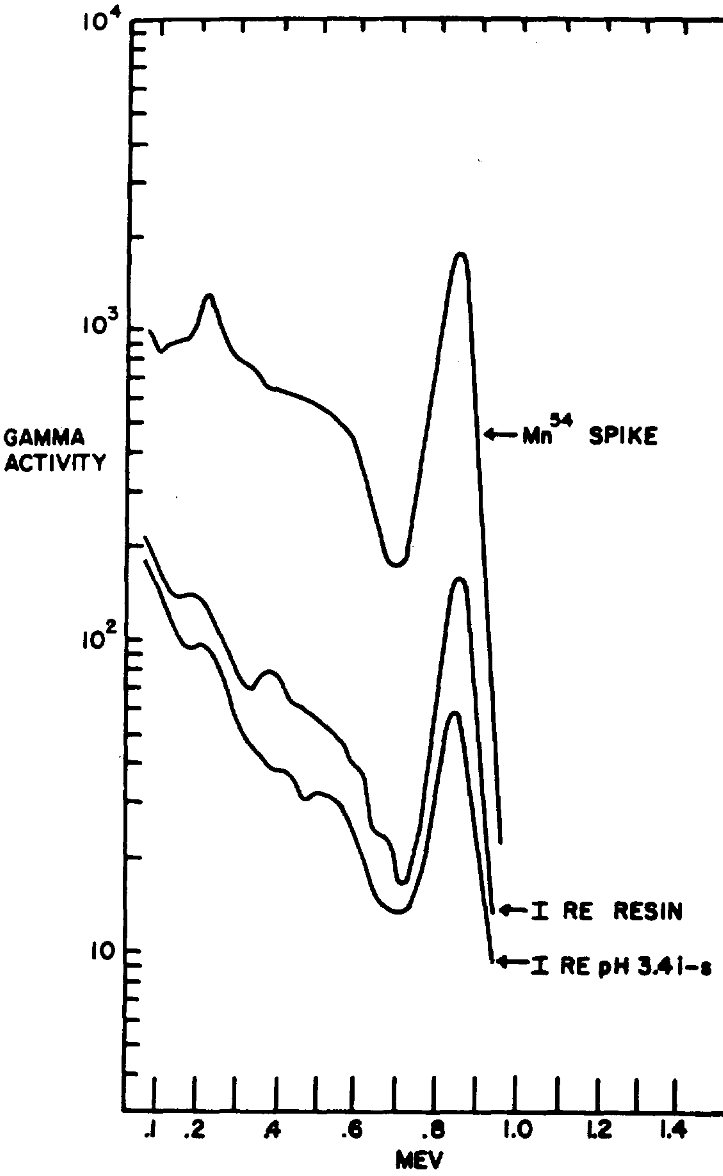


Fig. 5 Gamma spectrum curves on the I-RE 3.4 i-s fraction (second elution) and on the I-RE resin from clam kidney sample I eluted twice on Dowex 50. The gamma spectrum curve for a Mn^{54} spike is also shown.

The fourth peak in the original elution curve occurred at pH 4.1. As above, the isotope comprising the peak was Mn^{54}. The total Mn^{54} in clam kidney sample I amounted to 38,500 d/m.

The last peak of the original elution experiment contained a total of only 20 c/m over background and occurred at pH 4.1, fraction i. Because of the low level of activity the identity of the isotope or isotopes comprising the fraction was not determined; however, $Sr^{89,90}$ are known to be eluted at this position.

Sample II

Bonito liver sample II collected at Fox Island, Bikini Atoll, September 23, 1956. The elution curve for this sample is shown in Fig. 6. The anions were not identified but had maximum β energies of .03 and .45 MEV and γ peaks of .67 and .89 MEV.

The oxalate fraction contained Fe^{55}, Fe^{59}, and Zr^{95}-Nb^{95}. The activities of Fe^{59} (488 d/m) and Zr^{95}-Nb^{95} (100 d/m) were determined from the γ spectrum curve.

The identification of Fe^{55} was made by mass absorption observations. The procedures used for oxalate fractions b and c may be cited as examples of the method.

One-ml samples of oxalate fractions b and c, after drying under an infrared heat lamp, yielded counts of 11,400 and 1,240 c/m respectively in the windowless methane gas-flow counter. After the plates were flamed their counts were 29,700 and 3,510 c/m respectively. After each plate was covered with aluminum foil of 4.7 mg/cm^2 weight the observed counts were 174 and 8 c/m respectively. In a similar experiment with six Fe^{55} spikes the averages of the counts were 6,190 c/m with only carrier-free Fe^{55}

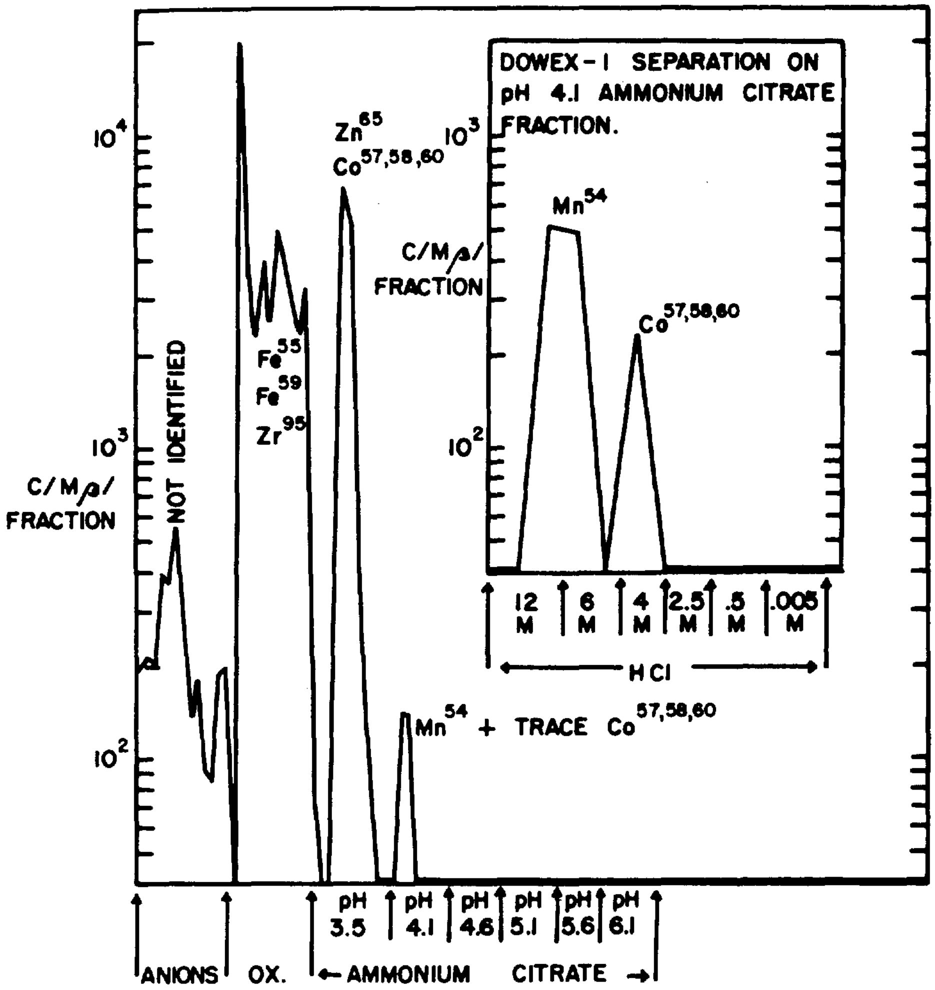


Fig. 6 Elution curve for bonito liver sample II, Fox Island, Bikini Atoll, September 23, 1956, from a Dowex 50 cationic resin column and the elution curve for a further elution of the pH 4.1 fraction from a Dowex 1 anionic resin column.

on the plates, 1,070 c/m after the addition of 1 cc of .5% oxalate (dried sample), and 15 c/m with the aluminum foil.

On the basis of the above observations, the total count on the flamed oxalate plates, after subtraction of the count contributed by Fe^{59} and Zr^{95}-Nb^{95}, was considered to be due to Fe^{55}. Because some self-absorption occurs, even on the flamed plates, the value of 250,000 d/m of Fe^{55} in the oxalate fraction is probably a conservative estimate.

The next elution peak for sample II occurred in the ammonium citrate pH 3.5 fractions d through i. The γ spectrum (Fig. 7) contained γ peaks of Co^{57} (.12 MEV, 12,900 d/m), Co^{58} (.81 MEV, 4,130 d/m), Co^{60} (1.17 and 1.33 MEV, 13,400 d/m), and Zn^{65} (.51 and 1.12 MEV, 78,100 d/m). Mass absorption curves were made from the sample and a maximum β energy of .34 MEV was observed. This is in agreement with the reported β energies for Co^{57} (.26 MEV β^+ 100%), Co^{58} (.48 MEV β^+ 15%), Co^{60} (.31 MEV β^- 100%), and Zn^{65} (.32 MEV β^+ 2.5%). The observed β count of 12,600 c/m in the fraction was identical with the β count calculated on the basis of the γ count.

The last peak observed in the elution curve for sample II occurred in the ammonium citrate pH 4.1 fractions a and b. On the basis of a low β to γ ratio, an observed maximum β energy of 1.0 MEV, and a strong γ peak at .84 MEV (Fig. 7), the isotope was identified as Mn^{54} (8,200 d/m). A subsequent re-elution of the 4.1 fraction according to the method of Kraus and Moore (1953) confirmed the presence of Mn^{54} which in this method is eluted by 12 M and 6 M HCl. A trace of $Co^{57,58,60}$ contamination was also detected.

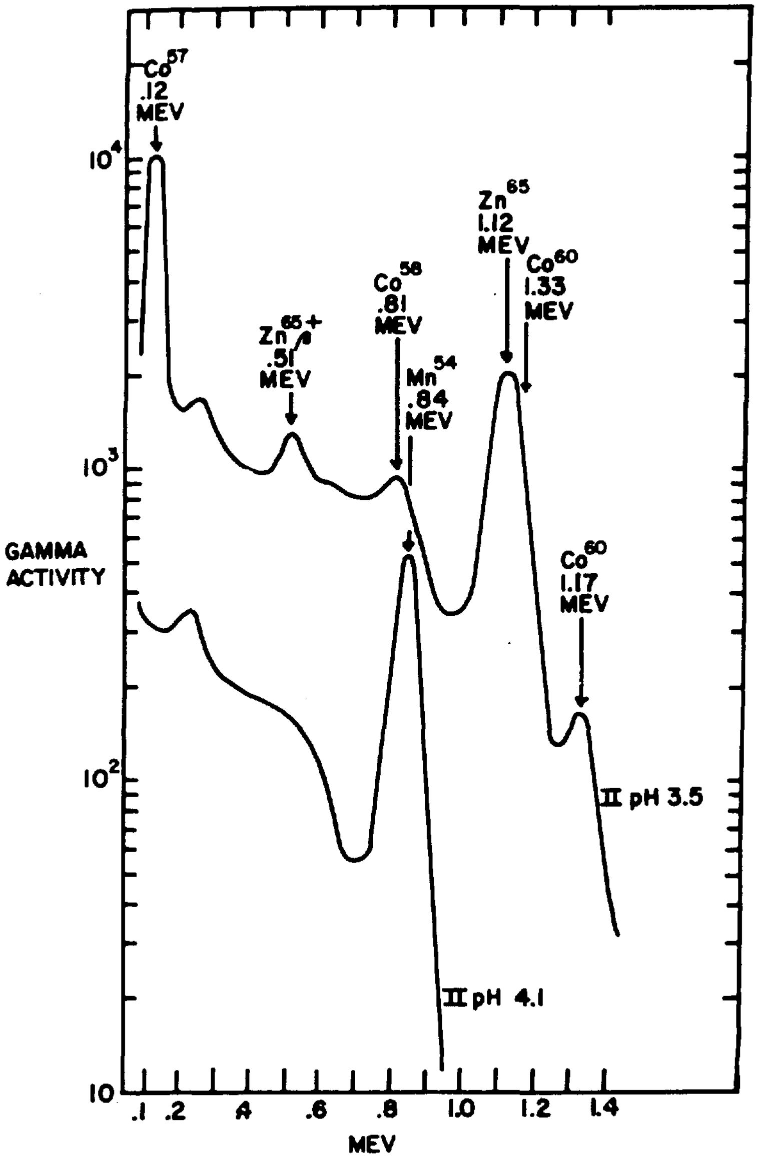


Fig. 7 Gamma spectrum curves of the pH 3.5 and 4.1 ammonium citrate fractions from bonito liver sample II eluted on Dowex 50.

Elutions from Dowex 50 were done on sample III (fish liver homogenate collected September 1956 near Bikini Island) and sample IV (fish liver homogenate collected in May and June, 1954, at Belle Island). The isotbpes contained in the samples were identified, as before, by use of the β mass absorption technique, γ spectra determinations, and comparison of the experimental elution curves with spike elution curves. In addition, cobalt separations were made on some fractions. Iron separations by the Los Alamos method were made on the oxalate fraction of sample IV. The results confirmed the ion-exchange findings.

The results of the studies on the four samples containing predominantly non-fission products and the sample of Belle Island soil which, for the most part, contained fission products are summarized in Table 4.

In order to confirm the results obtained by the ion exchange techniques, chemical separations were made for one or more of the elements, Mn, Fe, Co, Zn, and V, on selected samples. In each case the chemical findings corroborated those reported above.

(1) Chemical separations for manganese, cobalt and vanadium were done on two fish muscle samples:

a. Bonito muscle collected at Yvonne Island, Eniwetok Atoll, November 26, 1954.

b. Bonito muscle collected at the Eniwetok Deep Passage between Parry and Japtan Islands, November 27, 1954.

Gamma spectra on whole samples (a) and (b) were identical (Fig. 8) and exhibited γ peaks at

.08 (weak) and .12 MEV	considered to be from	Co^{57}
.67 MEV	" " " "	Cs^{137}
.84 MEV	" " " "	Mn^{54}
1.12 - 1.17 MEV	" " " "	Zn^{65}, Co^{60}
1.33 MEV	" " " "	Co^{60}

The results from the chemical separations are as follows:

Fraction	c/m β in methane gas-flow chamber		Reference for the chemical technique
	(a)	(b)	
Original sample	2,690	3,410	---
Co^{57}, Co^{60}	1,190	1,190	Kleinberg (1954)
Mn^{54}	747	213	Meinke (1949)
Vanadium	0	0	Meinke (1949)

In γ spectra of the manganese fractions, both samples exhibited the typical .84 MEV γ peak observed in Mn^{54} spikes (Fig. 8).

The cobalt separations exhibited the .12 MEV γ peak attributed to Co^{57} and the 1.17 and 1.33 γ peaks of Co^{60} (Fig. 8 and Fig. 8, inset). The .81 MEV peak of Co^{58} was not observed nor was it expected. The samples were collected in November 1954 and any Co^{58} present at that time would have decayed to an undetectable level by the time of analysis. The .67 and 1.12 MEV γ peaks observed in the whole samples were absent in the γ curves on the cobalt separations, thus indirectly confirming the presence of Zn^{65} and Cs^{137} in the bonito muscle samples (Fig. 8). The vanadium separation did not exhibit detectable β or γ activity.

In contrast to the absence of the .81 MEV Co^{58} γ peak in the cobalt fraction from the 1954 bonito muscle samples, strong

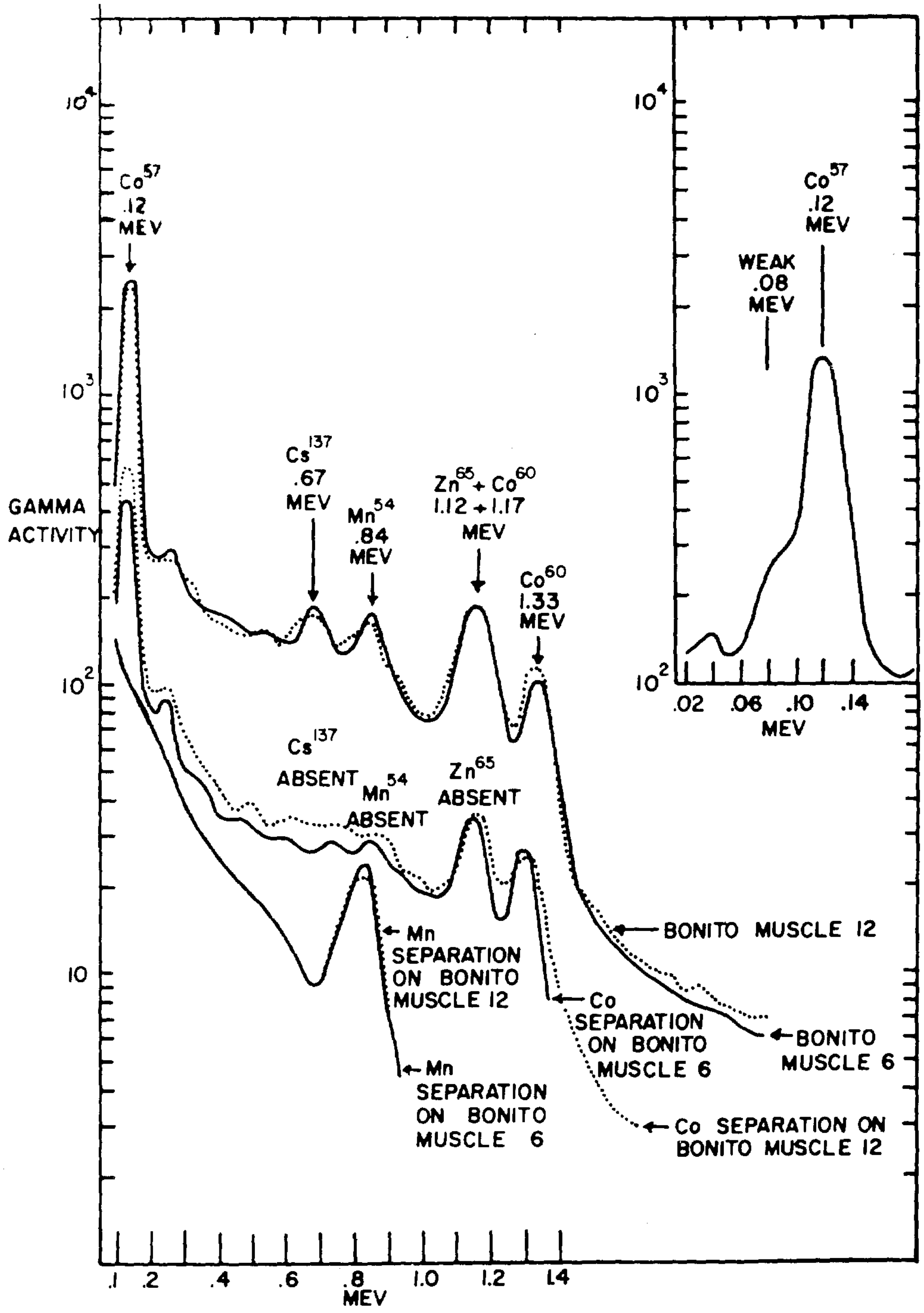


Fig. 8 Gamma spectrum curves on whole samples of bonito muscle from Eniwetok Atoll, November 26 and 27, 1954, and on cobalt and manganese chemical separations made on the muscle samples.

.81 MEV peaks were observed in cobalt separations on two different 1956 samples. Fish liver homogenate collected at Belle Island, Eniwetok, September 22, 1956, and a plankton sample (No. 42) taken 470 miles west of Eniwetok, September 13, 1956, (Seymour et al., 1957) both exhibited strong .81 MEV γ peaks in the whole samples as well as in the cobalt separation. The peaks attributed to Co^{57} and Co^{60} were also present in the whole samples and in the cobalt separations.

(2) Radiochemical separations for zinc were made on two fish liver samples:

a. Fish liver homogenate, Belle Island, Eniwetok Atoll, September 22, 1956.

b. Bonito liver, How Island, Bikini Atoll, September 22, 1956.

A comparison of the c/m β for the total sample and the Zn^{65} fraction could not be made for sample I because the total sample count was made with the methane gas-flow counter and the Zn^{65} count with the end window geiger tube. However, sample II and its Zn^{65} fraction were counted on the methane gas-flow counter and out of a count of 8,450 c/m β in the whole sample, 3,600 c/m β were obtained in the zinc separation. The γ curves for the two samples and that of a Zn^{65} spike are shown in Fig. 9. A small amount of contamination by cobalt radioisotopes was present in the zinc separations.

Supplementary evidence concerning the distribution of the non-fission products to that obtained by chemical and ion exchange

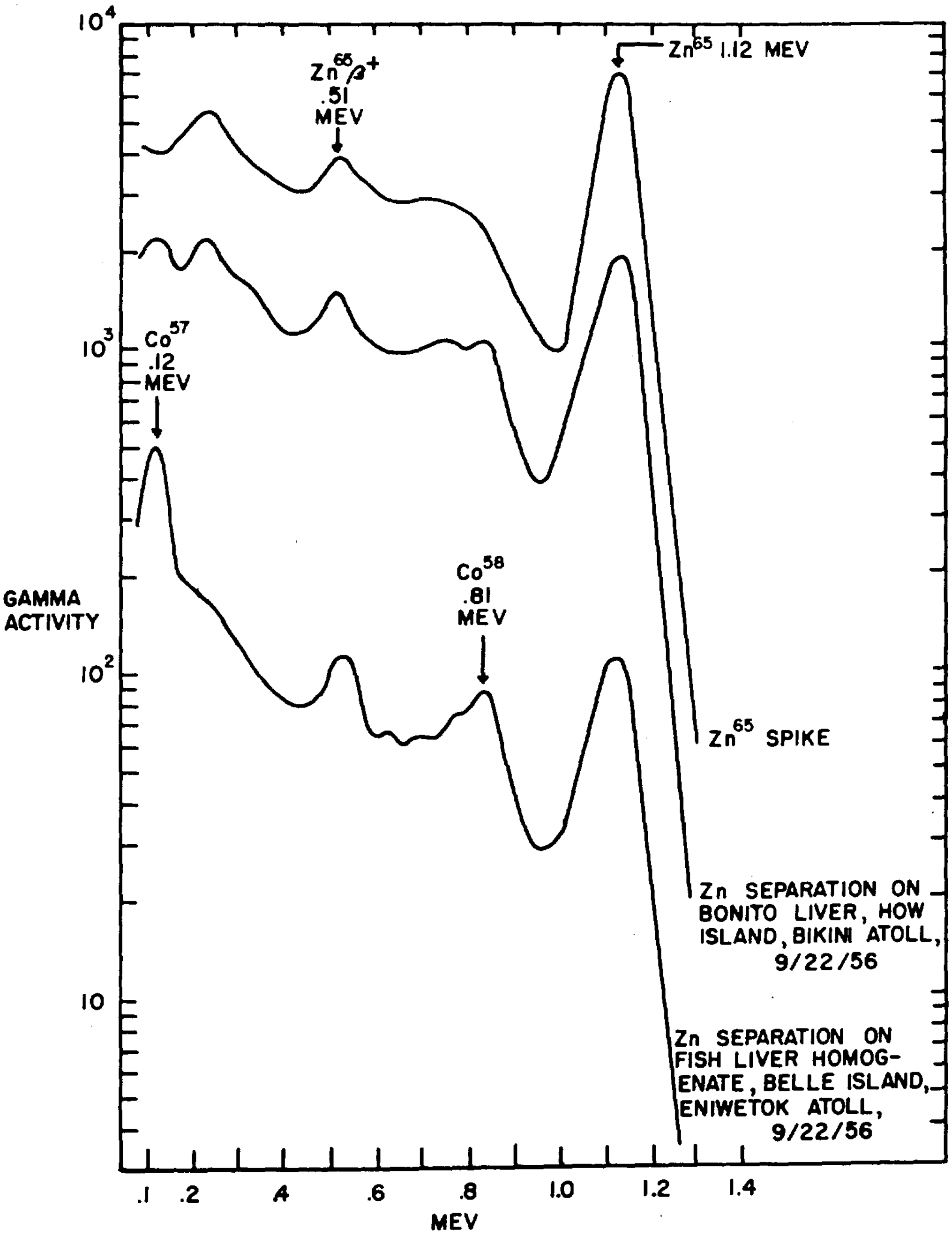


Fig. 9 Gamma spectrum curves from zinc chemical separations on fish livers collected at How Island, Bikini Atoll, and Belle Island, Eniwetok Atoll, on September 22, 1956. A gamma spectrum curve on a Zn^{65} spike is shown for comparison.

resin column techniques is provided by gamma spectrometric observations on whole samples.

Of the transition elements reported in this paper, Co^{60} and Zn^{65} were first detected by their γ peaks in spectra of whole samples. Fe^{55}, Fe^{59}, Co^{57}, Co^{58}, and Mn^{54} were not identified, however, until the samples containing them were subjected to chemical and ion exchange separation, although the γ peaks of Co^{57}, Co^{58}, and Mn^{54} were present in whole samples. The 1.17 and 1.33 MEV γ peaks of Co^{60} were first observed by the present workers in the following samples examined in late 1956 and early 1957:

Organism	Date of collection	Site of collection
Whole fish	Nov. 8, 1952	Janet Is., Eniwetok
Snail liver	June 19, 1954	Belle Is., Eniwetok
Bonito muscle	Nov. 26, 1954	Bruce Is., Eniwetok
Clam kidney	June 1954	Belle Is., Eniwetok
Clam kidney	July 1955	Belle Is., Eniwetok
Clam kidney	Sept. 1956	Belle Is., Eniwetok
Crab carapace	July 27, 1956	Belle Is., Eniwetok
Herring	July 27, 1956	Belle Is., Eniwetok
Plankton	Sept. 1956	West of Eniwetok
Fish liver	Sept. 22, 1956	Belle Is., Eniwetok

Gamma spectra from the first two samples (snail liver and bonito muscle) above are shown in Fig. 10. The Co^{60} γ peaks of the snail liver are not as well defined as those of the bonito muscle or the Co^{60} spikes. This is due to the penetration of the aluminum case of the NaI crystal by high energy β radi-

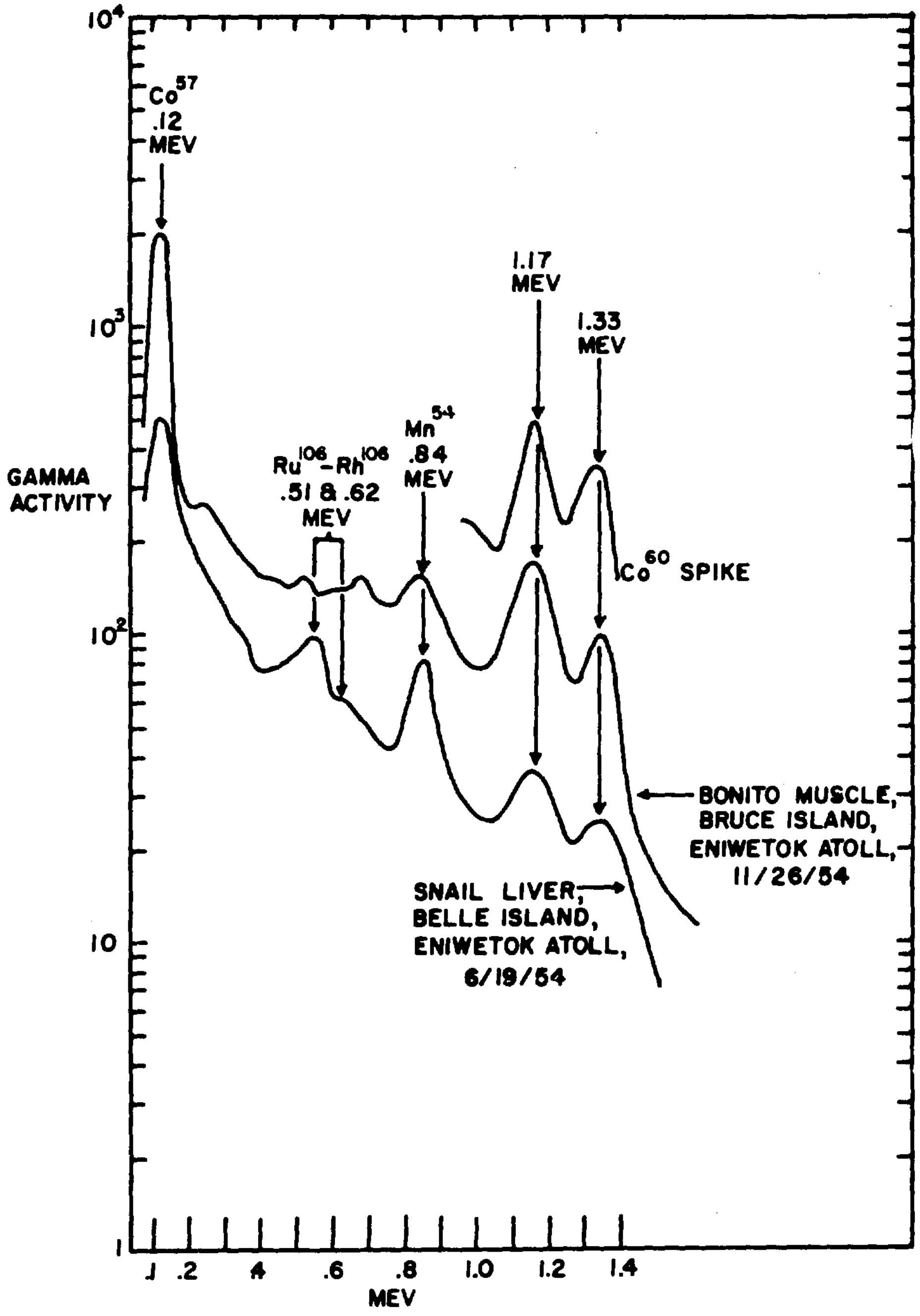


Fig. 10 Gamma spectra on bonito muscle and snail liver showing the gamma peaks of Mn^{54}, Co^{57} and Co^{60}. The gamma peaks of Ru^{106}-Rh^{106} are present in the snail liver curve.

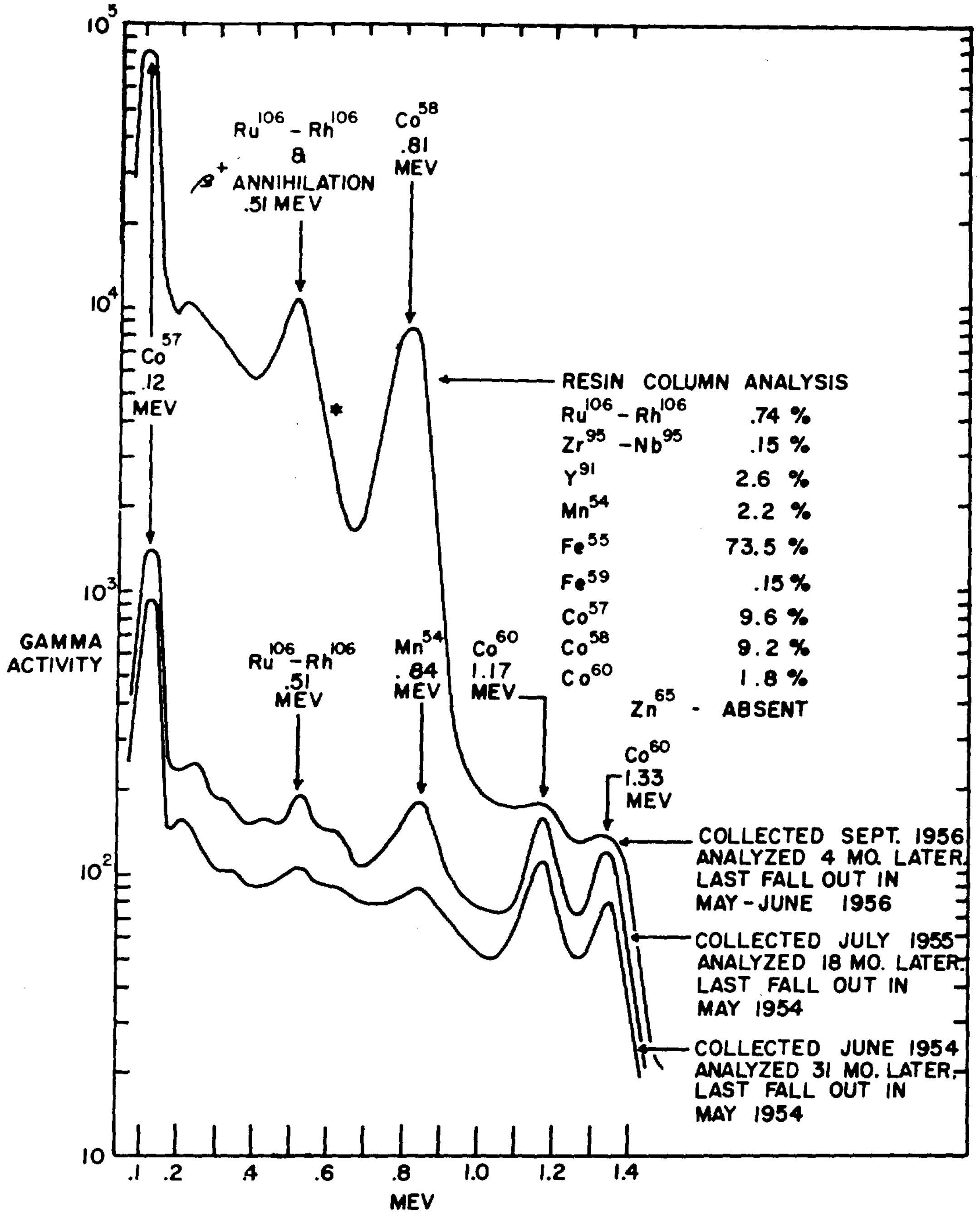
ation from Rh^{106} in the snail liver sample. Although the γ peaks for both Co^{57} and Co^{60} are present in the γ curves for both samples, the peak for Co^{58} is not present. This is expected. The Co^{58} in the samples would have passed through about 12 half lives and would be present in insignificant amounts. Mn^{54}, on the other hand, with a half life of 310 days would have gone through only about three half lives and its .84 MEV γ peak is evident in the curves from both samples.

The .84 MEV peak of Mn^{54} is easily confused with the .81 MEV peak of Co^{58}. In those samples containing both isotopes, the γ peak from Mn^{54} can be detected only after the Co^{58} has been allowed to decay out as shown in Fig.10, or after chemical separation.

Figure 11 shows γ spectrum curves from three clam kidney samples made 4, 18, and 31 months after the dates of collection. The Co^{57} and Co^{60} peaks are present in all three curves. In the curve analyzed 4 months after the date of collection the .81 MEV Co^{58} peak is very strong. In the other two curves this peak has decayed out and is replaced by the .84 MEV Mn^{54} peak.

Representative whole samples in which the 1.12 MEV γ peak of Zn^{65} has been observed include the following:

Organism	Date of collection	Site of collection
Whole fish	Nov. 8, 1952	Janet Is., Eniwetok
Fish skin	May, June, 1954	Belle Is., Eniwetok
Fish muscle	" " "	" " "
Fish bone	" " "	" " "
Fish liver	" " "	" " "



* Higher background due in part to high energy beta particles penetrating the aluminum shield on the NaI crystal (Rh^{106} 3.53, 3.1, 2.44 beta; Y^{91} 1.54 beta).

Fig. 11 Gamma curves on clam kidneys made 4, 18, and 31 months after the dates of collection.

Organism	Date of collection	Site of collection
Fish gut	May, June, 1954	Belle Is., Eniwetok
Whole mullet	May 19, 1954	Edna Is., Eniwetok
Plant leaves (Scaevola)	Sept. 22, 1956	Bikini Is.
Plankton	June 1956	West of Eniwetok
Plankton	Sept. 1956	" " "
Coconut crab liver	" "	Nan Is., Bikini
Coconut crab carapace	" "	" " "
Bonito liver	Sept. 22, 1956	Bikini Is.
Bonito muscle	" " "	" "
Fish liver homogenate	" " "	" "

Gamma spectra from the first five samples are shown in Fig. 12. Only in the fish liver sample is the Zn^{65} γ peak low, in which case it is masked by the 1.17 MEV peak of Co^{60}. The γ peak for Zn^{65} is, however, strong in the curves for the other fish tissues (gut, bone, skin, and muscle) indicating a high level of Zn^{65} in comparison with the other γ emitters. Mn^{54} is also present in the fish bone, skin, and muscle samples. The latter sample also exhibits a weak peak for Cs^{137}, an isotope often found in the muscles of many species of animals collected at the Pacific Proving Ground.

High levels of Zn^{65} are also present in fish collected recently (Sept. 1956, Fig. 13) as well as in the 1954 fish samples described above.

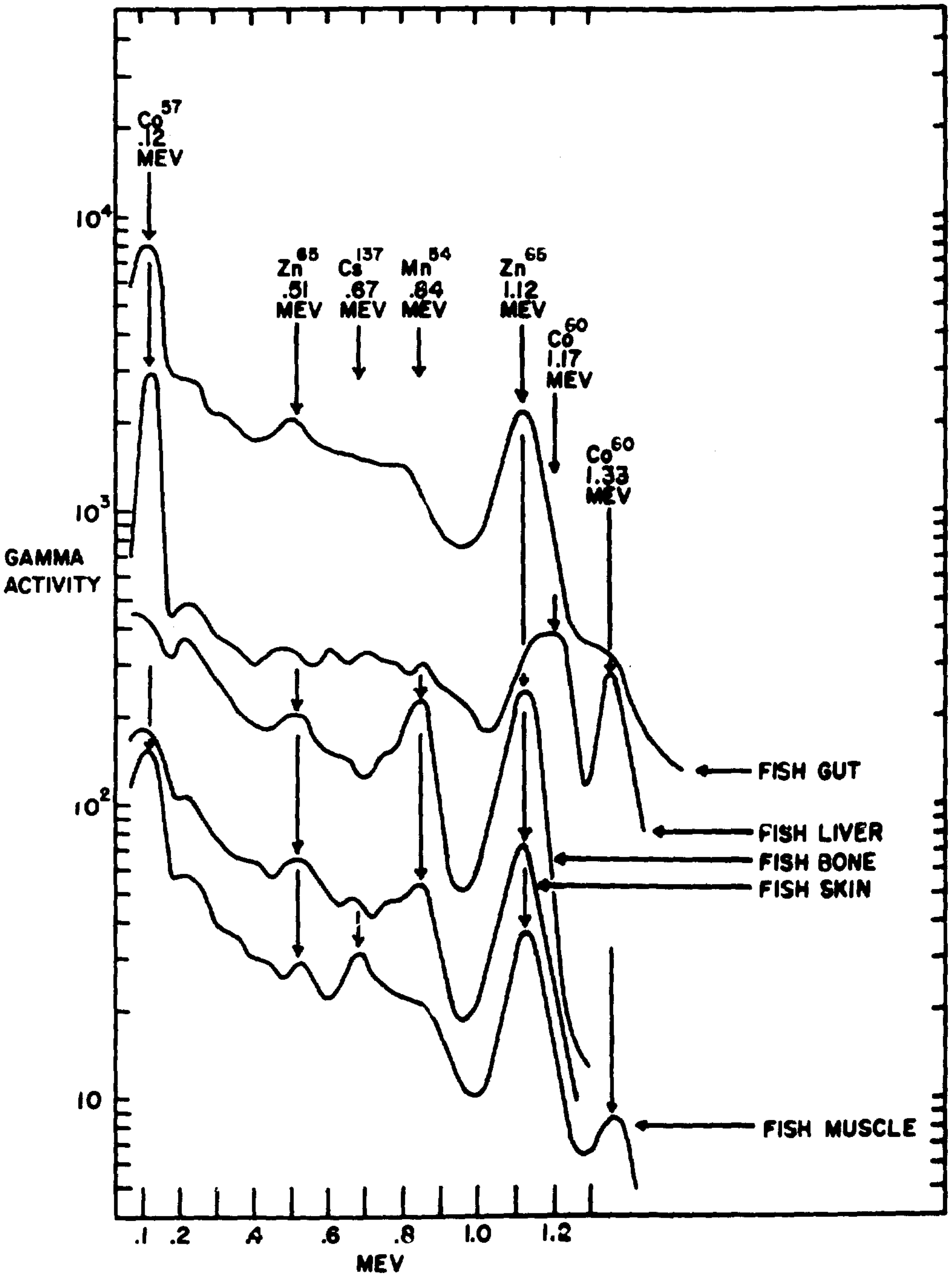


Fig. 12 Gamma spectrum curves of fish tissues collected at Belle Island, Eniwetok Atoll, May-June, 1954, and analyzed February 1957.

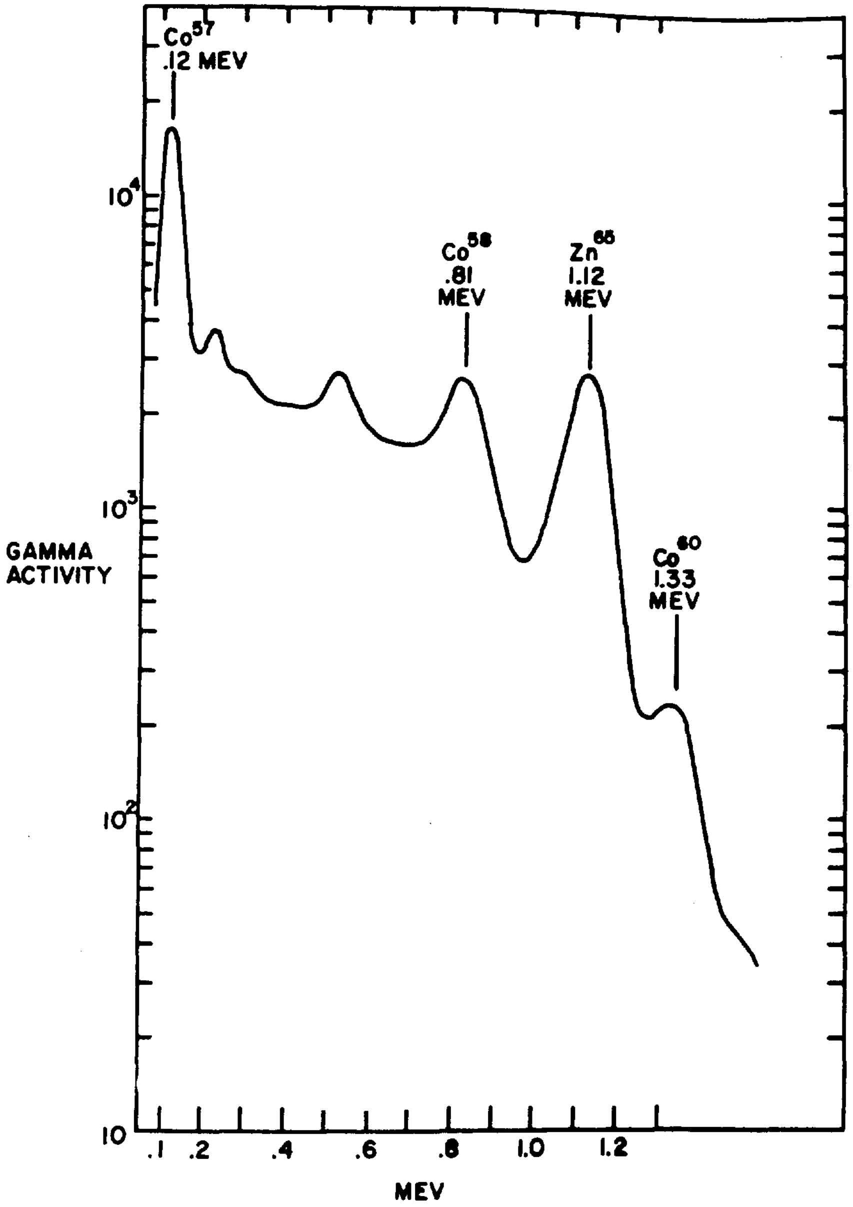


Fig. 13 Gamma spectrum curve on fish liver homogenate collected on How Island, Bikini Atoll, September 22, 1956.

The terns, or sea swallows, at the Pacific Proving Ground also contain Zn^{65} in their organs and tissues. Except during the time interval shortly after detonation, almost all of the radioactivity in these animals is due to Zn^{65}. In Fig. 14 the γ curves for tern muscle, lung, liver, kidney, and gut, collected in 1954 at Bikini, are compared with the curve for a Zn^{65} spike. In each case Zn^{65} contributes almost all of the total γ activity. In tern liver samples collected in September 1956 at Bikini, however, Co^{57}, Co^{58}, and Co^{60} were also present although almost all of the radioactivity was contributed by Zn^{65} (Fig. 15). In the corresponding 1954 Bikini tern liver sample shown in Fig. 15 all of the γ activity was due to Zn^{65}.

A pattern of distribution of the radioactive isotopes of the transition elements manganese, iron, cobalt and zinc in the plants and animals at the Pacific Proving Ground is evident even from the limited number of γ spectrum curves made thus far.

In general, these isotopes are found in the sea and not on the land, in the marine animals and not in the marine plants.

Table 5 shows the samples, on which γ spectra have been made, in which none of the above named transition elements could be detected. Table 6 shows the samples in which the isotopes in question have been observed by γ spectrometry.

On the land few or none of the non-fission products were detected in the soil, in the plants growing in the soil, nor in the herbivorous rats.

In the water surrounding the islands few or none of the non-fission products were present in the algae nor in the sea

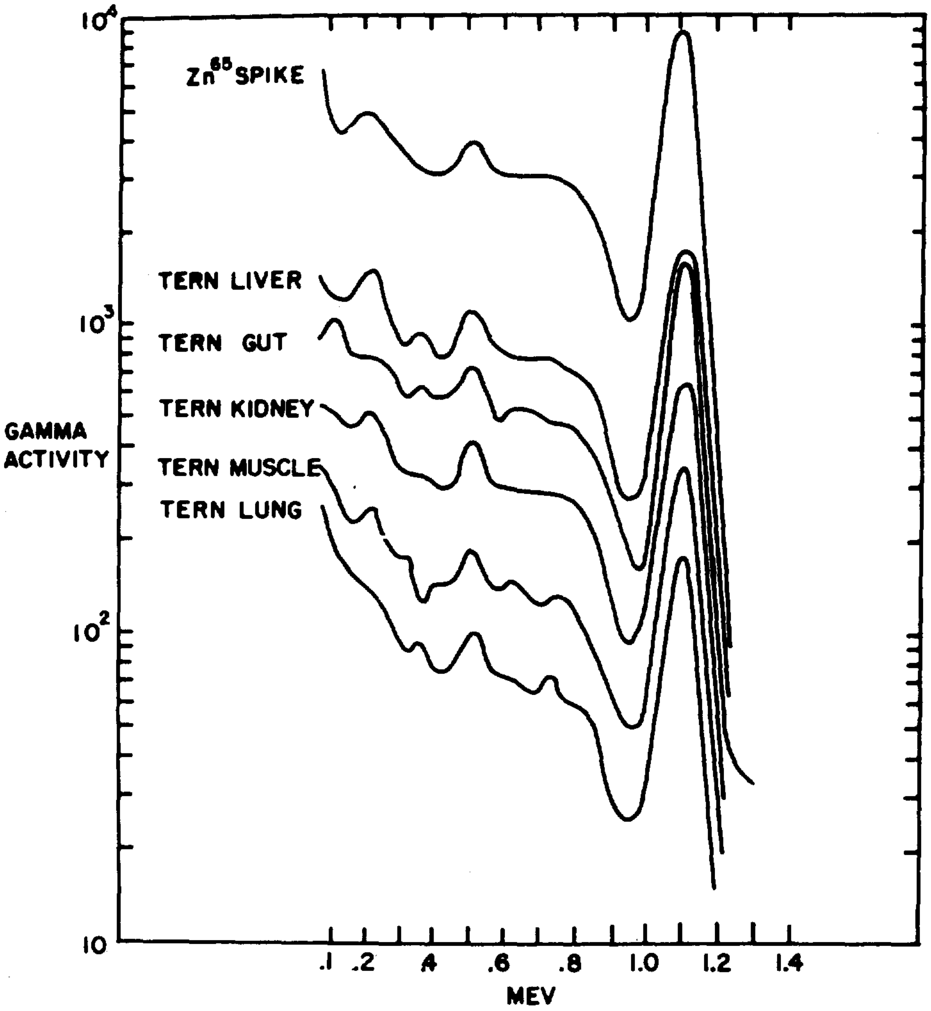


Fig. 14 Gamma spectrum curves for tern muscle, lung, kidney, liver and gut, Eniwetok Atoll, March, June, 1954. Gamma spectra made March 1957. A gamma spectrum curve for a Zn^{65} spike is also shown.

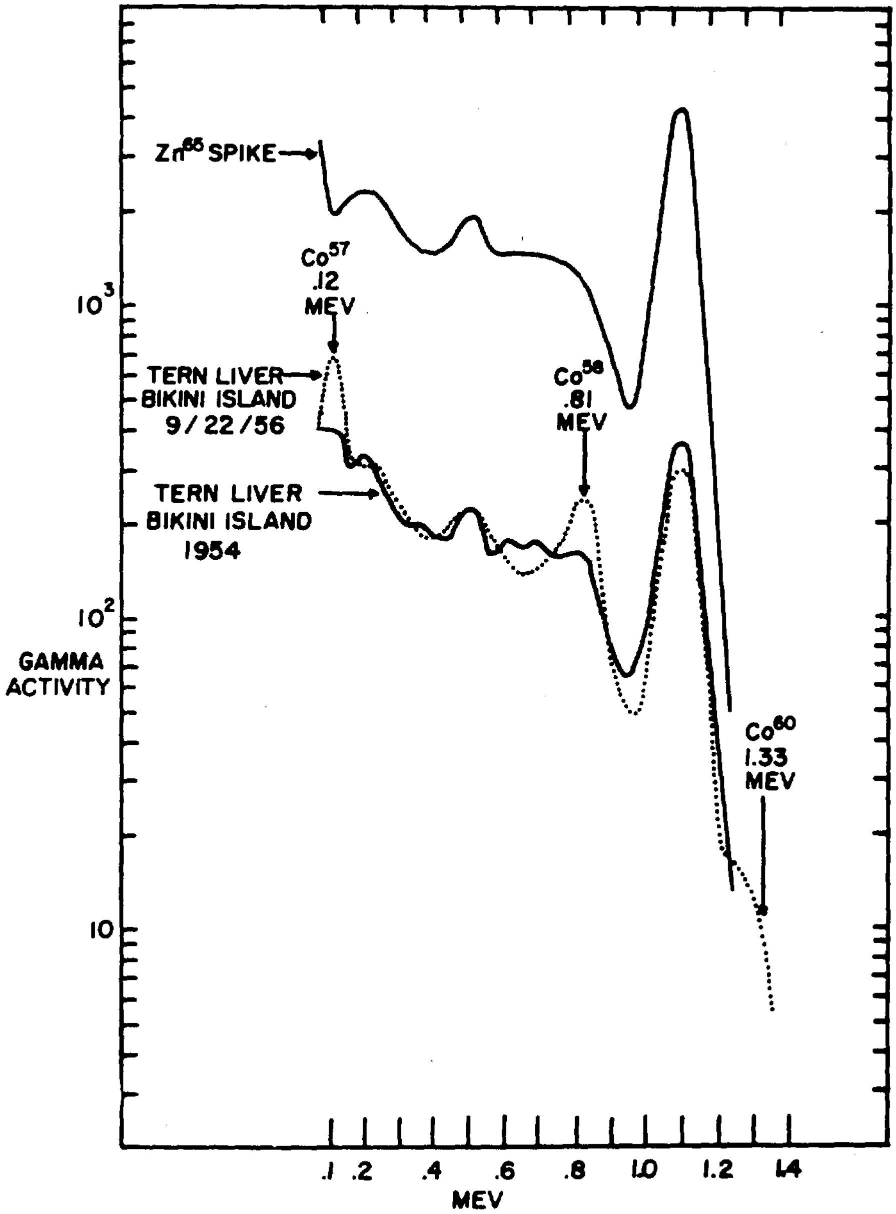


Fig. 15 Gamma spectra for tern livers collected at Bikini Atoll in 1954, and September 22, 1956. A gamma curve for a Zn^{65} spike is shown for comparison.

cucumber which feeds on organic detritus in the bottom sand.

The findings are entirely different, however, in the plankton, the invertebrate filter feeders and omnivores, and in the fishes. In all of these forms the non-fission products are present -- in some species representing almost 100% of the total radioactivity in the organism.

Table 5. Samples not containing Mn^{54}, Zn^{65}, Fe^{59}, Co^{60}, Co^{58}, and Co^{57}

Sample	Date	Place
	Algae	
Padina	Sept. 19, 1956	Belle Is., Eniwetok
Cladophora	Sept. 30, 1956	Elmer Is., Eniwetok
Halimeda	Sept. 19, 1956	Belle Is., Eniwetok
Porites	Sept. 22, 1956	" " "
Asparagopsis	July 28, 1956	Elmer Is., Eniwetok
Lyngbya	May 6, 1956	Vera Is., Eniwetok
	Land Plants	
Papaya seeds	Sept. 22, 1956	Nan Is., Bikini
Papaya pulp	" " "	How Is., Bikini
Papaya skin	" " "	" " "
Morinda meat	" " "	" " "
Coconut meat	" " "	" " "
Cenchrus leaves	April 24, 1956	Janet Is., Eniwetok
Cenchrus leaves	July 4, 1956	" " "
Sida leaves	" " "	" " "
Triumfetta leaves	" " "	" " "
Scaevola leaves	May 15, 1956	Belle Is., Eniwetok

Sample	Date	Place
	Invertebrates	
H. atra gut	July 27, 1956	Belle Is., Eniwetok
	Rats	
Muscle	Jan.-March, 1955	Janet Is., Eniwetok
Liver	" " "	" " "
	Miscellaneous	
Soil	May 6, 1956	Vera Is., Eniwetok
Soil	May 25, 1956	Edna Is., Eniwetok
Soil	Sept. 19, 1956	Belle Is., Eniwetok

Table 6. Non-fission products observed by gamma spectrometry

Specimen	Collection site	Collection date	Mn^{54}	Zn^{65}	Fe^{59}	Fe^{55}	Co^{60}	Co^{58}	Co^{57}
Algae									
Halimeda	Fox[1]	9-23-56		+					
Spyridia	"	"		+			?	+	?
Padina	"	"		+			+	+	
Halimeda	How[1]	9-22-56		+					
Spyridia	"	"		+					
Asparagopsis	Elmer[2]	9-1956		+					
Asparagopsis	"	9-30-56		weak			?	weak	
Rhizoclonium	Belle[2]	9-19-56					weak	weak	weak
Enteromorpha	"	"					weak	weak	weak
Spyridia	"	"					weak	weak	weak
Land Plants									
Messerschmidia	Fox[1]	9-23-56		+					
Scaevola leaves	How[1]	9-22-56	+	+				+	
Arrowroot corms	"	12-20-54		weak					
Coconut meat	Nan[1]	9-24-56						weak	
Land plants	Vera[2]	3-30-54	weak	weak					
Sida	Janet[2]	9-29-56		+					
Sida	"	4-24-56		weak					
Scaevola stem	Belle[2]	4-26-56						weak	
Messerschmidia	"	9-19-56		weak					
Plankton									
Plankton	Nan[1]	9-24-56		+					
Plankton	Tewa crater[1]	9-23-56		+					
Plankton	Deep Pass[2]	7-17-56		+					
Walton Plankton 8C	Open sea	6-1956		strong			+		
" Plankton 9D	Open sea	"		weak					

Table 6. (continued)

Specimen	Collection site	Collection date	Mn^{54}	Zn^{65}	Fe^{59}	Fe^{55}	Co^{60}	Co^{58}	Co^{57}
Plankton (continued)									
Marsh Plankton 55	Open sea	9-1956		strong				+	
" Plankton 42	" "	"		strong			+	+	+
" " 5	" "	"		strong					
" " 47	" "	"		+					
" " filt. 42	" "	"					+	+	+
" " 54	" "	"		strong					
Invertebrates									
Heliopora	How[1]	12-20-54		+					
Black sponge	Nan[1]	9-24-56	?	?	?		+	+	?
Coconut crab liver	"	9-1956		strong			+	+	
" crab carapace	"	"	?	strong				?	
Hermit crab feces	Seattle	11-3-56		strong					
Cenobita carapace	Belle[2]	9-27-56		weak			weak		
Cenobita muscle	"	"		+			+	strong	
Clam kidney	"	5 & 6-1954					+	+	+
Clam kidney	"	9-22-56					+	strong	strong
Clam kidney	"	3-21--11-1 1955					+	+	+
Sea cuc. gonad	"	7-27-56		+					
Sea cuc. gut	"	"							
Sea cuc. integument	"	"		?					
Fish									
Bonito muscle	Fox[1]	9-23-56		strong				+	
Bonito liver	How[1]	9-22-56		strong+				+	
Fish livers	"	9-1956		strong+			+	strong	strong
Bonito muscle	"	9-22-56		strong				+	
Goatfish liver	Kabelle[3]	7-24-56		strong	trace	?			

Table 6. (continued)

Specimen	Collection site	Collection date	Mn^{54}	Zn^{65}	Fe^{59}	Fe^{55}	Co^{60}	Co^{58}	Co^{57}
Fish (continued)									
Marsh flying fish liver	Open sea	9-1956		strong			+	+	+
Bonito muscle	Bruce[2]	11-26-54	+				strong		strong
Bonito liver	South of crater[2]	"		strong			weak		+
Bonito muscle	Mike crater[2]	"	+				strong		strong
Tuna liver	Mike crater[2]	"		strong+			weak		+
Blue bonito bone	Lagoon[2]	2-12-55	strong	strong					
Blue bonito muscle	"	"	?	strong			+	?	strong
Bonito muscle	"	11-26-54							strong
Bonito muscle	"	"	+	+			+		strong
Bonito muscle	"	"	+	+			+		strong
Mullet	Edna[2]	5-19-54		strong	?	?		+	+
Whole convict surgeon	"	"		strong	?				
Fish skin	Belle[2]	1954	+	strong+					
Fish liver	"	9-22-56		strong	+		+	+	+
Fish liver	"	"		strong	+			+	+
Herring	"	7-27-56		+			+	strong	strong
Fish livers .5 MEV scale	"	5-22-54 6-6-54							+
Fish liver	"	5-16-54		strong			strong		strong
Fish liver	" "	5-22--6-6 1954	?	strong			strong	weak	strong
Fish muscle	"	5-16--6-6 1954	?	strong+			+	weak	+
Fish gut	"	5-16--6-6 1954	?	strong+			weak	weak	+
Fish bone	"	5-16--6-6 1954	+	strong+					

Table 6. (continued)

Specimen	Collection site	Collection date	Mn^{54}	Zn^{65}	Fe^{59}	Fe^{55}	Co^{60}	Co^{58}	Co^{57}
Birds									
Tern liver	How[1]	6 & 7-1954		~100%			+	+	
Tern liver	"	9-22-56		strong			weak	+	+
Tern liver	Belle and Janet[2]	1954		~100%					
Tern muscle	" "	"		~100%					
Tern gut	" "	3 & 6-1954		~100%					
Tern lung	" "	" " "		~100%					
Tern liver	Eniwetok	1, 2, & 3-1955		strong			+		
Tern gut	Belle and Janet[2]	1, 2, & 3-1955		strong					
Tern kidney	" "	1, 2, & 3-1955		strong			+		

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[1] Bikini; [2] Eniwetok; [3] Rongelap

Discussion

The physiological roles of manganese, iron, cobalt, and zinc in plants and animals are not completely understood. However, these elements participate in many enzyme reactions and along with other elements have been conclusively demonstrated to be nutritionally essential to, and incorporated into, higher animals including humans (Underwood, 1956).

In the present work, detailed studies have been done on clam kidney and fish livers. Although the accumulation of the non-fission products is a process common to both animals, the concentration patterns for the different isotopes are not the same. These differences are to be expected between various phyla, and even between species within one phyla in which different metabolic patterns or food habits occur.

In the fish livers Zn^{65} amounted to 35-58% of the total activity but in the clam kidney this isotope was not present. On the other hand, the absolute levels of radioactive iron, cobalt, and manganese were much higher in the clam kidney than in the fish livers.

The presence of large amounts of heavy metals in the clam kidney is in agreement with the observations of members of this Laboratory that the clam kidney has a remarkably high specific gravity in comparison to soft tissues of other animals. This high specific gravity is reflected in the relation of ash weight to wet weight in the present experiments. In the clam kidney the ash fraction accounted for 13% of the wet weight, but in fish livers the ash accounted for only 2.6-2.9% of the wet weight.

Although accurate determinations of levels of fission and non-fission products have been made for only a few samples, summarized above, the gamma spectrum survey studies indicate a widespread distribution of the non-fission isotopes in the contaminated area. The distribution is due, in part at least, to the uptake of the non-fission radioisotopes by plankton, the organisms that comprise the base of the food chain cycle. Not only are the non-fission products present in plankton samples from Eniwetok and Bikini lagoons, but they are present also in samples collected in the open sea several hundred miles west of the test atolls (see also Seymour _et al_., 1957).

The absolute levels of the different radioisotopes in the organs and tissues of the marine animals of the contaminated area are of interest, especially with regard to the possible utilization of the animals for food by humans. The levels of radioisotopes reported in the present work cannot, however, be used directly for health safety consideration since the samples under study are not representative of the average values for the food organisms in the region.

However, in the absence of a better comparative standard, the levels of the different isotopes in the four biological samples described in this paper may be compared with the maximum permissible concentrations (MPC)* for water as follows:

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* The MPC for water cannot be used directly for food items unless the diet of the individual under consideration is known

	Ratio of observed activity to MPC (water)*								
Sample**	Ru^{106}-Rh^{106}	Y^{91}	Mn^{54}	Fe^{55}	Fe^{59}	Co^{57}	Co^{58}	Co^{60}	Zn^{65}
Clam kidney Belle Is. 9-22-56	0.05	0.1	12.	140.	11.	9.7	44.7	.65	---
Fish liver homogenate Fox Is. 9-23-56	---	---	0.42	0.32	---	.1	.2	.03	0.08
Bonito liver Fox Is. 9-23-56	---	---	0.32	3.2	---	.09	.14	.03	0.13
Fish liver Belle Is. 5 and 6, 54	---	---	0.08	5.3	---	.005	---	.008	0.01

In the clam kidney the levels of Mn^{54}, Fe^{55}, Fe^{59}, Co^{57} and Co^{58} are greater than the maximum permissible concentration in water for human consumption. In an average of the fish livers, however, only the level of Fe^{55} is greater than the MPC.

The levels of activity in these samples, however, are undoubtedly greater than the average for the animals of the Pacific Proving Ground, since the samples were collected in regions subjected to greater than average amounts of contamination. The general levels of non-fission product radioisotopes in the marine organisms and the variation in radioactivity content between indi-

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* MPC/ml H_2O for $Mn^{54}=1.3x10^{-3}$ uc (Seymour, 1957), $Fe^{55}=4x10^{-3}$, $Fe^{59}=1x10^{-4}$ uc, $Co^{57}=7.2x10^{-3}$, $Co^{58}=1.5x10^{-3}$, $Co^{60}=2x10^{-2}$, $Zn^{65}=6x10^{-2}$ (Handbook-52)

** See appendix table

viduals of the same species taken within the contaminated region are being investigated at this Laboratory at the present time.

Origin of the non-fission products

A survey of the samples in which the non-fission product radioisotopes have been found reveals the following:

(1) Snail liver samples collected prior to the Nectar shot and whole fish samples collected November 8, 1952, and examined in 1957 contained Co^{57}, Co^{60}, and Mn^{54}, thus indicating that the Mike shot (Nov. 1, 1952) produced these isotopes.

(2) Fish samples collected after the Nectar test (May 14, 1954) near the crater contained higher levels of radioactivity in comparison with the pre-Nectar levels, all of which were contributed by non-fission product isotopes, suggesting the production of these isotopes by the Nectar test.

(3) Fish and clam samples collected at Bikini and Eniwetok Atolls in 1956 and examined early in 1957 contained the non-fission products mentioned previously, including the short lived isotope Co^{58}, thus indicating the production of these isotopes during the summer of 1956.

The isotopes Mn^{54}, Fe^{55}, Fe^{59}, Co^{57}, Co^{58}, Co^{60}, and Zn^{65} all belong to the so-called transition elements and are characterized chemically by variable valence, easy oxidation and reduction, and marked tendency to form complexes. An examination of the interrelationship of these elements in a table of nuclides reveals that three, Mn, Fe, and Co, fall in one group with the elements Ni and Cu separating them from the remaining element, Zn. In addition, all of the isotopes under consideration except Fe^{59} and Co^{60} fall on or to the left of the line of beta stability, suggesting the process of spallation as a possible source of production. Of course, their formation by lower energy neutron reactions is also possible, although a greater number of elemental precursors would be required.

Summary

1. Chemical separations by ion-exchange resin column and precipitation techniques were made during January to April, 1957, on selected samples collected at the Pacific Proving Ground in June 1954 and September 1956. The radioactive isotopes contained in the separated fractions were identified by determination of rates of decay, maximum beta energies, and gamma energies.

2. The distribution pattern of radioactive non-fission products in plants and animals at the Pacific Proving Ground was studied by means of gamma spectrometry on whole samples. The gamma peaks of Mn^{54}, Co^{57}, Co^{58}, Co^{60}, and Zn^{65} were used for identification in this survey.

3. In a soil sample from Belle Island, Eniwetok Atoll, collected September 19, 1956, almost all of the radioactivity was due to fission products, with Ru^{106}-Rh^{106} and trivalent rare earths accounting for approximately 90 per cent of the total activity. Of the non-fission radioisotopes only cobalt was detected -- at a level of one to two per cent.

4. In a clam kidney sample collected at Belle Island, Eniwetok Atoll, September 22, 1956, fission products accounted for only 3.5 per cent of the total radioactivity. Fe^{55}, Co^{57}, Co^{58}, Co^{60}, Mn^{54}, and Fe^{59} comprised the remaining 96.5 per cent of the radioactivity.

5. In two fish liver samples collected at Bikini Atoll September 22 and 23, 1956, fission products were not present.

Fe^{55} and Zn^{65} were the principal radioactive constituents, with Mn^{54}, Co^{57}, Co^{58}, and Co^{60} contributing the remainder of the activity.

6. In fish liver samples collected at Belle Island, Eniwetok Atoll, May and June, 1954, Fe^{55} accounted for 95.3 per cent and Zn^{65} 3.1 per cent of the total activity. Mn^{54}, Co^{57}, and Co^{60} also were detected.

7. The pattern of distribution of the transition elements manganese, iron, cobalt, and zinc in the plants and animals at the Pacific Proving Ground is as follows. In general, they are present in the sea and not on the land, in marine animals and not in marine plants. Few or none of the radioactive transition elements are present in island soil, in plants growing in the soil, or in the herbivorous field rats (<u>Rattus exulans</u>). However, in the plankton, the marine invertebrate filter feeders and omnivores, and in the fishes, the above named isotopes are present and may contribute up to 100 per cent of the total radioactivity.

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Appendix Table 1. Volumes in milliliters of eluted fractions obtained in the ion-exchange experiments with Dowex 50

	Experiment Number						
Fraction	I	II	III	IVA	IVB	V	Spikes
Anions	99	49	50	230	4.5	25	25
HCl wash	60	20	30	---	---	22	20
0.5% Oxalic acid	200	40	40	37	28	42	40
0.15 N NH_4Cl + 0.1 N HCl	0	0	0	40	26	38	40
Ammonium citrate - 5%							
pH 3.5	260	60	64	62	60	55	60
3.1	140						
3.3	360						
3.4	400						
pH 4.1	200	57	40	33	46	42	40
pH 4.6	200	30	40	30	32	30	30
pH 5.1	200	20	40	20	20	20	20
pH 5.6	200	29	40	30	31	30	30
pH 6.1	200	22	40	22	21	20	20

Appendix Table 2. Volumes in milliliters of eluted fractions obtained in the ion-exchange experiments with Dowex 1

	Experiment Number	
Fraction	III	IV
12 M HCl	11.7	28
6 M	9.2	10
4 M	8.3	10
2.5 M	8.2	15
0.5 M	9.8	20
0.005 M	11.6	30
Distilled water	9.8	--

Appendix Table 3. Summary of data and calculations on four biological samples in which chemical separations were made

I Clam kidney (1.1 gm wet weight; .142 gm ash weight)
Belle Is.,
Eniwetok Atoll
9-22-56

Isotope	d/m in sample	d/m/g wet weight	uc/g wet weight	uc/cc MPA	observed MPA
$Ru^{106}Rh^{106}$	13,000	11,800	5.4x10^{-3}	1x10^{-1}	.05
$Zr^{95}Nb^{95}$	2,710	2,460	1.1x10^{-3}	---	---
Y^{91}	46,400	42,200	1.9x10^{-2}	2x10^{-1}	.1
Mn^{54}	38,500	35,000	1.6x10^{-2}	1.3x10^{-3}	12
Fe^{55}	1,300,000	1,180,000	5.4x10^{-1}	4x10^{-3}	140
Fe^{59}	2,680	2,440	1.1x10^{-3}	1x10^{-4}	11
Co^{57}	170,000	155,000	7.0x10^{-2}	7.2x10^{-3}	9.7
Co^{58}	162,000	147,000	6.7x10^{-2}	1.5x10^{-3}	44.7
Co^{60}	32,200	29,300	1.3x10^{-2}	2x10^{-2}	.65

II Bonito liver (8.9 gm wet weight; .202 gm ash weight)
Fox Is.,
Bikini Atoll
9-22-56

Isotope	d/m in sample	d/m/g wet weight	uc/g wet weight	uc/cc MPA	observed MPA
Mn^{54}	8,200	920	4.2x10^{-4}	1.3x10^{-3}	.32
Fe^{55}	250,000	28,100	1.3x10^{-2}	4x10^{-3}	3.2
Fe^{59}	488	55	---	---	---
Co^{57}	12,900	1,450	6.6x10^{-4}	7.2x10^{-3}	.09
Co^{58}	4,130	464	2.1x10^{-4}	1.5x10^{-3}	.14
Co^{60}	13,400	1,510	6.9x10^{-4}	2x10^{-2}	.03
Zn^{65}	156,200	17,600	8x10^{-3}	6x10^{-2}	.13

Appendix Table 3. (continued)

III Fish liver homogenate (7.29 gm wet weight; .213 gm ash weight)
How Is.,
Bikini Atoll
9-22-56

Mulloidichthys samoensis	10	specimens
Acanthurus triostegus	2	"
Caranx melampygus	1	"
Epinephalus merra	2	"
Epinephalus macrospilos	2	"
Variola louti	1	"
Priacanthus carolines	1	"

Isotope	d/m in sample	d/m/g wet weight	uc/g wet weight	uc/cc MPA	observed MPA
Mn^{54}	8,600	1,180	$5.4x10^{-4}$	$1.3x10^{-3}$	.42
Fe^{55}	20,600	2,830	$1.3x10^{-3}$	$4x10^{-3}$	.32
Fe^{59}	---	---	---	$1x10^{-4}$	---
Co^{57}	11,000	1,510	$6.9x10^{-4}$	$7.2x10^{-3}$	.10
Co^{58}	4,800	658	$3.0x10^{-4}$	$1.5x10^{-3}$	.20
Co^{60}	9,800	1,340	$6.1x10^{-4}$	$2x10^{-2}$	.03
Zn^{65}	78,700	10,800	$4.9x10^{-3}$	$6x10^{-2}$	.08

IV Fish liver homogenate (19.1 gm wet weight; .491 gm ash weight)
Belle Is.,
Eniwetok Atoll
May, June, 1954

Isotope	d/m in sample	d/m/g wet weight	uc/g wet weight	uc/cc MPA	observed MPA
Mn^{54}	4,280	224	$1.0x10^{-4}$	$1.3x10^{-3}$	.08
Fe^{55}	888,000	46,500	$2.1x10^{-2}$	$4x10^{-3}$	5.3
Co^{57}	1,400	73	$3.3x10^{-5}$	$7.2x10^{-3}$	.005
Co^{60}	7,300	382	$1.7x10^{-4}$	$2x10^{-2}$	.008
Zn^{65}	29,000	1,320	$6.0x10^{-4}$	$6x10^{-2}$	.01