Paper Chromatography of Radioactive Substances. (Radiochemical Studies on "Bikini Ashes" (March 1, 1954), Part III). (Studies of the Analytical Chemistry on Filter Paper XVI)

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Since Consden, Gordon and Martin[1] found in 1944 that filter paper sheets can be used as support of a stationary phase in partition chromatography, radioactive tracer techniques are applied to the identification of the substances developed on paper without the aid of reagents. For example, Wieland et al.[2] determined amino-acids by chromatographing their Cu salts containing radioactive Cu.

The methods for the separation of radioactive substances by means of ion exchange resins developed very rapidly by the studies carried out in connection with the work on atomic energy in the U. S. A.[3-13]. Tompkins, Khym and Cohn[14] found that a mixture of fission products which passed through a column of Amberlite IR-I (HR-form) to separate anions could be further separated into three groups by washing the column with oxalic acid to remove Zr and Hf, then with citrate at pH 3 to remove rare earths, and then with citrate at pH 5 to remove alkaline earths.

On the other hand, the separation of radioactive substances by paper chromatography has also been studied by several workers, but not so fully as ion exchange resins. For example, radioactive ^{42}K and ^{24}Na were separated by Frierson and Jones[15]. Bouissieres and Lederer[16] separated Ra and Ba using ethanol- water (80 : 20) as solvent, ^{22}Na and Mg with ethanol-20% water. By paper electromigration, Sato, Norris and Strain[17] obtained a good separation of many radioactive elements with 0.1 M lactic acid. In Japan, the separation of the radioactive substances adhering on the surface of tunny was also studied with the aid of paper chromatography by Kiba et al[18].

As above mentioned, the separation of radioactive substances by paper chromatography has not been investigated in detail, therefore many problems remain to be solved in a fundamental field. The important variables which have influence on the degree of separation, R_f values, purity and spot shape are: (I) carrier and its quantity; (2) other inactive substances (such as scavenger, analogous element, impurities in paper and solvent); (3) filter paper; (4) solvent; (5) self-absorption of radiation in paper (thickness of paper)[19]; (6) quantity spotted on original point; (7) temperature.

The author studied paper chromatography of radioactive substances in connection with ion-exchange resins so as to examine these effects in detail.

Experimental and Results

Separation by Ion-exchange Resins into four Groups.—According to Tompkins, Khym and Cohn[14], a mixture of fission products are separated into about four groups, and the nuclides in each group are further separated by ion exchange resins. The author tried the separation of nuclides in each group, which had been previously separated by ion exchange resins as shown in Fig. 1, with the aid of paper chromatography. The author treated the nuclides which are described in each group in Fig. 1. The obtained result is as follows.

Filter Paper.—Sheets of the Toyo filter paper

1) R. Consden, A. H. Gordon and A. J. P. Martin, *Biochem. J.*, 38, 224 (1944).
2) T. Wieland, K. Schmeiser, E. Fischer and H. Maier-Laibnitz, *Naturwissenschaften*, 36, 280 (1949).
3) B.H. Ketelle and G.E. Boyd, *J. Am. Chem. Soc.*, 69, 2800 (1947).
4) F. H. Spedding, *Discuss. Faraday Soc.*, 7, 214 (1949).
5) L.B. Werner and I. Perlman, *USAECD*, 1898.
6) S.G. Thompson, R. A. James and L. O. Morgar, *USAECD*, 1907.
7) K. Street Jr. and G.T. Seaborg, *J. Am. Chem. Soc.*, 72, 2790 (1950).
8) S.G. Thompson, B.B. Cunningham and G.T. Seaborg, ibid., 72, 2798 (1950).
9) K. Street Jr., S.G. Thompson and G.T. Seaborg, ibid., 72, 4832 (1950).
10) L.B. Magnusson, S.G. Thompson and G.T. Seaborg, *Phys. Rev.*, 78, 363 (1950).
11) R. Bouchez and G. Kayas, *Compt. rend.*, 228, 1222 (1949).
12) G. Kayas, *J. Chim. Phys.*, 47, 408 (1950).
13) E.R. Tompkins, ibid., 70, 3520 (1948).
14) E.R. Tompkins, J.X. Khym and W.E. Cohn, *J. Am. Chem. Soc.*, 69, 2769 (1947).
15) W.J. Frierson and J.W. Jones, *Anal. Chem.*, 23, 1447 (1951).
16) G. Bouissieres and M. Lederer, E. Lederer, M. Lederer and A.T. James, "Chromatography", Elsevier Publishing Co., Amsterdam, (1954), p. 331.
17) T.R. Sato, W.P. Norris and H.H. Strain, *Anal. Chem.*, 26, 267 (1954).
18) T. Kiba, S. Ohashi, M. Sibata and T. Mizube, *Japan Analyst*, 3, 361 (1954).
19) F. P. W. Winteringham, A. Harrison and R.G. Bridges, *Analyst*, 77, 19 (1952).

No. 5A or No. 5B (40×32 cm., with up to 8 samples on one sheet) are commonly used so as to prevent the influence of paper. When the development is completed (25 cm.), the sheet is hung for drying andc ut into strips (4×32 cm.) for measurement.

mm. in width and trailing the paper strip 10 mm. to every five minutes. Furthermore, the determination of each nuclide is done by the use of Nuclear Type 183, G. M. Counter having a mica window with a thickness of 1.4 mg./cm².

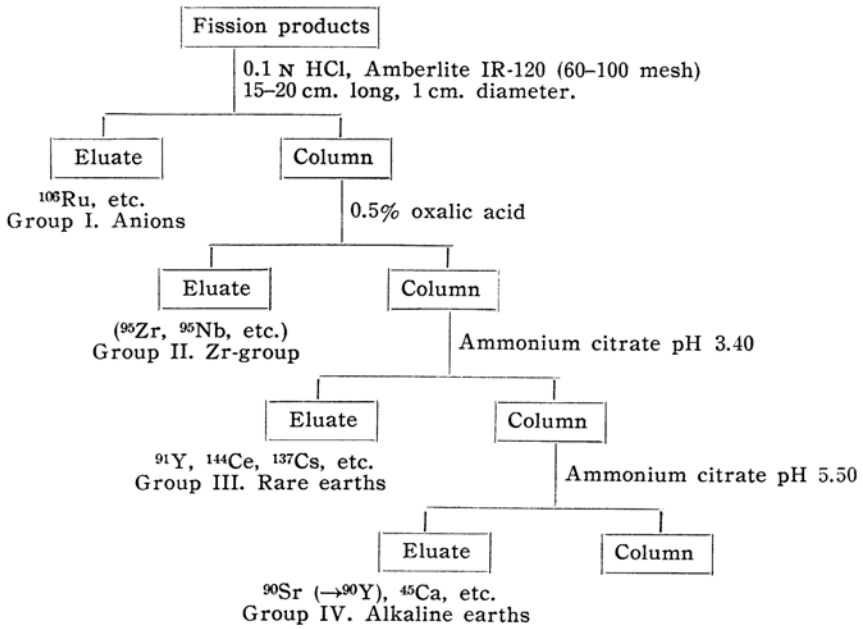


Fig. 1. Separation by ion exchange resins into four groups.

Test Solution.—The test solution separated from ion exchange resins or by distillation are concentrated to 3–10 cc. on the water bath, therefore, those solutions contain ammonium citrate, etc. (pH about 2–6).

Measurement of Radioactivity.—The distribution of radioactive substances on filter paper is measured by the Lauritzen electroscope with the author's continuous scanning device as shown in Fig. 2. It consists of lead plate (3 g./cm²) with paper holder and a slit which can be regulated by a screw. It is inserted into the gutter of the ionization chamber, and a filter paper strip is moved between the slit and the holder.

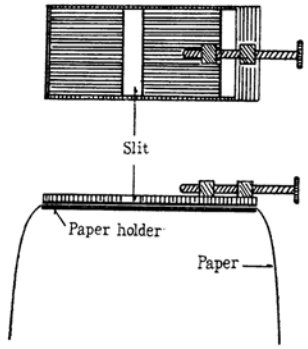
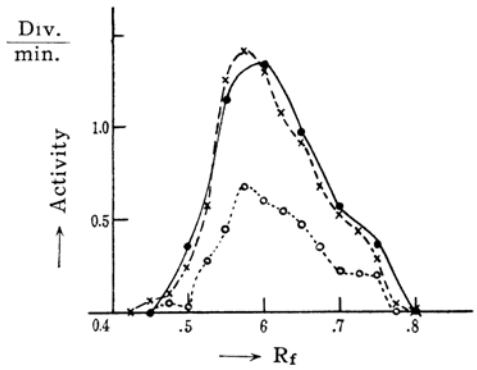


Fig. 2. Scanning slit of lead plate.

As shown in Fig. 3, it was observed that the best result is obtained by keeping the slit to 10



Methanol 9, c. NHO_3 1, Toyo filter paper No. 5B 15°–16°C, 4 hr./25 cm.

Fig. 3. Measurement of radioactivity using Lauritzenelectroscope.

•—•—• Slit: 10 mm. in width, trailing; 10 mm./5 min.

o···o···o Slit: 10 mm. in width, trailing; 5 mm./5min.

×···×···× Slit: 10 mm. in width, trailing: 5 mm./5 min.

(1) Separation of Group I (Anions).—At first, the author tried the separation of anions by using a sample solution separated with the aid of ion exchange resins from "Bikini Ashes" by the above mentioned method on May 24, 1954 (Fig. 4). Iso-amylalcohol-HCl (74 : 26) was used

as solvent. But the radioactivity was so low that nuclides could not be determined thoroughly. ^{35}S was reported on June 25, 1954 by the Tokyo University later.

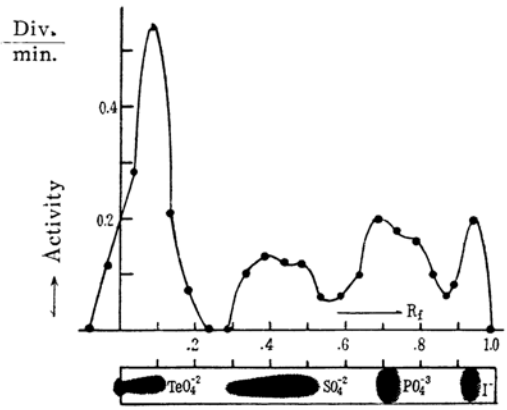


Fig. 4. Radio-chromatogram of anions (Group I).

{iso-Amylalcohol 74
{conc. HCl 26

Toyo filter paper No. 5A; 22°C.; 22 hr./25 cm. (SO_4^{-2} and PO_4^{-3} could be detected by chemical reactions.)

Fig. 5 is the radio-chromatogram of carrier-free ^{106}Ru separated from "Bikini Ashes" and ^{235}U-fission products by ion exchange resins. It is interesting that ^{106}Ru appears at two separated spots. As shown in Fig. 6, however, ^{106}Ru is detected as a spot extending over the whole paper with methanol-HNO_3 (9:1). This figure contains the radio-chromatograms of carrier-free ^{106}Ru derived from ion exchange resins and the distillation method.

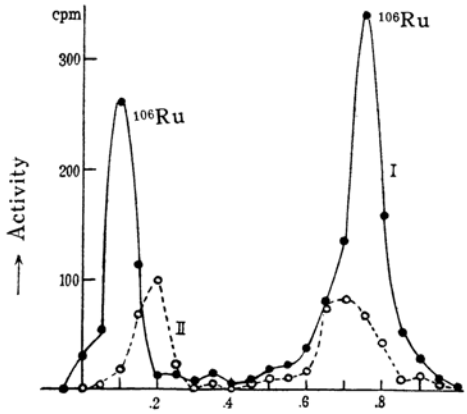


Fig. 5. Radio-chromatograms of ^{106}Ru (Group I).

{iso-Amylalcohol 74
{conc. HCl 26

From "Bikini Ashes". Quantity spotted on O.P.:
I: Large amounts.
II: Small amounts.
The same result was obtained in respect of a sample from ^{235}U-fission products.

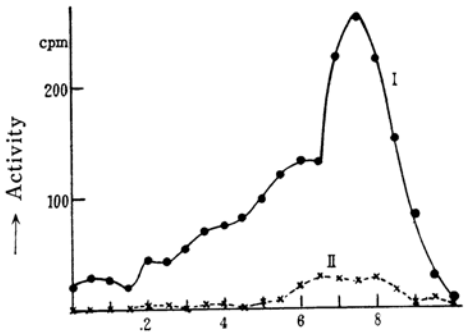


Fig. 6. Radio-chromatograms of ^{106}Ru.

{Methanol 9
{conc. HNO_3 1

Toyo filter paper No. 5B.
Sample solution: I: From ion exchange resins: II: From distillation.

(2) Separation of Group II(Zr-group).—The author has not obtained results to report here about this group yet, because the test solution was used for an other experiment by another student.

(3) Separation of Group III(Rate Earths).—A sample solution separated with the aid of ion exchanger from ^{235}U-fission products by the above mentioned method was developed with methanol-HNO_3(9:1). It is observed clearly in Fig. 7 that ^{137}Cs (alkali metal) is contained within this portion of eluates from the ion exchanger, and that ^{144}Ce and ^{137}Cs are separated completely. But also that ^{137}Cs was not found in the "Bikini Ashes" by this method.

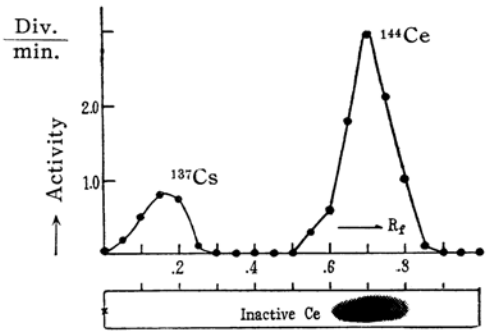


Fig. 7. Radio-chromatogram of rare earths (Group III).

{Methanol 9
{conc. HNO_3 1

Toyo filter paper No. 5B; 15°–16°C.; 4 hr./25 cm.
From ^{235}U-fission products.

A radio-chromatogram of ^{91}Y obtained from "Bikini Ashes" using ion exchange resins is shown in Fig. 8.

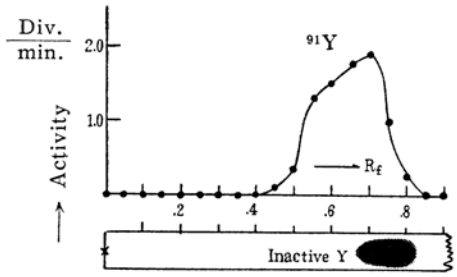


Fig. 8. Radio-chromatogram of rare earths (Group III).

{Methanol 9
{conc. NHO_3 1

Toyo filter paper No. 5B; 15°–16°C; 4 hr. /25 cm. From "Bikini Ashes".

In both chromatograms developed with methanol-HNO_3 (9:1), R_f values of carrier-free ^{144}Ce and ^{91}Y agree with R_f values of inactive Ce and Y nitrates (spot on paper strip in figure, $R_f=0.70$ and 0.75 respectively). When 1% oxine in glacial acetic acid is used as solvent, however, R_f values of carrier-free radioactive ^{144}Ce and ^{91}Y are not in agreement with R_f values of inactive Ce and Y respectively. In Figs. 9 and 10, ^{144}Ce containing ^{137}Cs and ^{91}Y are developed with 1% oxine in glacial acetic acid. In Fig. 9, inactive Ce separates to two spots ($R_f=0.00$ and 0.86), but carrier-free radioactive ^{144}Ce travels to $R_f=0.85$ only. The behavior of ^{144}Ce containing inactive Ce as carrier has also a different appearance as shown later. The peak of $R_f=0.5$–0.65 may be due to ^{137}Cs. In Fig. 10, the peak of ^{91}Y is found at $R_f=0.30$, but inactive Y has an $R_f=0.48$.

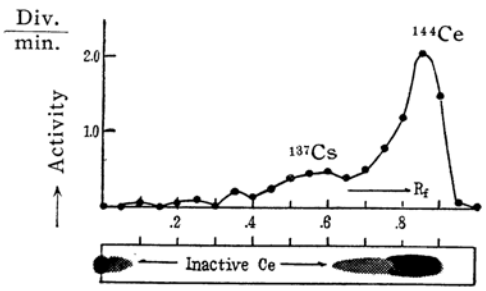


Fig. 9. Radio-chromatogram of rare earths (Group III).

1% oxine in gl. acetic acid. Toyo filter paper No. 5A; 15°–16°C; 5 hr./25 cm. From ^{235}U-fission products.

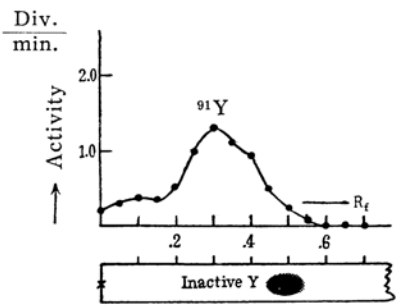


Fig. 10. Ralio-chromatogram of rare earths (Group III).

1% oxine in gl. acetic acid. Toyo filter paper No. 5A; 15°–16°C; 5 hr./25 cm. From "Bikini Ashes".

(4) Separation of Group IV(Alkaline Earths).—The typical results of separation of alkaline earths using methanol-HNO_3 (9:1) or 1% oxine in glacial acetic acid as solvent are shown in Figs. 11, 12, 13 and 14. A sample solution of carrier-free ^{90}Sr (containing ^{90}Y as its daughter) was separated by means of ion exchange resins from ^{235}U-fission products by the above mentioned method. A solution of ^{45}Ca was obtained from the liver of a tunny and separated using ion exchanger. It may contain inactive Ca. But the radioactivity was so low that ^{45}Ca could not be determined thoroughly (whether 0.7–0.8 or 0.0–0.4). In case of methanol-HNO_3 (9:1), R_f values of radioactive carrier-free ^{90}Sr, ^{90}Y and pure ^{90}Y separated from ^{90}Sr nearly agree with R_f values of inactive Sr and Y. In case of 1% oxine in glacial acetic acid, however, R_f values of radio-

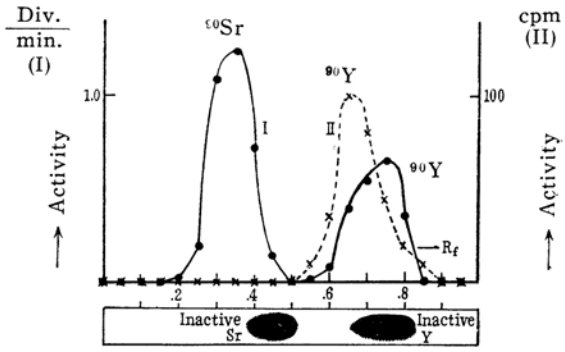


Fig. 11. Radio-chromatogram of alkaline earths (Group IV).

{Methanol 9
{conc. HNO_3 1

Toyo filter paper No. 5B; 15°–16°C; 4 hr. /25 cm. From ^{235}U-fission products.
I: ^{90}Sr containing ^{90}Y. II; Pure ^{90}Y.

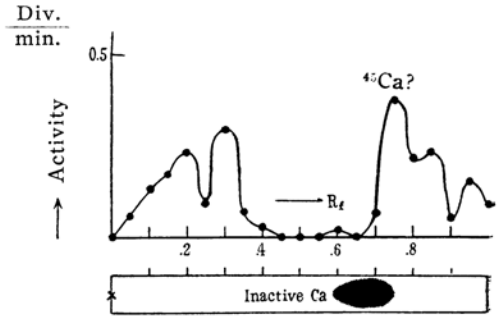


Fig. 12. Radio-chromatogram of alkaline earths (Group IV).

Toyo filter paper No. 5B; 4 hr./25 cm. From liver of tunny.

{Methanol 9
{conc. HNO_3 1

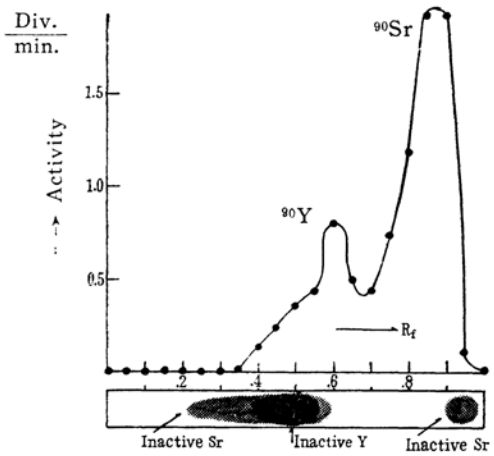


Fig. 13. Radio-chromatogram of alkaline earths (Group IV).
1% oxine in gl. acetic acid. Toyo filter paper No. 5A; 15°–16°C; 5 hr./25 cm.
From ^{235}U-fission products.

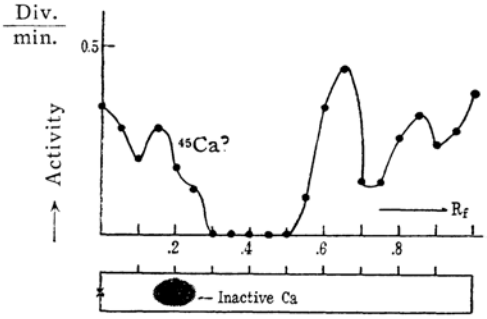


Fig. 14. Radio-chromatogram of alkaline earths (Group IV).
1% oxine in gl. acetic acid. Toyo filter paper No. 5A; 15°–16°C; 5 hr./25 cm.
From liver of tunny.

active and inactive elements are different from those shown in Fig. 13. That is, ^{90}Sr travels to R_f=0.87 which corresponds to one of the spots of inactive Sr (R_f=0.40 and 0.95), and R_f values of radioactive ^{90}Y and inactive Y are 0.60 and 0.45 respectively. The position of ^{45}Ca could not be determined as low counts in both of the solvents. In addition, as shown in Figs. 10 and 13 with 1% oxine in glacial acetic acid, it is interesting that R_f values of radioactive ^{91}Y, ^{90}Y and inactive Y are 0.30, 0.60 and 0.45 respectively.

(5) Influence of Carrier or other Elements.— As above shown, in 1% oxine in glacial acetic acid, the behavior of radioactive elements differs from that of the inactive elements. Therefore, it is very clear that the existence of carrier or other elements have some influence upon R_f value of a radioactive element. The qualitative results obtained are shown in Fig. 15. In this experiment, 1% oxine in glacial acetic acid is used as solvent. The strips ,a, b and c in Fig. 15 represent the movements of inactive Ce, ^{144}Ce (containing

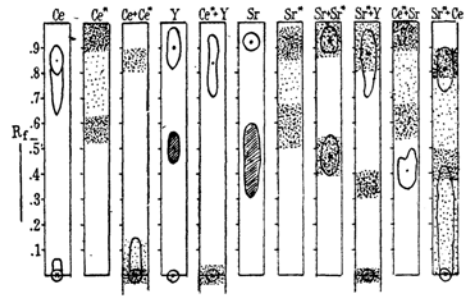


Fig. 15. Influence of carrier or other elements.
1% oxine in gl. acetic acid. Toyo filter paper No. 5A; 15°–16°C. 5hr./25 cm.
Ce* contains Cs*. Sr* contains ^{90}Y.

: Represents inactive element (Ce, Y and Sr).
: Represents position of a large part of ion.
* : Represents radioactive elements.
: Represents radioactivity

^{137}Cs), and ^{144}Ce with inactive Ce as carrier respectively. These strips show the influence of the carrier on ^{144}Ce containing ^{137}Cs (radioactivity of an R_f of 0.00). From the strips, b, d and e, it is clear that an addition of inactive Y to ^{144}Ce alters the R_f of ^{144}Ce remarkably. The results with respect to Sr and ^{90}Sr (containing ^{90}Y) are shown in the strips, f, g and h. The influence of inactive Y and Ce upon radioactive ^{90}Sr are remarkable as shown in the strips, i and k. This phenomenon may be due to ^{90}Y in ^{90}Sr. There is no influence of inactive Sr on ^{144}Ce (containing ^{137}Cs), and the former is not detected at R_f=0.92 (the strip, j). But a limit of detection of inactive Sr, as above inactive Ce and Y, is a question.

Discussion

The present author studied the paper chromatography of radioactive substances in connection with ion exchange resins, and obtained the results that nuclides can rapidly be determined by applying the paper chromatography to the separation of radioactive elements of the four groups eluated from ion exchange resins. In particular, the author knew that the factors influencing on R_f value and degree of separation of radioactive substances are (I) carrier or analogous element; (2) quantity spotted on original point of filter paper. These effects are shown in Figs. 15 and 5. Furtheremore, the R_f values of inactive and active elements are generally ^{144}Ce≤Ce (inactive) (Fig. 7); ^{90}Sr<Sr (inactive), ^{90}Y<Y (inactive) (Fig. 11) in methanol-HNO_3, and are ^{144}Ce≤Ce (Fig. 9); ^{91}Y<Y (Fig. 10); ^{90}Sr<Sr, ^{90}Y>Y (may be due to ^{90}Sr, Fig. 13) in 1% oxine in glacial acetic acid. That is, as shown in the reports with respect to

the influence of cations on the R_f values of anions (Cl^-, NO_3^- and SO_4^{2-})[20], the R_f values of the element of a large mass are smaller than those of the element of a small mass. But it can not be concluded in these experiments that the difference of an R_f of inactive and active element is due to a mass number. Because a difference of quantity spotted on the original point of paper between inactive and active solutions, a different complex formation in very dilute solution and the existence of some other substance in an active sample solution (ammonium citrate) in place of nitrate of inactive sample solution. It is the author's wish to study these questions quantitatively.

The author wishes to express his heartfelt thanks to Prof. Takanobu Siokawa, Sizuoka University, for his kind advice and encouragement.

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20) S. Nakano, *J. Chem. Soc. Japan*, **75**, 150 (1954).