

# Potential Column Chromatography for Ionic Ga-68. II: Organic Ion Exchangers as Chromatographic Supports

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**A potential Ge-68 → Ga-68 generator system, based on the adsorption of Ge-68 from a dilute hydrofluoric acid eluent onto a commercially available organic anion-exchange resin, is described. The distribution coefficients between the resin and dilute hydrofluoric acid were measured for germanium and gallium, and the breakthrough of germanium from a generator column determined. Using 0.01 N HF, the  $K_D$  values are 27 and >4,000 for gallium and germanium, respectively. Gallium-68 can be quantitatively collected with 4 ml of 0.01 N HF. After neutralization, this provides a biologically safe concentration of NaF for injection. The breakthrough levels of germanium-68 remain lower than  $10^{-4}$  for up to 600 collections.**

**J Nucl Med 21: 81-83, 1980**

The availability of a suitable generator for Ga-68 is of importance in the development of clinically useful positron emission tomography. Among previously developed generator systems (1-7) only a few (3,4) deliver ionic Ga-68 by a chromatographic process. In an earlier paper (8) some of the conditions a suitable generator should meet were enumerated and several chromatographic systems, based on inorganic substrates, were discussed with respect to their application as Ge-68 → Ga-68 generators. In this paper we describe a potential Ga-68 generator, based on a commercially available organic anion-exchange resin.

The anion-exchange behavior of most of the stable elements on Dowex AG1-X8 in hydrochloric acid has been measured by Kraus and Nelson (9). The  $\text{GaCl}_6^{3-}$  complexes are adsorbed strongly by the very basic resin from hydrochloric acid concentrations greater than 3 N, whereas  $\text{GeCl}_4$  is always more weakly adsorbed and only from 6 N or more concentrated hydrochloric acid, presumably due to ready hydrolysis of the germanium tetrahalides in weakly acidic conditions. Such a system, therefore, would not be acceptable as the basis for a ra-

dionuclide generator. From a pH 4 medium, germanium has been adsorbed on the anionite EDE-10P in the chloride form (10), but distribution coefficients have not been measured.

Anion-exchange behavior in hydrofluoric acid on a strongly basic anion-exchange resin, as measured by Faris (11), shows a far different picture. The heavier elements of the IIIB series are only weakly complexed by the fluoride, whereas the IVB elements form very stable complexes that are strongly adsorbed from hydrofluoric acid. In the case of germanium, this is due to the formation of germanium hexafluoride, which can readily replace the anions on the anion-exchange resin. The complexing of Ga(III) with fluoride as a function of fluoride concentration, and the stability constants of the complexes, have been studied (12,13). There are no anionic gallium fluoride complexes for fluoride concentrations less than  $10^{-3}$  molar. The separation of ionic gallium from germanium, which should still form a stable  $\text{GeF}_6^{2-}$  complex under these circumstances, appears possible on an anion-exchange resin.

## MATERIALS AND METHODS

**Measurement of distribution coefficients.** Anion-exchange resin\* in the chloride form (AG1-X8 200-400

Received May 29, 1979; revision accepted July 18, 1979.

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mesh) was used to measure the adsorption of both germanium and gallium from aqueous hydrofluoric acid and from mixtures of dilute hydrofluoric and hydrochloric acid. Before use, the resin was washed three times with distilled water and the fines decanted. It was then shaken with an excess of eluent, and this process was repeated three times. Distribution coefficients were determined by batch equilibration of 100-mg amounts of the resin with 5 ml of the eluents to which 1  $\mu$ Ci of a Ge-68  $\rightarrow$  Ga-68 equilibrium mixture had been added. After shaking for 10 min, the samples were centrifuged and 1-ml aliquots of the supernatants removed and assayed for Ga-68. After correction for decay between the end of equilibration and the activity measurement, this activity was compared with that of the original solution of the Ge-68  $\rightarrow$  Ga-68 equilibrium mixture. Distribution coefficients ( $K_D$ ) were calculated as:

$$K_D = \frac{\text{activity/gram of adsorbent}}{\text{activity/gram of eluent}}$$

The  $K_D$  values for germanium were obtained by counting the same samples for Ga-68 24 hr after equilibration.

**Experimental columns.** Two columns of AG1-X8 resin, chloride form, 200–400 mesh, were used: 1.3 ml (4 mm diameter by 10 cm high) and 0.25 ml (4 mm diameter by 2 cm high). They were loaded with 1  $\mu$ Ci of a Ge-68  $\rightarrow$  Ga-68 equilibrium mixture in aqueous hydrofluoric acid. The columns were eluted with dilute HF, and 2.0-ml and 1.0-ml fractions, respectively, were collected. The elution profile of Ga-68 and the breakthrough of Ge-68, as a function of the total eluted volumes, were measured. The breakthrough was expressed as percentage of the total Ge-68 loaded onto the resin.

A preliminary toxicity screening of the 0.01 *N* HF eluate from the small columns was performed after neutralization of the eluate with 0.3 *N* NaOH and addition of sufficient NaCl to make the solution 0.15 *N* in sodium salts. Mice were injected (i.v.) with single 0.5-ml fractions of this solution and observed over a 2-day period.

## RESULTS

The results of the batch equilibration studies are summarized in Fig. 1. The distribution coefficients ( $K_D$ ) for germanium on Bio-Rad AG1-X8 in dilute hydrofluoric acid were higher than 4,000 for acid normalities between  $10^{-3}$  and  $10^{-1}$ . The  $K_D$  values for gallium varied between 44 and 14, respectively. When hydrochloric acid mixtures in different concentrations were added to depress the gallium adsorption, the adsorption of germanium was also decreased, and it was found that the concentration of the hydrochloric acid had to be kept below 0.005 *N* in order to avoid significant germanium breakthrough.

Figure 2 shows the elution profile of Ga-68 from a

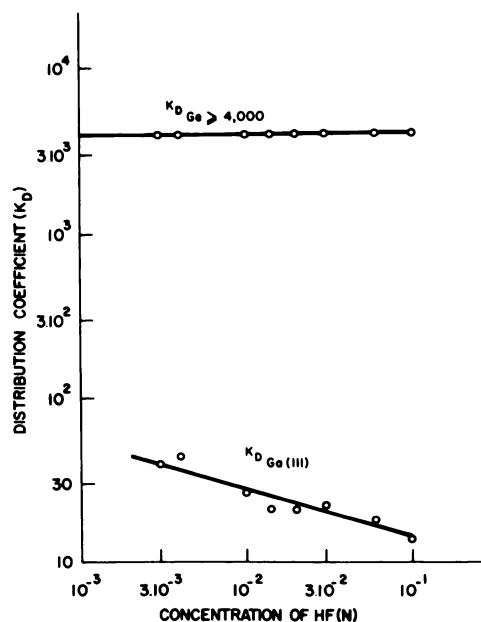


FIG. 1. Distribution coefficients (w/w) for germanium and gallium between AG1-X8, \* 200–400 mesh, and aqueous HF.

1.3-ml AG1-X8 anion-exchange\* column (Curve 3). The cumulative Ga-68 collection yield showed that 95% of all Ga-68 on the 1.3-ml column could be collected in 20 ml of 0.01 *N* HF (Curve 2), and on the 0.25-ml column in 4 ml (Curve 1).

Table 1 shows Ga-68 yield and the breakthrough of Ge-68 in 4-ml collection volumes, as a function of the total number of elutions from the column of Ga-68 with 0.01 *N* HF.

The injection of the neutralized isotonic eluates into mice caused no discernible effects. On a normalized weight basis, the injectate represented 400 times the dose received by a 70 kg human given an injection of 4 ml.

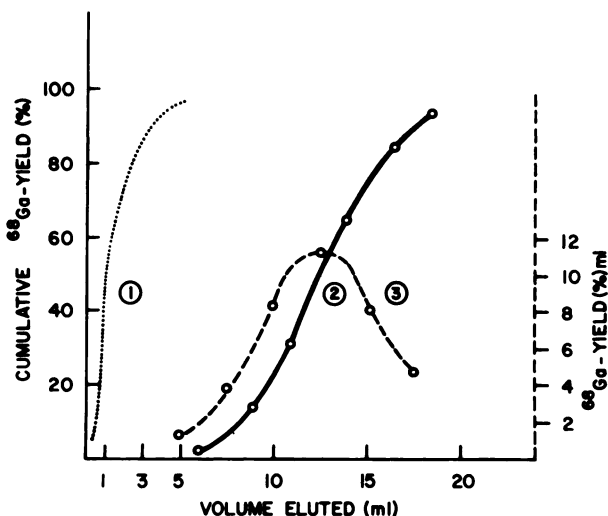


FIG. 2. Cumulative yield of Ga-68, eluted with 0.01 *N* HF from AG1-X8\* anion-exchange columns, with 0.25-ml bed (Curve 1) and 1.3-ml bed (Curve 2). Elution profile of Ga-68 from anion-exchange column with 1.3-ml bed (Curve 3).

**TABLE 1. ELUTION YIELD AND BREAKTHROUGH VALUES FROM A PROTOTYPIC Ge-68 → Ga-68 RADIONUCLIDE GENERATOR\***

Collection No.	Ga-68 yield (%)	Ge-68 breakthrough (% of Ge-68 loaded)
0-300	90 ± 3	<0.008
300-500	90 ± 3	<0.01
500-600	90 ± 3	<0.01

\* 0.25 ml AG1-X8, 200-400 mesh, anion-exchange resin columns eluted with 4-ml volumes of 0.01 N HF.

#### DISCUSSION

The high distribution coefficient for germanium between the anion-exchange resin and hydrofluoric acid should allow hundreds of collections of Ga-68 from a generator without appreciable loss of germanium. The hydrolysis of the  $\text{GeF}_6^{2-}$  complex, leading to early losses of germanium, is unlikely in dilute hydrofluoric acid. This was confirmed by the germanium breakthrough measurements as a function of the total volume eluted through the 0.25-ml column, which represented 600 collection volumes for Ga-68 from the generator.

The choice of the hydrofluoric acid concentration for the collection of Ga-68 depends on two conflicting considerations. The  $K_D$  for gallium is lowest for higher normalities of hydrofluoric acid. However, higher HF concentrations could mean increased toxicity of the final eluate. Lower HF concentrations necessitate the use of larger collection volumes for gallium, and the longer elution times and larger eluate volumes would make the generator less practical. In addition, since injectable Ga-68-labeled compounds should have ionic strengths close to that of blood, the generator eluate should also be isotonic. We therefore limited the acidity of the eluents to 0.1 N or less.

With 0.01 N HF, the amount of sodium fluoride in a neutralized 4-ml eluate would be  $4 \times 10^{-5}$  mole, or 1.7 mg. This would increase the fluoride content in normal human whole blood by 0.4 mg/liter—namely to twice that normally present. The safety of these fluoride preparations will depend on the speed with which fluoride is excreted or adsorbed. Studies with F-18 have shown that fluoride uptake into bone of 50% of the injected dose is virtually complete within 1 hr, while the remaining fluoride is rapidly excreted by the urine. Since fluoride taken orally is adsorbed very rapidly, a daily prophylactic fluoride dose contributes as much fluoride to the blood in a day as would a typical Ga-68 preparation in dilute sodium fluoride.

The complex of gallium with fluoride ions is much less stable than that with hydroxyl ions. The  $\log \beta$  value for the complex with fluoride is 5.1, at an ionic strength of

0.5, against 11.1 for the complex with hydroxyl ions. Therefore, fluoride at the concentrations under consideration is not expected to interfere with the preparation of Ga-68-labeled radiopharmaceuticals that are stable in vivo.

The generator described here has shown lower breakthrough values for Ge-68 and higher Ga-68 yields for a larger number of elutions than any of the previously described systems. Its major drawback is the question of the radiolytic stability of the organic matrix. A full-scale acute and subacute toxicity study in mice, rabbits, and dogs has been initiated in order to elucidate the relationship between the integrated radiation dose to the resin and its radiolytic breakdown in terms of decreased adsorptive capacity and increased release of organic material.

#### FOOTNOTE

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#### ACKNOWLEDGMENTS

This work was supported by DOE Contract No. EY-76-S-4115 and by NIH Grant No. GM 26780. The material was presented in part at the Second International Symposium on Radiopharmaceuticals, March 1979, Seattle, Washington, and at the June 1979 Annual Meeting of the Society of Nuclear Medicine, in Atlanta. Thanks are due to Rebekah Taube for help in the preparation of the manuscript, and to Daniella Neirinckx for the art work.

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