# Manganese-52m, A New Short-Lived, Generator-Produced Radionuclide: A Potential Tracer for Positron Tomography

Robert W. Atcher,* Arnold M. Friedman, John R. Huizenga, G. V. S. Rayudu, Edward A. Silverstein, and David A. Turner

*Argonne National Laboratory, Argonne, Illinois, University of Rochester, Rochester, New York, and Rush University Medical Center, Chicago, Illinois*

**A new generator system has been developed using the Fe-52 → Mn-52m parent-daughter pair. Fe-52, half-life 8.3 hr, is isolated on an anion-exchange column, and Mn-52m is eluted in hydrochloric acid. Breakthrough is less than 0.01% and the yield is 75%. The 21.1-min half life of Mn-52m is ideal for use in sequential studies, but is long enough to permit radiochemical manipulations to control biodistribution. Animal studies indicate that Mn-52m is an ideal nuclide for myocardial imaging, combining rapid blood clearance and high concentration in the myocardium. An added advantage is that Mn-52m decays 98% by positron emission and is useful for positron computer tomography.**

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Interest in the use of nuclear medicine techniques for dynamic or sequential studies has pointed out the limitations of Tc-99m. Its relatively long (6 hr) half-life, reduces its applicability for studies in which the tracer has a biological half-time on the order of minutes.

The recent advances in three-dimensional imaging with radionuclides, and the introduction of commercial imaging systems, have increased the interest in positron emitters for such studies. The drawback has been that C-11, N-13, O-15, and F-18 all require that the user be near a cyclotron.

In order to combine the advantages of three-dimensional imaging with those of short-lived radionuclides, we have developed a generator system based on the parent-daughter pair Fe-52 → Mn-52m. The parent (half-life 8.3 hr) was produced by a number of nuclear reactions in either cyclotrons or high-current proton linear accelerators (linacs) (*1*).

Fe-52 decays by positron emission (56.5%) and by electron capture (*2*). It emits a 168.8-keV gamma photon (99.2%). Mn-52m decays by positron emission (98.3%) with a 21.1-min half-life. The positron energy is 2.631 MeV. In addition to the annihilation radiation, Mn-52m emits a 1434-keV gamma (98.3%). The remainder of the decay is by isomeric transition to Mn-52, which has a 5.59 day half-life (Fig. 1).

Recent studies (*3*) have indicated that radionuclides of manganese have potential as myocardial imaging agents. The short half-life of Mn-52m should make it suitable for such studies, and biodistribution data for manganese are presented here.

## MATERIALS AND METHODS

**Preparation of the generator.** Fe-52 is currently prepared in a cyclotron by He-3 or He-4 bombardment of a chromium target, proton bombardment of a manganese target, or proton spallation reactions on a nickel target, usually in a high-current proton linac (*1*).

The yield of Fe-52 at 0.5 saturation (8.3 hr) ranges from 4 mCi for 30 MeV, 30 $\mu$A He-3 particles on chromium, to 8 Ci for a spallation reaction at the Los Alamos Meson Physics Facility's (LAMPF) isotope producer.

Fe-52 produced at Argonne National Laboratory's 60-in. cyclotron and at the Brookhaven Linac Isotope Producer (BLIP) has been used to test the generator.

* Present address: Dept. of Radiology, Peter Bent Brigham Hospital, 721 Huntington Ave., Boston, MA 02115.

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For reprints contact: A. M. Friedman, Chemistry Div., Argonne National Laboratory, Argonne, IL 60439.

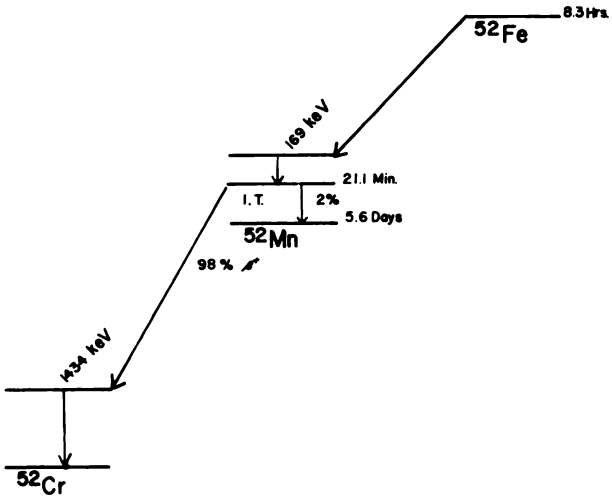


**FIG. 1.** Decay scheme of Fe-52 and Mn-52m.

Though the Fe-52 produced at BLIP for patient injection had been reduced with ascorbic acid to the II oxidation state, its treatment with concentrated nitric acid and hydrogen peroxide successfully oxidized it to the III oxidation state.

Separation schemes include either the isolation of Fe-52 on anion-exchange resin or extraction from hydrochloric acid into ether. The product is Fe-52 in the III oxidation state.

For this study, typically 2 mCi of Fe-52 were used in generator tests. Fe-52 was dissolved in 8 *N* HCl and percolated through an anion-exchange resin† (100–200 mesh, chloride form) that had been pre-equilibrated with 8 *N* HCl. The resin was supported by glass wool plugs in a 2-mm (i.d.) × 40-mm glass column. Unless a large concentration of carrier iron was present in the Fe-52 sample (from impurities in the reagents or targets) this size of column had sufficient capacity to bind the Fe-52 completely. Distribution coefficients for Fe(III) and Mn(II) are shown in Fig. 2 (*4*).

**Elution.** Mn-52m was eluted from the generator in 8 *N* HCl, two column volumes being sufficient (Fig. 3). Studies in 6 *N* and 9 *N* HCl were also performed.

The yield of Mn-52m was determined by gamma spectroscopy using a Ge(Li) detector and a multichannel analyzer. A spectrum was made from the column before elution and (using the same geometry) of the eluate after elution, and the 1434-keV gamma peak was integrated in both spectra. Breakthrough was determined by integrating the 169-keV gamma peak either at elution or after the Mn-52m had decayed.

The eluant was evaporated to dryness to remove HCl. The Mn-52m was dissolved in an 0.2 *M* acetate buffer, pH 5, for administration to animals. No suitable images were obtained.

In order to test the behavior of the generator over time, a generator was loaded with reactor-produced Fe-59 (15.4 mCi/mg) in the form of $FeCl_3$ and eluted 100 times in succession—more than a typical generator would experience in normal use.

**Tissue distribution studies.** Animal distributions were performed using the longer-lived Mn-54 to eliminate sample-counting problems associated with the short-lived Mn-52m. [$^{54}$Mn]manganous chloride in dilute HCl was evaporated to dryness and dissolved in 0.2 *M* acetate buffer, pH 5. This solution was passed through an anti-

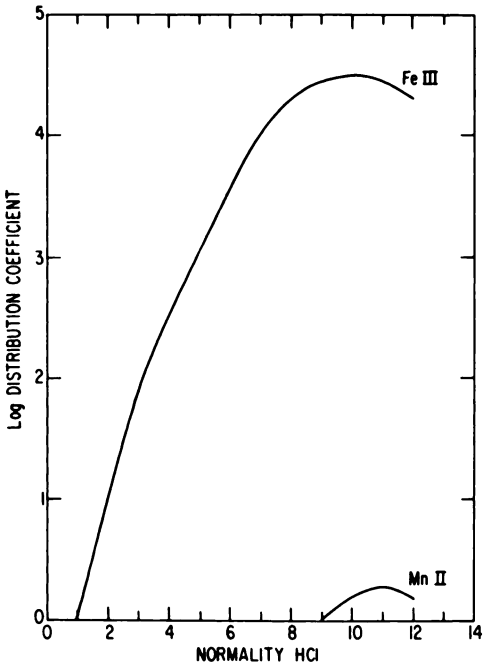


**FIG. 2.** Distribution coefficients for Fe(III) and Mn(II) in HCl on anion-exchange resin (*4*).

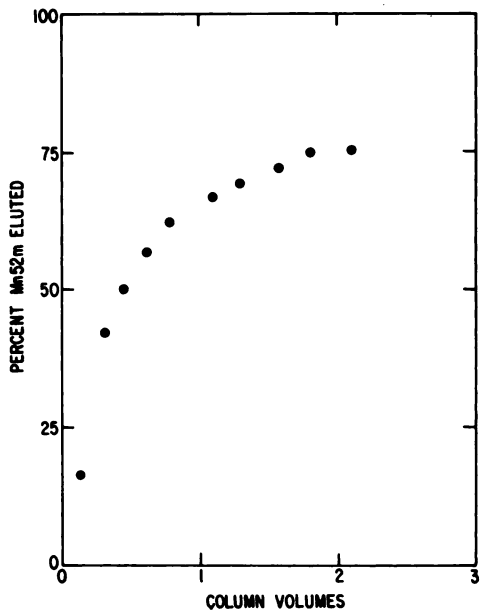


**FIG. 3.** Elution of Mn-52m in 8 *N* HCl from anion-exchange resin, 100–200 mesh.*

bacterial filter and injected into the tail vein of ICR female, white mice, 20–30 g, about 8 wk old.

Three mice each were killed at 5, 10, and 20 min, and 2 hr after injection. Organs were excised and counted in a NaI(Tl) well detector. About 1 $\mu$Ci was injected into each animal.

## RESULTS

Figure 3 shows a typical elution curve for the generator in 8 *N* HCl. Previous studies of the generator system (*5*) indicated that the Mn-52m yield was lower, but our yield was 75% per elution. In addition, the activity eluted in two column volumes rather than three, as previously reported.

With elution in 6 *N* HCl, the yield increased to an average of 89%, but breakthrough also increased to a maximum of 0.04%. In 9 *N* HCl, the yield decreased to 60% while breakthrough decreased to less than one part per million.

Figure 4 shows Ge(Li) spectra of the generator before elution, and of the eluate. The 169-keV gamma peak from the Fe-52 decay is not evident in the eluate. Additional information on the Fe-52 breakthrough was obtained by counting the samples 4 hr later, after the Mn-52m had decayed. With correction for Fe-52 decay, assay by integrating the 169-keV photopeak showed a breakthrough of less than 0.01% of the Fe-52 on the column.

Figure 5 shows the results for the performance of the column during 100 elutions. With few exceptions, breakthrough is less than 0.01%. This column was later loaded with 2 mCi of Fe-52 and performed as the others. Yield was 75% and breakthrough was less than one part per million.

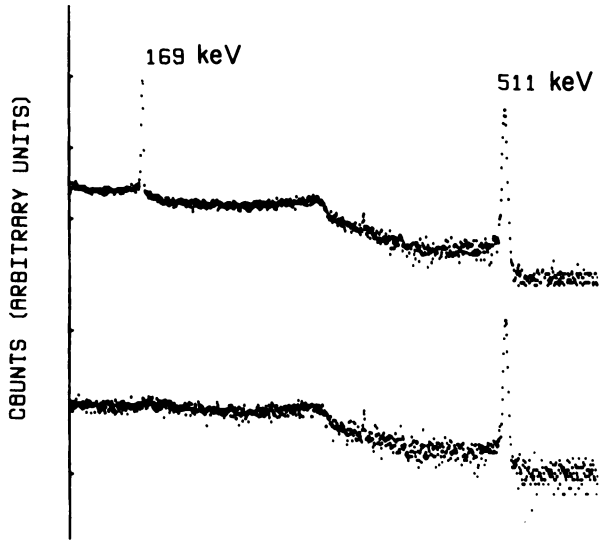


**FIG. 4.** Ge(Li) spectra (semilog) of the generator before elution (top), and of the eluate, show clean separation of Mn-52m from generator. Note absence of 169-keV peak in eluate.

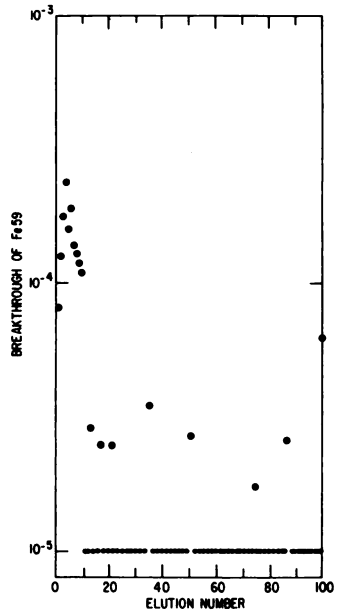


**FIG. 5.** Anion-exchange resin column loaded with Fe-59 was eluted 100 times with two column volumes per elution.

The results of the biodistribution studies are shown in Fig. 6 and Table 1. Values in the table include the ranges for the data. [$^{54}$Mn]manganous chloride clears the blood quickly after injection into the tail vein of the mice.

## DISCUSSION

Cyclotron production of Fe-52 yields a product with high radionuclidic purity. However, Fe-52 produced by proton reactions suffer enough from contamination with

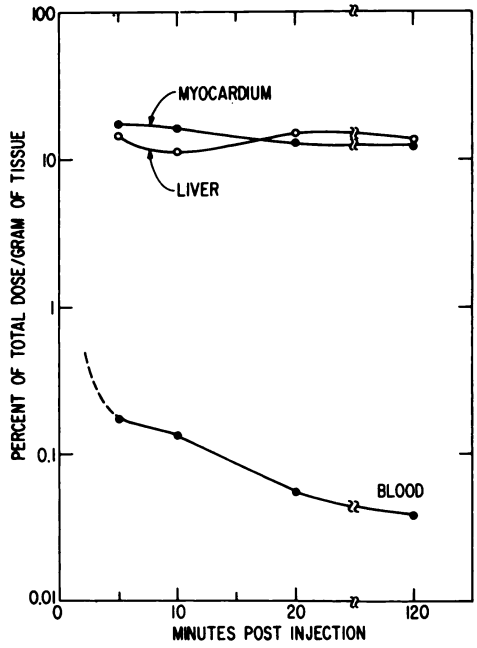


**FIG. 6.** Body distributions of Mn-54 as a function of time.

**TABLE 1. TISSUE CONCENTRATION OF Mn-54 IN MICE: PERCENTAGE OF TOTAL DOSE/GRAM OF TISSUE AND RANGE**

| | Time after injection | | | |
|---|---|---|---|---|
| Organ | 5 min | 10 min | 20 min | 2 hr |
| | 0.16 | 0.16 | 0.06 | 0.04 |
| Blood | 0.18–0.10 | 0.13–0.08 | 0.07–0.05 | 0.06–0.02 |
| | 17.34 | 16.53 | 2.79 | 2.89 |
| Myocardium | 18.54–16.14 | 17.63–15.43 | 13.00–2.89 | 12.85–3.31 |
| | 5.20 | 1.34 | 3.80 | 4.37 |
| Liver | 14.82–6.00 | 11.38–1.60 | 11.60–6.04 | 13.18–4.30 |
| | 3.30 | 1.35 | 7.29 | 10.01 |
| Kidney | 40.57–2.30 | 37.89–1.10 | 34.90–7.96 | 33.57–5.19 |
| | 1.40 | 0.45 | 4.67 | 1.06 |
| Lung | 8.25–1.20 | 7.03–0.26 | 4.83–4.51 | 6.07–1.59 |
| | Ratios of percent total dose/gram of tissue | | | |
| Myocardium/blood | 97.5 | 123.5 | 236 | 328.9 |
| Myocardium/liver | 1.17 | 1.47 | 1.12 | 0.97 |
| Myocardium/lung | 2.12 | 2.36 | 2.78 | 2.12 |
| Myocardium/kidney | 0.43 | 0.44 | 0.37 | 0.38 |

Fe-55—and, for spallation, Fe-59—to cause potential breakthrough problems (*1*). For example, a production run that produced 100 mCi of Fe-52 by spallation would also produce 2 mCi of Fe-55 and 0.15 mCi of Fe-59. While decay losses are significant for Fe-52, the longer half-lives of Fe-55 (2.7 yr) and Fe-59 (44.6 days) mean negligible losses of these nuclides over the life of the generator.

This potential problem from the longer-lived iron radionuclides dictated that breakthrough be held to a minimum. The HCl anion-exchange resin system has the highest distribution coefficient for iron of any of the commonly used eluants or exchange resins. At a normality of 8, the distribution coefficient for iron is about 10,000—near its maximum of 40,000 in 10 *N* HCl. Thus breakthrough problems will be minimal at this concentration of HCl.

At 8 *N* HCl, manganese has a distribution coefficient less than one, enabling one to recover nearly all the Mn-52m on the column. In fact, the ratio of the distribution coefficients of Fe(III) and Mn(II) goes through a maximum in 8 *N* HCl. Hydrochloric acid adds the advantages of being easily evaporated and yielding manganous chloride, a stable form of manganese. Manganese stays in the (II) oxidation state in acidic chloride media, whereas in nitric acid other oxidation states, mostly (IV), can form. The $MnO_2$, if formed, acts as a radiocolloid and is unreactive. The chloride left after the evaporation of the hydrochloric acid would present no toxicological hazard to the patient.

In 9 *N* HCl the reduced breakthrough of iron radionuclides was offset by the decreased Mn-52m yield. Conversely, in 6 *N* HCl the yield of Mn-52m increased, but the breakthrough increased as well.

The high breakthrough initially in the column loaded with Fe-59 could be attributed to the carrier iron present, since the nuclide is reactor-produced. The gradual decrease in breakthrough would be consistent with this, i.e., a peak of iron is coming off the column.

If 100 mCi produced by spallation were put on a generator, and the generator eluted in 8 *N* HCl, the maximum breakthrough of Fe-55 would be 200 nCi per elution, and for Fe-59 it would be 15 nCi per elution. Of course, the characteristics of a generator loaded with 100 mCi of Fe-52 may be different because of the higher radiation dose to the resin or other radiolytic effects.

The production capabilities of LAMPF, 10 Ci of Fe-52, enable one to produce generators that could be used over several days. If a generator were not eluted for several hours, the buildup of Mn-52 from the decay of Mn-52m could increase patient's radiation dose. A pre-elution of the generator about an hour before its intended use would remove the longer-lived Mn-52. Our attempts at using the generator product for imaging with conventional equipment were not satisfactory. The Compton scatter from the 1434-keV gamma of Mn-52m, and the high level of septal penetration, resulted in poor images. Recent reports have shown that Mn-52m can be used in emission computed tomography systems with good results (*6*).

For this reason, the mouse data were taken using Mn-54. Many agents were tested as potential myocardial imaging agents. Manganous chloride in acetate buffer gave the best results of the group tested, which included manganous phosphate and manganese complexes with ethylenediamine, ethylenediamine-tetraacetate, and diethylenetriaminepenta-acetic acid. The biodistribution showed that carrier-free radionuclides of manganese

cleared the blood rapidly, since when the dose is injected into the tail vein, 99+% is removed from the blood in 5 min. A study of myocardial distribution can be begun 5 min after administration of the radiopharmaceutical. The previous study (*3*) did not present data for a time shorter than 30 min.

The authors did point out that an infarct in a dog heart did show decreased uptake of Mn-54; uptake in the center of the infarct was 24% of the uptake in normal myocardium. The authors also pointed out that the myocardium-to-blood ratios were equal to or exceeded those for Tl-201.

It has been shown that isotopes of manganese are concentrated by the mitochondria (*7*). Nearly three fourths of all manganese was found in the organelles of the cell.

Using the organ distribution in mice for manganous chloride in acetate buffer, we have calculated the radiation doses for the organs of greatest interest by the method of Snyder et al. (*8*). The results are: 418 mrad/mCi to the kidneys, 174 mrad/mCi to the liver, 42 mrad/mCi to the bone, and 58 mrad/mCi to the whole body.

The rapid blood clearance of manganese is especially suited to a radionuclide with a short physical half-life. Indeed, if it is found that manganese redistributes in the heart, an improved exercise study could be performed with Mn-52m, whose physical half-life is better matched to biological half-life of blood flow to the heart than is that of Tl-201. Or, if the radionuclide does not redistribute, an exercise study could be performed 4 hr later; the rapid blood clearance would not extend the study's length.

Recent advances in attaching chelating agents to proteins open many exciting possibilities for radiopharmaceutical development (*9,10*). DTPA-labeled HSA microspheres could be used for visualization of lung perfusion.

While the attachment of chelating agents to macromolecules shows great promise for the development of new radiopharmaceuticals using inorganic radionuclides, care must be taken to deionize the water used in preparations and to use analytical-grade reagents. The small number of chelating units attached to the molecules could be saturated by metal ions in solution. The relative nonreactivity of Mn(II) compared with other transition elements dictates that all solutions be relatively free of ions competing for the chelating groups.

The potential application for Mn-52m in myocardial studies has been shown by its rapid clearance from the blood and high myocardial uptake. Further studies are being undertaken to develop radiopharmaceuticals for procedures that are better suited to Mn-52m's short half-life.

## FOOTNOTE

† Bio-Rad AG1X8, Bio-Rad Laboratories, Richmond, CA.

## ACKNOWLEDGMENT

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## REFERENCES

1. ATCHER RW, FRIEDMAN AM, HUIZENGA JR: Production of Fe 52 for use in a radionuclide generator. *Int J Nuc Med Biol,* in press
2. LEDERER CM, SHIRLEY VS: *Table of Isotopes.* 7th Ed. New York, John Wiley and Sons. 1978, pp 146–147
3. CHAUNCEY DM, SCHELBERT HR, HALPERN SE, et al: Tissue distribution studies with radioactive manganese: a potential agent for myocardial imaging. *J Nucl Med* 18: 933–936, 1977
4. KRAUSE KA, NELSON F: P/837, Proceedings of the International Conference on the Peaceful Uses of Atomic Energy, Geneva, 1955, 7, 113–116, (1956)
5. ATCHER RW, FRIEDMAN AM, HUIZENGA JR, et al: Mn-52m. A new short-lived, generator-produced radionuclide. *J Nucl Med* 19: 689, 1978 (abst)
6. HUI JCK, ATKINS HL, SOM P, et al: Manganese-52 positron emission transaxial tomography for detecting myocardial infarction. *J Nucl Med* 20: 648, 1979 (abst)
7. MAYNARD LS, COTZIAS GC: The partition of manganese among organs and intracellular organelles of the rat. *J Biol Chem* 214: 489–495, 1955
8. SNYDER WS, FORD MR, WARNER GG, et al: S, absorbed dose per unit cumulated activity for selected radionuclides and organs. MIRD Pamphlet No 11. New York, Society of Nuclear Medicine, 1975
9. KREJCAREK GE, TUCKER KL: Covalent attachment of chelating groups to macromolecules. *Biochem Biophys Res Commun* 77: 581–585, 1977
10. SUNDBERG MW, MEARES CF, GOODWIN DA, et al: Selective binding of metal ions to macromolecules using bifunctional analogs of EDTA. *J Med Chem* 17: 1304–1307, 1974