# Radiochemical Analysis of Commercial MDP Bone Kits

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***We performed radiochromatographic analysis of four commercial MDP bone kits to evaluate their performance under normal and less favorable conditions. Specifically, we evaluated the effects of (1) varying the type of storage container, (2) adding 3 cc of room air, and (3) adding high amounts of Tc-99m (400 mCi). Results indicate that the four commercial MDP kits were stable in vials and syringes up to 6 hr after preparation, but those kits containing ascorbate were better able to withstand the stress of high levels of radioactivity (400 mCi), storage in syringe, and addition of air.***

The provision of unit doses of radiopharmaceuticals from a centralized nuclear pharmacy has gained wide acceptance over the last few years. The rapidly increasing number of commercial nuclear pharmacies and the formation of central nuclear pharmacies in many large academic institutions attest to this. Hospitals purchasing radiopharmaceuticals from centralized facilities must rely on the nuclear pharmacy to perform adequate quality assurance procedures to ensure that the products they receive meet acceptable standards for radiochemical purity. This is critical because these radiopharmaceuticals could be subjected to more stressful conditions both during preparation and delivery than radiopharmaceuticals that are prepared in a hospital's own nuclear medicine department.

We undertook a study to evaluate the radiochemical purity of four MDP preparations by determining the levels of free pertechnetate and hydrolized reduced Tc-99m (Tc-HR) levels up to 24 hr after formulation under a variety of stressful conditions.

## Materials and Methods

Four MDP bone imaging agents were evaluated: MPI MDP Kit (Medi-Physics Inc.), Osteolite (New England Nuclear), MDP-Squibb (E.R. Squibb & Sons, Inc.), and Amerscan MDP (Amersham Corp.). The materials listed in each vial are shown in Table 1. Each vial of MDP was prepared according to package directions. The Squibb and Amersham kit package directions placed limits of 150 and 200 mCi of [$^{99m}$Tc] pertechnetate to be added to each kit, respectively. The Medi-Physics and New England Nuclear MDP kits did not list a maximum mCi amount of Tc-99m that could be added. They listed a suggested maximum volume for [$^{99m}$Tc] pertechnetate of 8.0 ml. For the Medi-Physics and New England Nuclear MDP kits, we injected 200 mCi of activity to simulate routine preparation in most hospital laboratories. All MDP kits were prepared so that the final volume was 5.0 ml. After injecting the [$^{99m}$Tc] pertechnetate and diluting to 5.0 ml with sterile, preservative-free 0.9% saline, the kits were allowed to incubate for 20 min.

We then evaluated the effect of the following parameters on the stability and radiochemical purity of the MDP kits up to 24 hr after reconstitution: (1) varying the storage container (vials versus syringes), (2) stressing the kits by injecting 3 cc of room air, and (3) adding excess Tc-99m (400 mCi). To simulate routine use, three samples of 0.8 ml each were drawn into 3-ml syringes. Depending on the experiment, the 0.8-ml samples were either discarded or placed in a syringe shield and kept at room temperature for radiochemical analysis at a later time.

The chromatographic procedures we used are summarized in Table 2. Whatman no. 3 chromatography paper (Whatman Chromatography Products, Clifton, NJ) and Gelman ITLC-SG sheets (Gelman Instrument Co., Ann Arbor, MI) were cut into 1 × 15 cm strips. Pencil lines were drawn on the strips at points 1.5, 6.5, and 11.5 cm from the bottom of the strip. The Whatman no. 3 strips were spotted with a dot of permanent ink and the Gelman ITLC-SG strips with a dot of water-soluble ink on the line 1.5 cm from the bottom (origin). The ink migrates with the solvent front and provides a visible indication of when it reaches the pencil line drawn 10 cm from the origin. In this system, with the Whatman no. 3 paper developed in a 1:1 mixture of anhydrous methanol and acetone, free pertechnetate migrates in close proximity to the solvent front while the labeled radiopharmaceutical and the Tc-HR remain at the origin. When the Gelman ITLC-SG strips are developed in 0.9% NaCl, the Tc-HR remains at the origin while the remaining activity migrates close to the solvent front. One vial of each brand of MDP was used for each experiment.

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**TABLE 1. Materials in the MDP Preparations**

| Product | MDP | Stannous chloride | Stannous fluoride | Ascorbic acid | pH |
|---|---|---|---|---|---|
| MPI MDP | 10 mg | 0.17 mg | 0 | 2 mg | 4–8 |
| Osteolite | 10 mg | 0.5 mg | 0 | 0 | 7–7.5 |
| MDP-Squibb | 20 mg | 0 | 0.33 mg | 1 mg | 6.5 |
| Amerscan MDP | 15.6 mg | 0 | 0.85 mg | 0 | ? |

**TABLE 2. Procedure for Determining Free Pertechnetate and Hydrolized Reduced Tc-99m in Labeled MDP Radiopharmaceuticals**

1. Cut 1 × 15 cm strips of Whatman no. 3 or Gelman ITLC-SG chromatography material and mark with a pencil at 1.5, 6.5, and 11.5 cm from one end.
2. Place 15 ml of a 1:1 mixture of acetone and methyl alcohol (anhydrous).
3. Place 15 ml of 0.9% NaCl in an 8-oz. jar with lid.
4. Place a small drop of radiopharmaceutical on the line 1.5 cm from the bottom.
5. Develop Whatman no. 3 strip in acetone-methanol mixture and Gelman ITLC-SG strip in 0.9% NaCl until solvent migrates to the line drawn 10 cm from the origin.
6. *Immediately* cut Whatman no. 3 and Gelman ITLC-SG strips into two sections at the line drawn 5 cm above the origin.
7. Count all sections for activity per unit time using scintillation well counter and subtract background. Determine levels of free pertechnetate and hydrolized reduced Tc-99m.

For each preparation of MDP, time period, and condition, five samples were chromatographically evaluated. The data were analyzed statistically to obtain the mean and standard deviation.

## Results

Table 3 shows the results of the radiochemical evaluation of the MDP preparations that were prepared under normal conditions and kept in the vials. Free pertechnetate levels of less than 0.5% were observed up to 6 hr after preparation. However, free pertechnetate levels of 1.48 and 6.87% were observed in the Osteolite and Amerscan kits at 24 hr while the MPI MDP and MDP-Squibb kits remained stable. The Tc-HR levels in all four preparations remained at less than 2% throughout. Results of radiochemical evaluation of these same four MDP preparations using syringes as storage containers were similar (Table 4); the Tc-HR levels in all four preparations were always less than 1.5%.

Table 5 shows the results of radiochemical evaluation of free Tc-99m and Tc-HR when the four MDP preparations were stressed by injecting 3 cc of room air into each vial 30 min after preparation. All four preparations exhibited levels of free pertechnetate of less than 0.6% up to 6 hr, but at 24 hr, two of the four products exhibited high levels of free pertechnetate and two had values not much different from those seen under normal conditions at 24 hr. The Tc-HR levels remained at less than 1.2% up to 24 hr after formulation throughout.

Table 6 summarizes the results of determining levels of free pertechnetate and Tc-HR in vials of four MDP preparations when they were injected with 400 mCi of [$^{99m}$Tc] pertechnetate from the first elution of a newly received generator. Table 7 summarizes the results obtained when a sample from the vial was stored in a syringe. The stress of adding high amounts of Tc-99m (400 mCi) showed that, again, all four products were stable when remaining in the vial up to 8 hr after preparation, but at 24 hr, three of the four preparations contained considerable amounts of free pertechnetate. The samples in the syringes were not as stable, showing signs of breakdown in two of the four products at 6 and 8 hr after preparation.

When the vials of MDP were prepared using 400 mCi of Tc-99m, levels of Tc-HR, while slightly higher for all products in both vials and syringes, were still at levels of less than 4% throughout.

## Discussion

Centralized nuclear pharmacies prepare radiopharmaceuticals with larger mCi amounts of Tc-99m, and dispense individual doses in syringes that may not be used for patient injection for several hours. We tried to evaluate kit performance under both normal and less-than-favorable conditions. An unfavorable condition was considered to be using high levels of radioactivity (400 mCi) from the first pertechnetate eluates obtained from a newly received Tc-99m generator. The first eluate contains high quantities of total technetium (Tc-99 and Tc-99m) and upon addition to stannous labeled radiopharmaceuticals, the usable stannous ion may become the limiting factor in the labeling reaction and may result in depressed

**TABLE 3. Radiochemical Evaluation of MDP Preparations Kept in Vial**

Free Tc-99m (mean percent ± SD of five samples)

| Time (hr) | Product: MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.005 ± .0034 | 0.12 ± .08 | 0.32 ± .31 | 0.11 ± .02 |
| 4 | 0.05 ± .01 | 0.21 ± .13 | 0.18 ± .06 | 0.06 ± .005 |
| 6 | 0.07 ± .01 | 0.23 ± .09 | 0.20 ± .10 | 0.08 ± .004 |
| 24 | 0.70 ± .05 | 1.48 ± .14 | 0.45 ± .27 | 6.87 ± .50 |

Hydrolized-reduced Tc-99m (mean percent Tc-HR ± SD of five samples)

| Time (hr) | Product: MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.28 ± .26 | 0.20 ± .20 | 0.43 ± .15 | 0.08 ± .03 |
| 4 | 1.22 ± 1.12 | 0.30 ± .19 | 0.23 ± .21 | 1.73 ± .10 |
| 6 | 0.26 ± .10 | 0.32 ± .13 | 0.43 ± .12 | 0.30 ± .08 |
| 24 | 0.28 ± .07 | 0.76 ± .14 | 0.80 ± .22 | 0.45 ± .12 |

**TABLE 4. Radiochemical Evaluation of MDP Preparations Kept in Syringes**

Free Tc-99m (mean percent pertechnetate ± SD of five samples)

| Time (hr) | Product MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.06 ± .04 | 0.50 ± .46 | 0.56 ± .27 | 0.82 ± .04 |
| 4 | 0.06 ± .005 | 0.60 ± .70 | 0.16 ± .18 | 0.42 ± .05 |
| 6 | 0.08 ± .006 | 0.34 ± .14 | 0.34 ± .25 | 0.98 ± .46 |
| 24 | 0.41 ± .25 | 1.46 ± 1.06 | 0.36 ± .20 | 5.74 ± .85 |

Hydrolized-reduced Tc-99m (mean percent Tc-HR ± SD of five samples)

| Time (hr) | Product MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.48 ± .40 | 1.24 ± .89 | 0.56 ± .59 | 0.84 ± .41 |
| 4 | 0.81 ± .47 | 0.69 ± .93 | 0.31 ± .12 | 0.55 ± .23 |
| 6 | 0.38 ± .09 | 0.22 ± .08 | 0.23 ± .15 | 0.94 ± .44 |
| 24 | 0.44 ± .06 | 1.18 ± .60 | 0.62 ± .48 | 0.66 ± .30 |

**TABLE 5. Radiochemical Evaluation of MDP Preparations When 3 cc of Air Is Injected Into Vial**

Free Tc-99m (mean percent pertechnetate ± SD of five samples)

| Time (hr) | Product MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.08 ± .05 | 0.08 ± .007 | 0.13 ± .02 | 0.11 ± .06 |
| 4 | 0.37 ± .42 | 0.55 ± .34 | 0.41 ± .35 | 0.04 ± .01 |
| 6 | 0.11 ± .003 | 0.55 ± .34 | 0.12 ± .02 | 0.14 ± .02 |
| 24 | 13.13 ± 1.8 | 1.97 ± .80 | 0.60 ± .14 | 9.04 ± .80 |

Tc-hydrolized reduced (mean percent Tc-HR ± SD of five samples)

| Time (hr) | Product MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.14 ± .11 | 0.22 ± .10 | 0.15 ± .09 | 0.27 ± .04 |
| 4 | 0.38 ± .12 | 0.46 ± .22 | 0.54 ± .56 | 0.44 ± .17 |
| 6 | 0.23 ± .02 | 0.47 ± .29 | 0.20 ± .28 | 0.43 ± .11 |
| 24 | 0.43 ± .16 | 1.12 ± .31 | 0.94 ± .43 | 0.55 ± .04 |

**TABLE 6. Radiochemical Evaluation of MDP Preparations Kept in Vials after Reconstituting Unit with 400 mCi of [$^{99m}$Tc] Pertechnetate**

Free Tc-99 (mean percent ± SD of five samples)

| Time (hr) | Product MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.10 ± .02 | 0.72 ± .007 | 0.03 ± .005 | 0.05 ± .002 |
| 4 | 0.11 ± .04 | 0.09 ± .02 | 0.04 ± .015 | 0.17 ± .04 |
| 6 | 0.44 ± .56 | 0.17 ± .02 | 0.04 ± .01 | 0.13 ± .02 |
| 8 | 0.14 ± .02 | 0.20 ± .15 | 0.06 ± .01 | 0.78 ± .26 |
| 24 | 11.53 ± .96 | 24.80 ± 1.80 | 0.18 ± .02 | 10.73 ± 1.35 |

Hydrolized-reduced Tc-99m (mean percent Tc-HR ± SD of five samples)

| Time (hr) | Product MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
|---|---|---|---|---|
| 1 | 0.59 ± .20 | 1.99 ± .43 | 0.90 ± .45 | 1.90 ± .28 |
| 4 | 0.45 ± .05 | 1.38 ± .46 | 0.50 ± .08 | 3.46 ± 1.00 |
| 6 | 0.49 ± .36 | 1.00 ± .48 | 0.31 ± .03 | 0.67 ± .32 |
| 8 | 1.12 ± 1.13 | 0.96 ± .21 | 0.46 ± .39 | 0.42 ± .11 |
| 24 | 0.33 ± .11 | 1.18 ± .09 | 0.45 ± .08 | 0.35 ± .05 |

or incomplete labeling (*1*).

Another unfavorable condition results when air is introduced into the vial during dose withdrawal. Previous studies have shown that the presence of oxidants and oxygen in radiopharmaceuticals containing low concentrations of stannous ion has an adverse effect on their stability (*2–4*). Until 1976 some manufacturers tried to overcome such problems by maintaining a nitrogen atmosphere or increasing the stannous ion concentration in kits; however, the increased levels of tin could adversely affect other nuclear medicine studies (*5*). In 1976, Tofe (*6*) reported the use of ascorbic acid as a stabilizing agent for bone imaging agents and some manufacturers have incorporated this into their product formulations. In our experiment two of the products contained ascorbic acid as a stabilizing agent and two did not (Table 1). Our findings indicate that the products with ascorbic acid are more stable. They are also able to withstand the stress of high levels of pertechnetate and injection of room air into the vials with very little effect on their stability.

When we began our study, we sometimes detected abnormally high levels of Tc-HR, but there was no consistent pattern to them. They would appear in one or two of the five samples for each product and at various times after preparation. After various experiments were run to evaluate this problem, we determined the cause: extended air drying times of the ITLC-SG strips after removal from the developing chamber and before cutting into sections. If the strips of ITLC-SG are placed flat on absorbent paper but not cut immediately after removal from the solvent, the radioactivity was found to migrate over the strips giving rise to abnormally high and false results for the Tc-HR levels. This redistribution of activity occurs because of the large volume of aqueous solvent retained by the ITLC-SG strips. It does not occur with the Whatman no. 3 paper developed in organic solvents such as acetone or methyl-ethyl-ketone because they are nonaqueous, evaporate rapidly, and have a low solvent capacity in the paper strips. The drying time between spotting the radioactivity and developing the strips was found to have an insignificant effect on the levels of Tc-HR. Once we had determined the exact cause of these abnormally high levels of Tc-HR, all the experiments were repeated and the chromatography strips were cut into two sections immediately upon removal from the developing chamber. The data included here were obtained using this procedure.

## Conclusions

All four MDP preparations exhibited excellent stability under both normal and stress conditions in vials and syringes up to 6 hr after preparation. However, the two MDP preparations containing ascorbate as an antioxidant appear better able to withstand the stress of high levels of radioactivity, storage in syringe, and accidental addition of air. These are very important considerations for a centralized nuclear pharmacy and are equally important for a nuclear medicine department in a large hospital that wishes to maximize the number of doses it can obtain from a single vial. This increased stability is also an important consideration for hospitals that rotate their technologists through the "hot lab" because individual kit preparation technique may vary slightly. Many technologists may find it useful to know that if an emergency scan is requested in the afternoon, a new kit will not have to be prepared.

**TABLE 7. Radiochemical Evaluation of MDP Preparations Kept in Syringes after Reconstituting with 400 mCi of Tc-99m**

Free Tc-99m (mean percent ± SD of five samples)

| Time (hr) | Product | | | |
|---|---|---|---|---|
| | MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
| 1 | 0.11 ± .01 | 0.11 ± .03 | 0.05 ± .03 | 0.06 ± .007 |
| 4 | 0.17 ± .02 | 0.42 ± .13 | 0.08 ± .007 | 0.26 ± .16 |
| 6 | 0.18 ± .03 | 0.12 ± .0008 | 0.06 ± .009 | 2.56 ± .15 |
| 8 | 0.18 ± .002 | 10.40 ± 2.29 | 0.06 ± .01 | 3.21 ± .25 |
| 24 | 5.86 ± 2.63 | 37.91 ± .76 | 0.40 ± .04 | 16.73 ± 1.0 |

Hydrolized-reduced Tc-99m (mean percent of Tc-HR ± SD of five samples)

| Time (hr) | Product | | | |
|---|---|---|---|---|
| | MPI MDP | Osteolite | MDP-Squibb | Amerscan MDP |
| 1 | 0.78 ± .16 | 1.79 ± .63 | 0.26 ± .04 | 1.94 ± .16 |
| 4 | 0.68 ± .25 | 1.79 ± .63 | 0.72 ± .62 | 0.89 ± .20 |
| 6 | 0.50 ± .12 | 0.60 ± .08 | 0.57 ± .43 | 0.76 ± .73 |
| 8 | 0.71 ± .19 | 0.71 ± .12 | 0.43 ± .12 | 0.83 ± .25 |
| 24 | 0.65 ± .18 | 0.63 ± .04 | 0.38 ± .09 | 0.68 ± .23 |


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