affect the rate of migration from the injection site. To maximize the rate of migration and the overall migration success rate of the tracer to the sentinel node in our clinical setting, we used kit preparations in which the ^{99m}Tc-sulfur colloid was heated for 3 min, allowed to cool for 2 min and used pertechnetate that had the highest amount of ingrowth of [^{99m}Tc]pertechnetate. Use of this preparation procedure in addition to withdrawing the patient dose through a sterile 5-μm filter and making 4–6 intradermal injections, 50–100 μl each, around the primary site, resulted in an excellent agent for lymphoscintigraphy studies for malignant melanoma. We have successfully imaged 97% (106/109) of our patients and have also used rapid dynamic scans to map and image the lymphatic drainage system (*17–20*). In addition to rapid movement, the preparation demonstrated prolonged retention within the nodes. At our institution, this preparation, in combination with imaging and a gamma hand-held detector, plays an important role in the detection, localization and excision of the sentinel node.

Currently, several studies are underway to identify additional methods that would further reduce the average particle size formed during the preparation of ^{99m}Tc-sulfur colloid. Particle size studies utilizing [^{99m}Tc]-sodium pertechnetate obtained from generators having up to 7 days of ingrowth of [^{99m}Tc]-pertechnetate are currently being investigated. Also, studies in which an additive is placed into the kit before heating, which provides more nucleation sites for particles to form, are underway.

ACKNOWLEDGMENT

We thank CIS-US for supplying the sulfur colloid kits.

REFERENCES

1. Krag DN, Weaver DL, Alex JC, Fairbank JT. Surgical resection and radiolocalization of the sentinel lymph node in breast cancer using a gamma probe. *Surg Oncol* 1993;2:335–340.
2. Alex JC and Krag DN. Gamma-probe guided localization of lymph nodes. *Surg Oncol* 1993;2:137–143.
3. Alex JC, Weaver DL, Fairbank JT, Rankin BS, Krag DN. Gamma-probe-guided lymph node localization in malignant melanoma. *Surg Oncol* 1993;2:303–308.
4. Morton DL, Wen D-R, Wong H, et al. Technical details of intraoperative lymphatic mapping for early stage melanoma. *Arch Surg* 1992;127:392–399.
5. Bergqvist L, Strand S-E, and Perssom BRR. Particle sizing and biokinetics of interstitial lymphoscintigraphic agents. *Semin Nucl Med* 1983;12:9–19.
6. Product package insert. Antimony sulfide colloid. Cadema Medical Products Inc, Middletown, NY.
7. Product package insert. Solco Nanocoll. Manufacturer Sorin Biomedi, Vercelli, Italy.
8. Hauser W, Atkins HL, Richards P. Lymph node scanning with ^{99m}Tc-sulfur colloid. *Radiology* 1969;92:1369–1371.
9. Dragotakes SC, Callahan RJ, LaPointe LC, et al. Particle size characterization of a filtered ^{99m}Tc-sulfur colloid preparation for lymphoscintigraphy. *J Nucl Med* 1995; 36(suppl):80P [Abstract].
10. Hung JC, Wiseman GA, Wahner HW, et al: Filtered technetium-99m-sulfur colloid evaluated for lymphoscintigraphy. *J Nucl Med* 1995;36:1895–1901.
11. Larson SM, Nelp WB. Radiopharmacology of a simplified technetium-99m colloid preparation for photoscanning. *J Nucl Med* 1966;7:817–826.
12. Kelly WN, Ice RD. Pharmaceutical quality of technetium-99m sulfur colloid. *Am J Hosp Pharm* 1973;30:817–820.
13. Krogsgaard OW. Technetium-99m-sulfur colloid: in vitro studies of various commercial kits. *Eur J Nucl Med* 1976;1:31–35.
14. Steigman J, Soloman NA, Hwang L-Y. Technetium-sulfur colloid. *Appl Radiat Isot* 1986;37:223–229.
15. Steigman J, Eckelman WC. Technetium (VII) compounds. In: *The chemistry of technetium in medicine*. Washington DC: National Academy Press; 1992:10–15.
16. Henze E, Schelbert HR, Collins JD, et al. Lymphoscintigraphy with ^{99m}Tc-labeled dextran. *J Nucl Med* 1982;23:923–929.
17. Taylor A, Murray D, Herda S, et al. The importance of dynamic lymphoscintigraphy in detecting the sentinel node. *J Nucl Med* 1995;36(suppl):223P [Abstract].
18. Vansant J, Mudun A, Murray D, et al. Lymphoscintigraphy defines a surgical approach to patients with clinical stage I melanoma. *J Nucl Med* 1995;36(suppl):223P [Abstract].
19. Mudun A, Murray D, Herda S, et al. Reproducibility of lymphoscintigraphy and intraoperative surgical probe to identify the sentinel node in patients with melanoma. *J Nucl Med* 1995;36:263P [Abstract].
20. Mudun A, Murray D, Herda S, et al. Lymphoscintigraphy and the sentinel node(s) in patients with melanoma: reproducibility and effectiveness of the intraoperative probe. *Radiology* 1996;199:171–175.

Technetium-99m-Tetrofosmin as a Substrate for P-Glycoprotein: In Vitro Studies in Multidrug-Resistant Breast Tumor Cells

James R. Ballinger, James Bannerman, Izzie Boxen, Patricia Firby, Neil G. Hartman and Malcolm J. Moore

Departments of Nuclear Medicine, Experimental Therapeutics and Medicine, Ontario Cancer Institute/Princess Margaret Hospital, Toronto, Ontario; Faculties of Pharmacy and Medicine, University of Toronto; and Department of Nuclear Medicine, Notre Dame Hospital, Montréal, Québec, Canada

The accumulation of ^{99m}Tc-tetrofosmin (TFos) was studied in wild-type (WT) and doxorubicin-resistant (Adr^R) variants of the rat MatB and human MCF-7 breast tumor cell lines to determine whether TFos, like ^{99m}Tc-sestamibi (MIBI), is a substrate for P-glycoprotein (P-gp), a multidrug-resistance transporter. **Methods:** The time course of accumulation of TFos and MIBI in WT and Adr^R cells over 1 hr was studied using single-cell suspensions at 1×10^6 cells/ml incubated at 37°C in the presence or absence of PSC833, a potent modulator of P-gp. Modulator dose-response curves were generated for PSC833, cyclosporin A, and verapamil. **Results:** In both MatB and MCF-7 cells, TFos and MIBI accumulated extensively in WT cells and accumulation was not affected by PSC833. In contrast, Adr^R cell lines accumulated very little of either tracer, but addition of PSC833 or other modulator increased this accumulation in a dose-dependent fashion. TFos and MIBI did not differ significantly in their behavior. **Conclusion:** TFos shares with MIBI the property of being a substrate for P-gp and thus TFos may be useful for functional imaging of tumor P-gp status.

Key Words: technetium-99m-tetrofosmin; technetium-99m-sestamibi; multidrug resistance

J Nucl Med 1996; 37:1578–1582

Multidrug resistance (MDR) involving overexpression of P-glycoprotein (P-gp), a transmembrane pump which acts as a natural defense mechanism by pumping xenobiotics out of

Received Jan. 12, 1996; revision accepted Apr. 4, 1996

For correspondence or reprints contact: J.R. Ballinger, PhD, Nuclear Medicine, Ontario Cancer Institute, 610 University Ave., Toronto, Ontario, Canada, M5G 2M9.

cells, is believed to be a factor contributing to the failure of some tumor's response to antineoplastic agents such as doxorubicin and paclitaxel (*1*). A variety of modulators, including cyclosporin A (CsA) and verapamil (Vrp), are capable of reversing resistance attributable to P-gp in model systems, although clinical efficacy has been difficult to demonstrate, possibly due to inadequate concentrations of the modulator reaching the tumor (*1,2*). Noninvasive assessment of the P-gp status of tumors and monitoring the effect of potent second-generation modulating agents such as PSC833 and GG918 with a nuclear medicine technique could lead to improved response in the treatment of resistant tumors (*3*).

Thallium-201 and ^{99m}Tc-sestamibi are myocardial imaging agents which are also useful for the detection of a variety of tumors (*4,5*). Both tracers accumulate in viable tumor cells; however, ^{99m}Tc-sestamibi is a substrate for P-gp and thus may provide additional information about the P-gp status of tumor cells. This property of ^{99m}Tc-sestamibi was discovered by Piwnica-Worms et al. using cultured tumor cells in vitro (*6*), validated in an animal model (*6*) and confirmed by gene transfection experiments (*7*). We have observed a three-fold difference in the washout rate of ^{99m}Tc-sestamibi from sensitive and doxorubicin-resistant breast tumors in rats (*8*), and have confirmed that ^{201}Tl is not a substrate for P-gp (*9*). Preliminary reports on the clinical use of functional imaging of tumor P-gp status with ^{99m}Tc-sestamibi are beginning to appear (*10–15*). Notably, in a series of patients with breast tumors, Ciarmiello et al. (*12*) determined ^{99m}Tc-sestamibi washout rates by quantification of serial images over 4 hr and P-gp expression was measured independently in biopsy specimens from the same patients. There was a statistically significant correlation between ^{99m}Tc-sestamibi washout rates and P-gp expression, and a three-fold difference in washout rate between tumors with baseline levels of P-gp and those with elevated levels.

Technetium-99m-tetrofosmin is a lipophilic phosphine cation like ^{99m}Tc-sestamibi, that is developed for myocardial perfusion imaging (*16*) and has recently been shown to accumulate in some tumors (*17–20*). Gros et al. (*21*) have suggested that lipophilic cations are a class of compounds which are substrates for P-gp. We therefore studied ^{99m}Tc-tetrofosmin in sensitive and multidrug-resistant rodent and human breast tumor cell lines in vitro to determine whether ^{99m}Tc-tetrofosmin is a substrate for P-gp and may potentially be useful for functional imaging of the P-gp status of tumors.

MATERIALS AND METHODS

The wild-type (WT) rat breast adenocarcinoma cell line MatB 13762, its doxorubicin-resistant variant MatB/Adr^R, and a doxorubicin-resistant variant of the human breast carcinoma cell line MCF-7 (MCF-7/Adr^R) were obtained from Dr. G. Batist, McGill University, Montréal, Canada (*22,23*). The wild-type MCF-7 cell line was obtained from the American Type Tissue Culture Collection (Rockville, MD). The MatB/Adr^R and MCF-7/Adr^R cell lines both showed 200-fold resistance to doxorubicin relative to the respective parental cell lines and their overexpression of P-gp has been characterized (*22,23*). Cells grown in monolayer in alpha minimum-essential medium that was supplemented with 10% fetal bovine serum were harvested with trypsin and resuspended at a concentration of 1×10^6 cells/ml in fresh medium. Technetium-99m-tetrofosmin and ^{99m}Tc-sestamibi were prepared from kits and diluted to 10 MBq/ml with saline. The radiochemical purity of the two agents was determined by TLC and on every occasion was > 90%. Accumulation experiments were performed as described previously (*8,9,24*) using stirred single-cell suspensions incubated at 37°C under room air. Aliquots of 100 μl (1 MBq) of the diluted tracer were added to vials containing 5-ml aliquots of cell suspension. At 1, 15, 30, 45 and 60 min after addition of the tracer, duplicate samples of 400 μl were removed from each vial and transferred to 1.5-ml microcentrifuge tubes containing 600 μl ice-cold saline and then centrifuged at 14,000 g for 2 min. The supernatant was aspirated and the cell pellet was carefully washed with 500 μl ice-cold saline. The tip of the tube containing the pellet was clipped off, placed in a counting tube and assayed for radioactivity in a gamma well counter. In experiments with CsA, Vrp, or the nonimmunosuppressive cyclosporin analog PSC833, the modulator was prepared by fresh serial dilution of a stock solution with saline and added to the cell suspension 5 min before the tracer. For experiments with different concentrations of modulator (dose-response curves), a single incubation time of 10 min was used; the ranges of modulator concentration evaluated were 0.1–10 μ*M* for PSC833 and 1–100 μ*M* for CsA and Vrp.

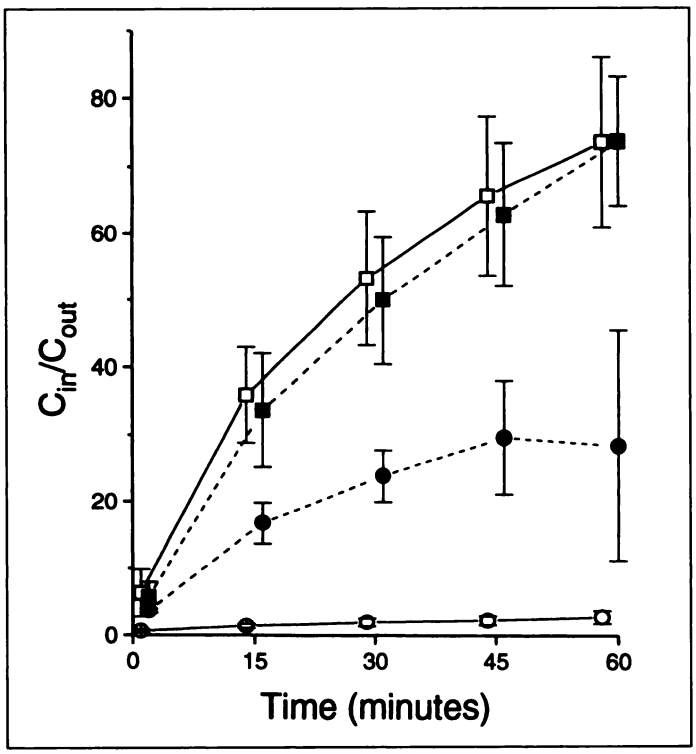

FIGURE 1. Time-course of accumulation of ^{99m}Tc-tetrofosmin in MatB/WT and MatB/Adr^R cells in the presence or absence of 1 μ*M* PSC833. Each point is the mean ± s.d. of 3–5 separate experiments. MatB/WT, □; MatB/WT + PSC833, ■; MatB/Adr^R, ○; MatB/Adr^R + PSC833, ●.

From the measurements of radioactivity in the cell pellets and a standard representing the supernatant concentration, together with an independent measurement of cell volume, the accumulation ratio could be calculated as the ratio of radioactivity concentration inside the cell to that outside the cell (C_{in}/C_{out}), as described previously (*8,24*). The trapping of supernatant liquid in the cell pellet was determined to contribute < 0.2 C_{in}/C_{out} unit. The C_{in}/C_{out} ratio can be converted to the more physiologically relevant unit fmol per mg protein per nM_0 by dividing by the cellular protein concentration. From the modulator dose-response curves, EC_{50} values were determined by interpolation on semilog plots.

RESULTS

Within 1 min of the addition of ^{99m}Tc-tetrofosmin to rat MatB/WT cells, the tracer had partitioned into the cells to the extent that the ratio of activity concentration inside to that outside the cells (C_{in}/C_{out}) was 5.2 ± 2.8 (Fig. 1). Accumulation continued over a 60-min period to reach a C_{in}/C_{out} value of 70.9 ± 15.0, although the rate of accumulation began to

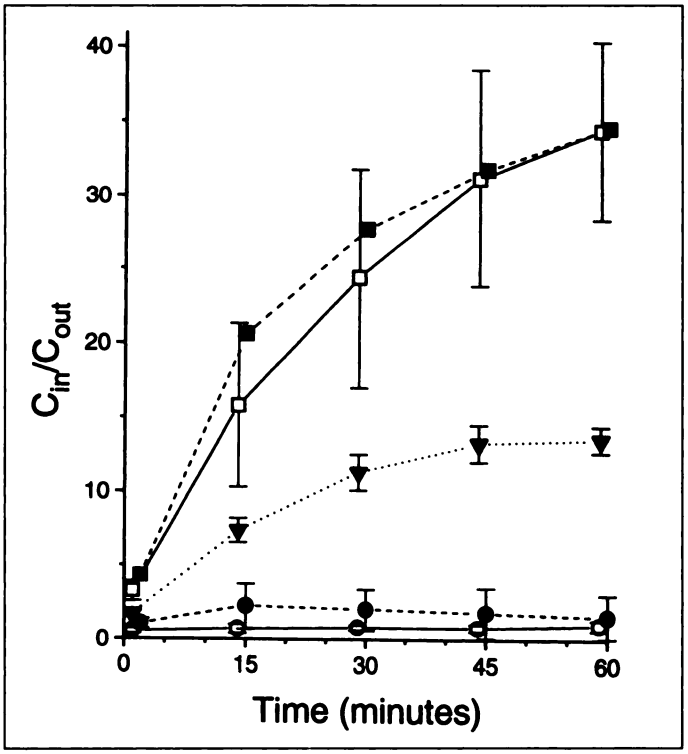

FIGURE 2. Time-course of accumulation of ^{99m}Tc-tetrofosmin in MCF-7/WT and MCF-7/Adr^R cells in the presence or absence of 1 μM or 10 μM PSC833. Each point is the mean ± s.d. of 3 separate experiments (except where noted). MCF-7/WT, □; MCF-7/WT + 1 μM PSC833, ■, (n = 1); MCF-7/Adr^R, ○; MCF-7/Adr^R + 1 μM PSC833, ●; MCF-7/Adr^R + 10 μM PSC833, ▼.

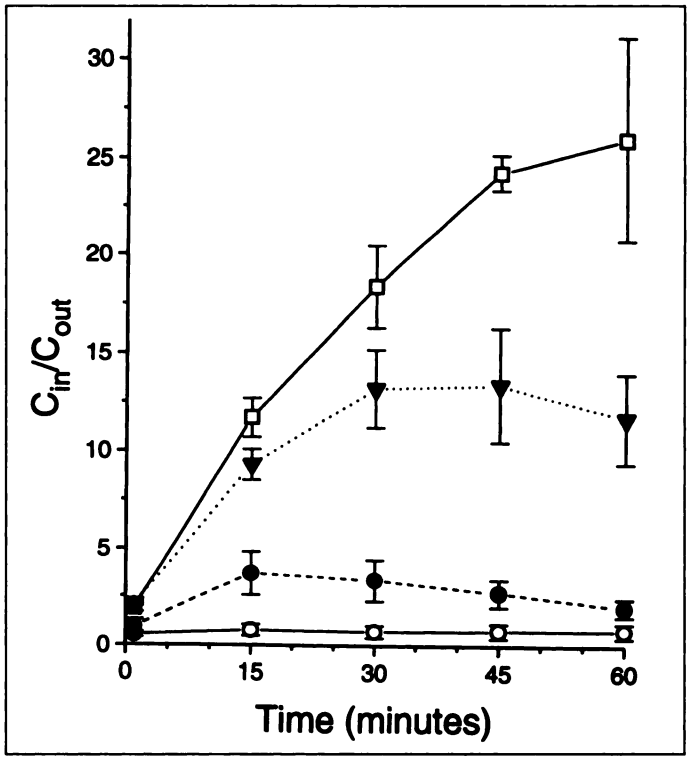

FIGURE 3. Time-course of accumulation of ^{99m}Tc-sestamibi in MCF-7/WT and MCF-7/Adr^R cells in the presence or absence of 1 μM or 10 μM PSC833. Each point is the mean ± s.d. of 3 separate experiments. MCF-7/WT, □; MCF-7/Adr^R, ○; MCF-7/Adr^R + 1 μM PSC833, ●; MCF-7/Adr^R + 10 μM PSC833, ▼.

decrease. Conversely, when ^{99m}Tc-tetrofosmin was added to MatB/Adr^R cells, the initial C_{in}/C_{out} was 0.58 ± 0.14 and accumulation increased only modestly over 60 min to a value of 2.77 ± 1.02, by which time there was a 26-fold difference in tracer accumulation between MatB/WT and MatB/Adr^R cells. The addition of 1 μM PSC did not significantly affect the accumulation of ^{99m}Tc-tetrofosmin in MatB-WT cells, whereas it did cause an increase in accumulation in MatB-Adr^R cells to a value of 11-fold over control MatB/Adr^R cells by 60 min, thereby reducing the difference between MatB/WT and MatB-Adr^R cells from 26-fold to 2.5-fold (Fig. 1). These results are similar to those we reported previously for ^{99m}Tc-sestamibi in the same pair of cell lines (*8*).

The pattern observed with ^{99m}Tc-tetrofosmin in human MCF-7/WT and MCF-7/Adr^R cells paralleled that in MatB/WT and MatB/Adr^R (Fig. 2). Accumulation of ^{99m}Tc-tetrofosmin in MCF-7/WT cells was high and not affected by the addition of 1 μM PSC833. Although accumulation of ^{99m}Tc-tetrofosmin in MCF-7/Adr^R cells was as low and flat as that in MatB/Adr^R cells (WT/Adr^R differential at 60 min was 35-fold), addition of 1 μM PSC833 produced only a modest and short-lived increase in accumulation tracer by MCF-7/Adr^R cells. However, 10 μM PSC833 produced a more extensive and prolonged increase in accumulation of tracer (WT/Adr^R differential at 60 min was 2.5-fold).

Technetium-99m-sestamibi was also studied in MCF-7/WT and MCF-7/Adr^R cells, and the results presented in Fig. 3 were both qualitatively and quantitatively similar to those obtained with ^{99m}Tc-tetrofosmin in the same cell lines (Fig. 2). As seen with ^{99m}Tc-tetrofosmin, 1 μM PSC833 produced a transient response while 10 μM produced a substantial increase in accumulation of ^{99m}Tc-sestamibi in MCF-7/Adr^R cells. The WT/Adr^R differentials for ^{99m}Tc-sestamibi accumulation at 60 min were 33-fold without the modulator and 2.2-fold in the presence of 10 μM PSC833.

Dose-response curves for three modulators (PSC833, CsA and Vrp) were generated for both tracers in both resistant cell lines. EC_{50} values for 10-min incubations at 1 × 10^6 cells/ml are reported in Table 1. The rank order of potency of modulators was PSC833 > CsA > Vrp for both tracers and both cell lines. With MatB/Adr^R cells, the differences in potency were approximately one order of magnitude between each modulator and the potencies were similar for both tracers. However, with MCF-7/Adr^R cells, the EC_{50} values for all three modulators were 2–4 times higher for ^{99m}Tc-tetrofosmin than for ^{99m}Tc-sestamibi, and the differences between modulators were not as pronounced.

DISCUSSION

Technetium-99m-tetrofosmin is a recently introduced agent and very limited information is available about its properties for

TABLE 1
Comparison of Potencies of P-gp Modulators for Accumulation of Technetium-99m-Tetrofosmin and Technetium-99m-Sestamibi in MatB/Adr^R and MCF-7/Adr^R Cells

		EC_{50} (μM)		
Cell line	Tracer	PSC833	Cyclosporin A	Verapamil
MatB/Adr^R	Tetrofosmin	1.2 ± 0.4	3.4 ± 0.7	25.0 ± 1.3
	Sestamibi	0.4 ± 0.1	4.5 ± 0.1	24.0 ± 3.6
MCF-7/Adr^R	Tetrofosmin	12.8 ± 1.8	14.3 ± 3.8	> 50
	Sestamibi	3.0 ± 1.1	7.5 ± 2.7	25.3 ± 0.2

Each value is mean ± s.d. for 3–4 replicates performed at 1 × 10^6 cells/ml.

tumor imaging (*17–20*). In vitro studies, particularly with respect to MDR, are particularly important to predict its potential clinical utility in this area.

The patterns obtained with ^{99m}Tc-tetrofosmin in rat MatB/WT and MatB-Adr^R cells in terms of time course of accumulation, WT versus Adr^R differences and responsiveness to modulation by 1 μM PSC833 (Fig. 1) were virtually identical to those previously reported with ^{99m}Tc-sestamibi in the same pair of cell lines (*8*). The only difference was in the absolute values for accumulation of the two tracers, with ^{99m}Tc-sestamibi values being ~3-fold higher than ^{99m}Tc-tetrofosmin values. This could reflect differences in responsiveness of the two tracers to membrane potentials (*6*).

The two tracers also behaved similarly in human MCF-7/WT and MCF-7/Adr^R cells (Figs. 2, 3). With this pair of cell lines, the absolute values for the two tracers were in the same range. However, MCF-7/Adr^R cells differed from MatB/Adr^R cells in that much higher concentrations of modulator were required to enhance accumulation of the tracers (i.e., 10 μM PSC833 produced a similar effect with MCF-7/Adr^R in Figs. 2 and 3 as 1 μM did with MatB/Adr^R in Fig. 1 and reference *8*). This difference is reflected in the EC_{50} values for PSC833 reported in Table 1. The data in Figures 2 and 3 and in Table 1 also suggest that ^{99m}Tc-tetrofosmin required higher concentrations of modulator than did ^{99m}Tc-sestamibi to enhance accumulation in MCF-7/Adr^R. Although both MatB/Adr^R and MCF-7/Adr^R were selected by growth in the presence of increasing concentrations of doxorubicin (adriamycin), and both are reported to show 200-fold resistance to doxorubicin compared to their parental WT lines (*22,23*), the extent to which P-gp is responsible for resistance may differ between MatB/Adr^R and MCF-7/Adr^R. Hendrikse et al. (*25*) reported that ^{99m}Tc-sestamibi is a substrate for the multidrug resistance-associated protein (MRP), an alternative transporter discovered by Cole et al. (*26*). It could be that ^{99m}Tc-tetrofosmin is also a substrate for MRP but has a different affinity than ^{99m}Tc-sestamibi.

The in vitro observation that ^{99m}Tc-sestamibi is a substrate for P-gp (*6*) has recently been extended into the clinic, with reports of correlation between ^{99m}Tc-sestamibi efflux rates and P-gp expression in breast tumors (*12*), and between ^{99m}Tc-sestamibi avidity and prognosis in lymphoma (*10*). The results of our study suggest that ^{99m}Tc-tetrofosmin shares with ^{99m}Tc-sestamibi the property of being a substrate for P-gp; thus, it is potentially useful for functional imaging of P-gp. This property of ^{99m}Tc-tetrofosmin is important for several reasons. First, ^{99m}Tc-tetrofosmin has some advantages over ^{99m}Tc-sestamibi as a radiopharmaceutical. Labeling of ^{99m}Tc-tetrofosmin is performed at room temperature, which is more convenient than the heating step required with ^{99m}Tc-sestamibi. Technetium-99m-tetrofosmin shows greater renal clearance and less hepatobiliary excretion than ^{99m}Tc-sestamibi, which offers advantages in imaging abdominal tumors. Second, since the primary use of these agents is for myocardial perfusion imaging and a clinic might switch from one agent to the other, it is important to know that both can be used for functional tumor imaging. Third, it can be useful to have a variety of agents available and each may turn out to have a different niche. Another new ^{99m}Tc-labeled myocardial imaging agent, ^{99m}Tc-furifosmin, also appears to be a substrate for P-gp, although not to the same extent as the other two (*27* and unpublished work from this laboratory). On the other hand, preliminary evidence suggests that ^{99m}Tc-teboroxime, a neutral lipophilic myocardial agent, is not a substrate for P-gp (Ballinger and Hartman, unpublished work); this is consistent with the work of Gros et al. (*21*).

CONCLUSION

The results of the present studies suggest that ^{99m}Tc-tetrofosmin shares with ^{99m}Tc-sestamibi the property of being a substrate for P-gp and thus is potentially useful for functional imaging of P-gp. These results form the groundwork for further investigation of ^{99m}Tc-tetrofosmin as a tumor agent in vitro and in the clinic.

ACKNOWLEDGMENTS

We thank Dr. Gerald Batist, McGill University, Montréal, for supplying resistant cell lines and Dr. A.M. Rauth, Ontario Cancer Institute, for assistance in establishing cell cultures. We thank Amersham Intl. plc, for donating the tetrofosmin, DuPont Pharma for donating the sestamibi and Sandoz Canada for donating cyclosporin A (Sandimmune) and PSC833. Preliminary reports of portions of this work were presented at the 15th annual meeting of the Eastern Great Lakes Chapter of the Society of Nuclear Medicine, Buffalo, NY, April 7, 1995, and at the 42nd annual meeting of the Society of Nuclear Medicine, Minneapolis, MN, June 12–15, 1995.

REFERENCES

1. Bradley G, Juranka PF, Ling V. Mechanism of multidrug resistance. *Biochim Biophys Acta* 1988;948:87–128.
2. Erlichman C, Moore MJ, Thiessen JJ, et al. Phase I pharmacokinetic study of cyclosporin A modulation of doxorubicin. *Cancer Res* 1993;53:4837–4842.
3. Moretti J-L, Caglar M, Duran-Cordobes M, Morere J-F. Can nuclear medicine predict response to chemotherapy? *Eur J Nucl Med* 1995;22:97–100.
4. Waxman AD. Thallium-201 in nuclear oncology. In: Freeman LM, ed. *Nuclear medicine annual 1991*. New York: Raven; 1991:193–209.
5. Piwnica-Worms D, Holman BL. Noncardiac applications of hexakis(alkylisonitrile) technetium-99m complexes. *J Nucl Med* 1990;31:1166–1167.
6. Piwnica-Worms D, Chiu ML, Budding M, Kronauge JF, Kramer RA, Croop JM. Functional imaging of multidrug-resistant P-glycoprotein with an organotechnetium complex. *Cancer Res* 1993;53:977–984.
7. Rao VV, Chiu ML, Kronauge JF, Piwnica-Worms D. Expression of recombinant human multidrug resistance P-glycoprotein in insect cells confers decreased accumulation of technetium-99m-sestamibi. *J Nucl Med* 1994;35:510–515.
8. Ballinger JR, Hua HA, Berry BW, Firby P, Boxen I. Technetium-99m-sestamibi as an agent for imaging P-glycoprotein-mediated multi-drug resistance: in vitro and in vivo studies in a rat breast tumor cell line and its doxorubicin-resistant variant. *Nucl Med Commun* 1995;16:253–257.
9. Ballinger JR, Sheldon KM, Boxen I, Erlichman C, Ling V. Differences between accumulation of ^{99m}Tc-sestamibi and ^{201}Tl-thallous chloride in tumor cells: role of P-glycoprotein. *J Nucl Biol Med* 1995:in press.
10. Dimitrakopoulou-Strauss A, Strauss LG, Goldschmidt H, Lorenz WJ, Maier-Borst W, van Kaick G. Evaluation of tumor metabolism and multidrug resistance in patients with treated malignant lymphomas. *Eur J Nucl Med* 1995;22:434–442.
11. Moretti JL, Caglar M, Boaziz C, Caillat-Vigneron N, Morere JF. Sequential functional imaging with technetium-99m hexakis-2-methoxyisobutylisonitrile and indium-111 octreotide: can we predict the response to chemotherapy in small cell lung cancer? *Eur J Nucl Med* 1995;22:177–180.
12. Ciarmiello A, Del Vecchio S, Potena MI, et al. Technetium-99m-sestamibi efflux and P-glycoprotein expression in human breast carcinoma [Abstract]. *J Nucl Med* 1995; 36(suppl):129P.
13. Bender H, Friedrich E, Zamora PO, Biersack H-J. Detection of multidrug resistance with ^{99m}Tc-sestamibi [Abstract]. *J Nucl Med* 1995;36(suppl):129P.
14. Varrella P, Lastoria S, Vergara E, Acampa W, Maurea S, Salvatore M. Technetium-99m-MIBI scintimammography for monitoring tumor response in patients with advanced breast cancer [Abstract]. *J Nucl Med* 1995;36(suppl):193P.
15. Dimitrakopoulou-Strauss A, Strauss LG, Goldschmidt H, Lorenz WJ, van Kaick G. Studies with ^{18}F-deoxyglucose (FDG) and ^{99m}Tc-sestamibi in patients with recurrent malignant lymphomas [Abstract]. *J Nucl Med* 1995;36(suppl):216P.
16. Kelly JD, Forster AM, Higley B, et al. Technetium-99m-tetrofosmin as a new radiopharmaceutical for myocardial perfusion imaging. *J Nucl Med* 1993;34:222–227.
17. Soricelli A, Cuocolo A, Tedeschi E, et al. Evaluation of brain tumors with ^{99m}Tc-tetrofosmin and ^{201}Tl: a preliminary report [Abstract]. *Eur J Nucl Med* 1994;21:S16.
18. Mansi L, Rambaldi PF, La Provitera A, Di Gregorio F, Procaccini E. Technetium-99m-tetrofosmin uptake in breast tumors [Abstract]. *J Nucl Med* 1995;36:83P.
19. Basoglu T, Sahin M, Coskun C, Koparan A, Bernay I, Erkan L. Technetium-99m-tetrofosmin uptake in malignant lung tumors. *Eur J Nucl Med* 1995;22:687–689.
20. Klain M, Maurea S, Lastoria S, Cuocolo A, Colao A, Salvatore M. Technetium-99m-tetrofosmin imaging of differentiated mixed thyroid cancer. *J Nucl Med* 1995;36: 2248–2251.
21. Gros P, Talbot F, Tang-Wai D, Bibi E, Kaback HR. Lipophilic cations: a group of model substrates for the multidrug-resistance transporter. *Biochemistry* 1992;31:1992–1998.

22. Schecter RL, Woo A, Duong M, Batist G. In vivo and in vitro mechanisms of drug resistance in rat mammary carcinoma model. *Cancer Res* 1991;51:1434–1442.
23. Batist G, Tulpule A, Sinha BK, Katki AG, Myers CE, Cowan KH. Overexpression of a novel anionic glutathione transferase in multidrug-resistant human breast cancer cells. *J Biol Chem* 1986;261:15544–15549.
24. Ballinger JR, Cowan DSM, Boxen I, Zhang ZM, Rauth AM. Effect of hypoxia on the accumulation of technetium-99m-glucarate and technetium-99m-gluconate by Chinese hamster ovary cells in vitro. *J Nucl Med* 1993;34:242–245.
25. Hendrikse NH, Franssen EJF, van der Graaf WTA, Meijer C, de Vries EGE. Reduced ^{99m}Tc-sestamibi accumulation in P-gp positive and MRP positive cell lines [Abstract]. *Proc Am Assoc Cancer Res* 1995;36:358.
26. Cole SPC, Bhardwaj G, Gerlach JH, et al. Overexpression of a transporter gene in a multidrug-resistant human cancer cell line. *Science* 1992;258:1650–1654.
27. Crankshaw CL, Marmion M, Burleigh BD, Deutsch E, Piwnica-Worms D. Non-reducible mixed ligand Tc(III) cations (Q complexes) are recognized as transport substrates by the human multidrug resistance (MDR) P-glycoprotein [Abstract]. *J Nucl Med* 1995;36(suppl):130P.

(continued from page 1556)

FIRST IMPRESSIONS

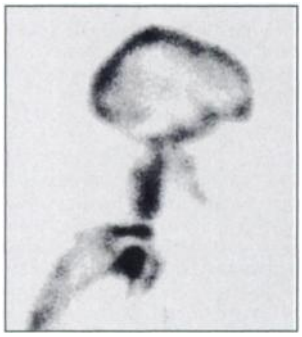

Figure 1.

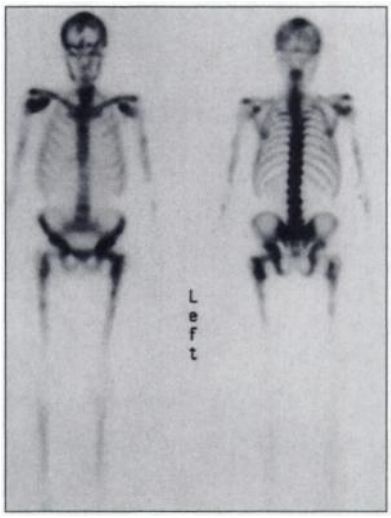

Figure 2.

Figure 3.

PURPOSE
Figure 1 is a right lateral view of the skull from a bone scan in an 82-yr-old man with metastatic prostate carcinoma. Anterior and posterior total-body images, however, are characteristic of extensive skeletal metastases (Fig.2). There is increased activity in the axial skeleton, proximal extremities and cranium. The lateral view demonstrates virtually absent activity in the facial bones. No activity is seen in the kidneys or bladder. This patient has a "superscan" with osseous metastases from prostate carcinoma which spare the distal extremities and facial bones and "disappear" on photographic display. With the intensity increased, activity in the facial bones can be detected (Fig. 3).

TRACER
Technetium-99m-MDP, 925 MBq

ROUTE OF ADMINISTRATION
Intravenous

TIME AFTER INJECTION
Three hours

INSTRUMENTATION
Prism (Picker) 2000 gamma camera

CONTRIBUTORS
Alan Siegel, Jill Hagerty, Dartmouth-Hitchcock Medical Center, Lebanon, NH