

Evaluation of the Stability of ^{99m}Tc -ECD and Stabilized ^{99m}Tc -HMPAO Stored in Syringes

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Objective: To determine and compare the stability of ^{99m}Tc -ECD and stabilized ^{99m}Tc -HMPAO when stored in syringes over an 8-h period.

Methods: ^{99m}Tc -ECD and stabilized ^{99m}Tc -HMPAO were prepared according to the manufacturers' protocols, with the following exception: eluate less than 60 min old was used to prepare ^{99m}Tc -HMPAO rather than the recommended 30 min. Once prepared, 185 MBq (5 mCi) of both products were drawn into 5-mL syringes and allowed to sit at room temperature. At 2, 4, 6, and 8 h after preparation, the radiochemical purity (RCP) of the contents of the syringes was determined and compared to the RCP of the products in vials. Retention of activity of each product in syringes was also evaluated by measuring activity remaining in each syringe (and filter, in the case of ^{99m}Tc -HMPAO) after expressing its contents.

Results: The RCP of stabilized ^{99m}Tc -HMPAO stored in syringes decreased from a mean of 87.7% at 2 h to 74.0% at 8 h after preparation. In contrast, ^{99m}Tc -ECD retained an RCP of greater than 94% throughout the time tested. The impurity that appeared to increase over time with ^{99m}Tc -HMPAO was found to be sodium pertechnetate. Total retention of activity remaining in the syringe and filter ranged from 11.6% at 2 h to 9.5% at 8 h for ^{99m}Tc -HMPAO; the syringe itself retained less than 5% of the total activity at all time periods. ^{99m}Tc -ECD exhibited 6.2% to 11.3% retention of activity in the syringe. The sorption of sodium pertechnetate to the syringe for the same time period was less than 1%.

Conclusions: ^{99m}Tc -ECD is a more stable product than stabilized ^{99m}Tc -HMPAO in a syringe. Both products demonstrate retention of radioactivity in the syringe. Some of this retention may denote sorption of the products to plastic.

Key Words: stabilized ^{99m}Tc -HMPAO; ^{99m}Tc -ECD; stability in syringe; retention in syringe

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SPPECT has become an important tool in the study of epileptogenic foci in those patients with refractory focal epilepsy (1-3). The radiopharmaceuticals most commonly used in North America for this purpose are technetium (^{99m}Tc)-labeled bicisate, or ethylcysteinate dimer (ECD)

(Neurolite; DuPont Pharmaceuticals Company, Billerica, MA), and stabilized ^{99m}Tc -exametazime, or hexamethylpropyleneamine oxime (HMPAO) (Ceretek; Amersham Healthcare, Arlington Heights, IL). For a patient to be injected during the ictal phase, the radiolabeled product should be prepared in advance and stored in a shielded syringe next to the patient's bedside.

From a pharmaceutical viewpoint, the stability of an injectable product should be evaluated if the product is transferred to and stored in another container for extended periods of time. In addition, the product should be tested for compatibility with its container before use (4,5). There is always a possibility that lipophilic products such as cerebral perfusion agents could interact with the syringe materials over time. Therefore, the intent of this study was to determine the radiochemical stability and retention of ^{99m}Tc -ECD and stabilized ^{99m}Tc -HMPAO in syringes over an 8-h period.

MATERIALS AND METHODS

^{99m}Tc -ECD was prepared as follows. To vial B (buffer/reaction vial), 3.7 GBq (100 mCi) sodium pertechnetate in 2 mL sterile saline were added. Only eluate from a generator eluted in the previous 24 h was used. Vial A was reconstituted with 3 mL sterile saline. The contents were gently agitated, and within 30 s, 1 mL of vial A was transferred to vial B. The mixture was allowed to sit at room temperature for 30 min. The contents were examined for particulates. Radiochemical purity (RCP) of the solution was determined, according to the manufacturer's package insert (6), by thin-layer chromatography using Baker-Flex silica gel IB-F plates (JT Baker Chemical Co., Phillipsburg, NJ) and ethyl acetate.

Stabilized ^{99m}Tc -HMPAO was prepared and tested in accordance with the manufacturer's insert (7), with the exception that pertechnetate was used within 1 h of elution from a generator that was eluted in the previous 24 h. Methylene blue, 0.5 mL, was added to 4.5 mL buffer solution (stabilizing solution). The reaction vial was reconstituted to 5 mL with 1.85 GBq (50 mCi) sodium pertechnetate in saline to which was added 2 mL of the stabilizing solution. The product was then allowed to incubate for 30 min at room temperature. The presence of free pertechnetate and hydrolyzed technetium was determined by using saline

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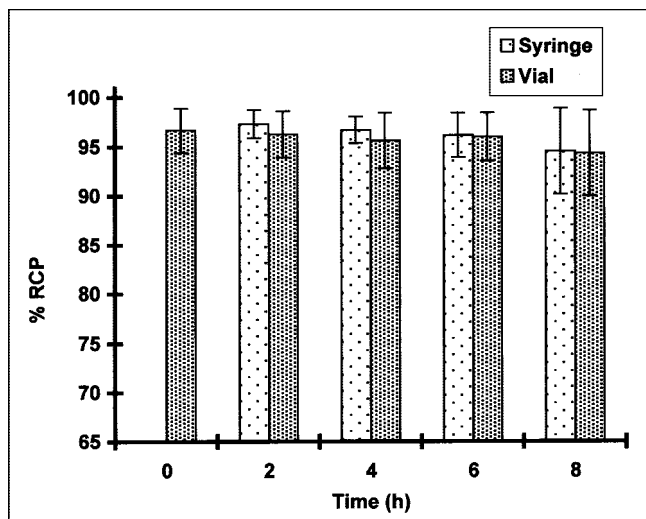


FIGURE 1. RCP of ^{99m}Tc-ECD in syringe and vial over time (mean \pm SD; $n = 10$). No significant difference was observed between RCP of samples at any time period regardless of container used.

and methyl ethyl ketone, and silica gel-impregnated glass fiber (ITLC-SG) paper. The percentage of secondary HMPAO was calculated using acetonitrile-to-water ratio of 50:50 and Whatman #1 paper (Whatman Biosystems, Kent, U.K.), as described in the manufacturer's insert.

Four latex-free syringes (Becton-Dickinson, Franklin Lakes, NJ) were prepared for each product immediately after quality control was completed, and each 5-mL syringe contained 185 MBq (5 mCi) activity. The syringes were stored at room temperature. RCP of the products was determined at 2, 4, 6, and 8 h after drawing up the doses into the syringes. The purity of the products in the syringes was compared with the RCP of the products in the vials for the same time period.

Retention of the products in the syringes was determined as follows. At the designated times, the full syringes were assayed in a dose calibrator, after which the syringe contents were expressed into a vial. The empty syringes were re-assayed in the dose calibrator and the remaining activity compared with that of the full syringes. The needles (and 0.45- μ m filter, for stabilized ^{99m}Tc-HMPAO) were then removed and the syringes re-assayed. The activity in the vial, empty syringe, needle, and filter (where applicable) was totaled and compared with the activity in the syringe before removal of the radiolabeled product, to ensure that geometry was not an influencing factor. For ^{99m}Tc-HMPAO, the radioactivity remaining in the filter was assayed separately. Both the RCP and the retention experiments were repeated 10 times for each product.

Controls were prepared by drawing up 185 MBq (5 mCi) of sodium pertechnetate into 5-mL syringes. The contents of the syringes were expressed at the selected times, and each syringe and filter was measured in the dose calibrator to assess the amount of radioactivity remaining.

The Student's *t* test was used to compare results between

the means of all groups. A value of $P < 0.05$ was considered statistically significant.

RESULTS

As shown in Figure 1, the RCP of ^{99m}Tc-ECD in the syringe and vial remained greater than 94% at all time periods tested. No statistically significant difference in the RCP was observed during the study, regardless of whether the product was stored in a syringe or vial. In contrast, the mean RCP of ^{99m}Tc-HMPAO decreased in both the vial and syringe with time (Fig. 2). The initial purity of ^{99m}Tc-HMPAO in the vial was observed to be 91.5% at the time of preparation, decreasing to 81.4% at 8 h after preparation. The product deteriorated more rapidly in the syringe, resulting in a mean RCP of 74.0% at the end of the study.

The predominant impurity corresponding with the decrease in RCP of ^{99m}Tc-HMPAO was found to be ^{99m}Tc-sodium pertechnetate (Fig. 3). ^{99m}Tc-sodium pertechnetate accounted for 3% of the activity in the vial, versus 4.5% in the syringe 2 h after preparation. After 4 h this difference became statistically significant, with 5.4% free pertechnetate in the vial compared with 7.6% in the syringe ($P < 0.05$). At 8 h after preparation, the percentage of pertechnetate in the syringe was almost twice that found in the vial (15.9% in the syringe vs. 9% in the vial; $P < 0.001$).

The radiochemical impurities of reduced, hydrolyzed technetium and secondary HMPAO did not change throughout the 8 h. ^{99m}TcO₂ represented 5.1% to 6% of the activity for both containers, whereas the percentage of secondary HMPAO ranged from 2.4% to 4.5%. These results were not statistically significant.

The retention characteristics of the products in syringes are provided in Table 1. The mean percentage of radioactivity remaining in the syringe/filter apparatus containing

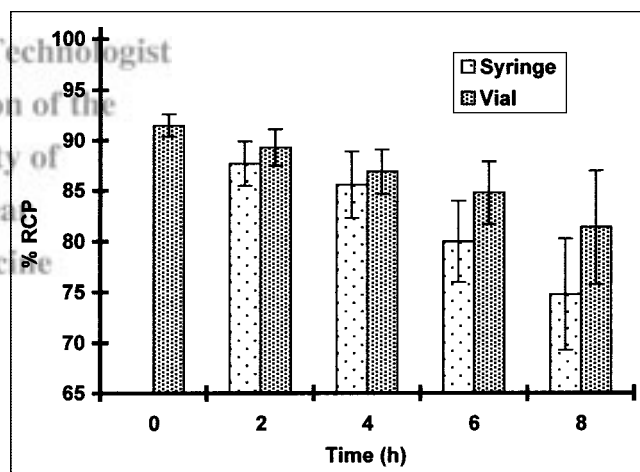


FIGURE 2. RCP of ^{99m}Tc-HMPAO in syringe and vial over time (mean \pm SD; $n = 10$). *Syringe vs. vial:* no significance for 4-h sample; otherwise, significance of at least $P < 0.01$. *Between syringes:* no significance between 2- and 4-h samples; $P < 0.001$ between other samples. *Between vials:* no significance between 4- and 6-h or between 6- and 8-h samples; otherwise, significance of at least $P < 0.05$.

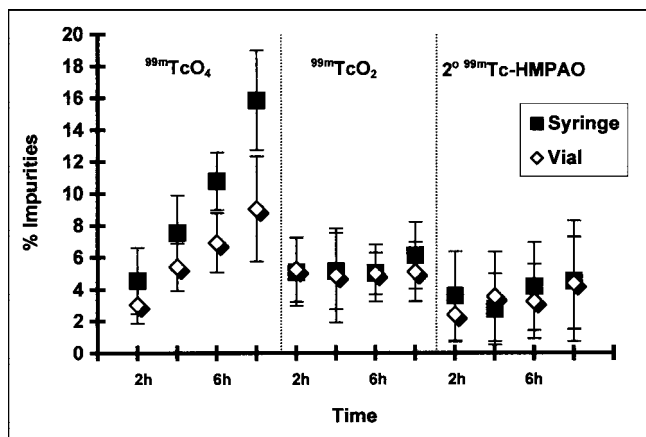


FIGURE 3. % Radiochemical impurities in ^{99m}Tc -HMPAO over time (mean \pm SD; $n = 10$). $^{99m}\text{TcO}_4$: no significant difference between 4- and 6-h sample, or between 6- and 8-h sample in vial; otherwise, $P < 0.05$ at all other time periods in vial, in syringe, and between vial and syringe. $^{99m}\text{TcO}_2$ and 2^{99m}Tc -HMPAO: no significance between samples of any time period.

^{99m}Tc -HMPAO was observed to be 11.6% at 2 h, decreasing to 9.5% at 8 h after preparation. With the filter removed, the syringe was found to contain 4.6% to 4.9% of the initial activity. In contrast, ^{99m}Tc -ECD retained 10.1% to 11.3% of initial activity in the syringe at 2, 4, and 6 h after preparation, with a decrease to 6.2% at 8 h. In comparison, sodium pertechnetate exhibited less than 1% retention in the syringe over the 8-h time period, and approximately 4% of the pertechnetate remained in the filter.

DISCUSSION

To effectively determine the foci of seizures in refractory epilepsy, patients are admitted to the hospital, taken off anticonvulsant medication, and monitored for seizure activity. Brain-imaging agents are injected within 20 s of seizure onset. The optimum agent for this type of procedure should be one that can be prepared early in the morning and remain stable in a syringe for at least 8 h. This experiment was performed to determine which agent, ^{99m}Tc -ECD or stabilized ^{99m}Tc -HMPAO, would be best suited for this type of procedure.

^{99m}Tc -HMPAO appears to be more sensitive than ^{99m}Tc -ECD to the transfer to and storage in another container. Although ^{99m}Tc -ECD maintained its stability of greater than 90% over 8 h in both the vial and syringe, stabilized ^{99m}Tc -HMPAO retained an RCP greater than 80% only when stored in a vial. Storage of the product in a syringe resulted in suboptimal RCP 8 h after preparation. The transfer of stabilized ^{99m}Tc -HMPAO to a syringe apparently increases the rate of reoxidation of bound technetium to sodium pertechnetate.

It is unlikely that the observed decrease in RCP of stabilized ^{99m}Tc -HMPAO 6 to 8 h after preparation would have improved had the manufacturer's recommendation been followed with respect to the use of generator eluate within 30 min of elution. Ballinger et al. (8) showed that the

initial stability of nonstabilized ^{99m}Tc -HMPAO was not affected either by generator ingrowth of 24 h or less, or by use of eluate within 4 h of elution. The 60-min elution limit used in this study was employed solely because of workload considerations in the radiopharmacy.

It should be noted that an 8-h study period is longer than the recommended expiration time for the prepared products (6 h for ^{99m}Tc -ECD; 4 h for stabilized ^{99m}Tc -HMPAO). In addition, the RCP of both products exceeded the manufacturers' requirements at expiration, regardless of the container used. To strictly comply with the manufacturer's package insert, it would be necessary to replace stabilized ^{99m}Tc -HMPAO every 4 h (and ^{99m}Tc -ECD at 6 h), regardless of the in vitro stability. It should also be noted that stability studies using a small sample size of 10 vials might be inadequate to establish an expiration time longer than that specified by the manufacturer.

Retention studies were performed to determine whether either product would be lost in the syringe, thus reducing the dose available for injection into the patient. Plastics that contain little or no phthalate plasticizers (such as polypropylene syringes used in this study) typically do not readily absorb lipid-soluble drugs (9). Adsorption of a product, however, must also be considered given that a clinically relevant amount of product could be easily removed from the solution. The risk of sorption increases with the length of time that the product is exposed to the surface. Any pharmaceutical product that is housed in a container for a prolonged period of time should be tested for compatibility with that container before use.

Both ^{99m}Tc -ECD and stabilized ^{99m}Tc -HMPAO retained approximately 10% of their total activity in the syringe (or syringe/filter, in the case of ^{99m}Tc -HMPAO). With ^{99m}Tc -HMPAO the activity was distributed equally between the syringe and the filter. Filters are not required for the injection.

TABLE 1
Percentage of ^{99m}Tc -ECD and ^{99m}Tc -HMPAO Retained in Syringe and Filter

Radiopharmaceutical	Time after product preparation (h)			
	2	4	6	8
^{99m}Tc -ECD*	10.9 (4.9)	11.3 (1.9)	10.1 (2.9)	6.2 (2.9)
^{99m}Tc -HMPAO				
Total	11.6 (2.1)	11.5 (2.6)	10.0 (2.1)	9.5 (2.5)
Syringe only*	4.6 (2.0)	4.6 (1.8)	4.9 (1.8)	4.9 (2.3)
Filter only†	7.1 (0.8)	6.8 (1.9)	5.1 (1.1)	4.6 (0.8)
$^{99m}\text{TcO}_4$				
Syringe	0.8 (0.4)	0.9 (0.3)	0.8 (0.2)	0.5 (0.2)
Filter†	4.0 (1.0)	4.0 (0.7)	3.2 (0.4)	4.2 (0.7)

Values represent the mean percentage radioactivity retained (\pm 1 SD); $n = 10$.

*Significance of at least $P < 0.01$ between radioactivity retained in syringes containing ^{99m}Tc -ECD and ^{99m}Tc -HMPAO at all time periods tested except at 8 h (no significance).

†Significance of at least $P < 0.001$ between radioactivity retained in filters of ^{99m}Tc -HMPAO and $^{99m}\text{TcO}_4$ at all time periods tested except at 8 h (no significance).

tion of ^{99m}Tc -ECD and therefore were not used in the ^{99m}Tc -ECD retention studies. ^{99m}Tc -ECD, however, exhibited significantly higher retention in the syringe compared with that of either ^{99m}Tc -HMPAO or the control syringes containing $^{99m}\text{TcO}_4$ (^{99m}Tc -ECD; 6.2%–11.3%; ^{99m}Tc -HMPAO, 4.6%–4.9%; $^{99m}\text{TcO}_4$, 0.5%–0.9%; $P < 0.01$). Higher retention of ^{99m}Tc -ECD is not likely caused by the lipophilic nature of the chemical, given that ECD is less lipophilic than HMPAO, based on the higher octanol/water partition coefficient of HMPAO ($\log P = 1.9$, HMPAO; $\log P = 1.64$, ECD) (10). The amount of ^{99m}Tc -ECD or ^{99m}Tc -HMPAO remaining in the syringe was observed to be significantly higher than that of the $^{99m}\text{TcO}_4$ control. This likely reflects some form of sorption to the syringe.

A statistical difference was also observed between the radioactivity retained in the filters containing ^{99m}Tc -HMPAO at 2, 4, and 6 h after preparation and that of the control filters ($P < 0.001$). This observed retention might represent adherence of the product to the filter. A portion of this activity may also reflect the lack of flushing of the syringe/filter after expression of the product from the syringe rather than interaction of the product with the filter. Flushing the syringe/filter with saline would have determined whether activity was irreversibly bound to the container, but this would not have represented the clinical situation (syringes are not flushed during injection because a bolus injection is desirable for the ictal study).

CONCLUSION

From a pharmaceutical point of view, ^{99m}Tc -ECD would be the preferred agent for use in ictal studies. The product displays greater stability than stabilized ^{99m}Tc -HMPAO when stored in a syringe for at least 8 h. Therefore, ^{99m}Tc -ECD is required to be prepared only once during the day. In contrast, two preparations of ^{99m}Tc -HMPAO would be required to provide availability of a product of specified

radiochemical purity throughout an 8-h period. This would increase the cost of the procedure as a result of the need for additional product, generator eluate, and labor.

Retention studies indicate that 10% of the product remains in the syringe/filter, regardless of the product selected. Although sorption of the brain agents does appear to occur, the radiochemical stability of the product should be the main determining factor in the selection of the most appropriate agent for seizure studies.

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REFERENCES

- Oliveira AJ, da Costa JC, Hilario LN, Anselmi OE, Palmieri A. Localization of the epileptogenic zone by ictal and interictal SPECT with ^{99m}Tc -ethyl cysteinate dimer in patients with medically refractory epilepsy. *Epilepsia*. 1999;40:693–702.
- Spanaki MV, Spencer SS, Corsi M, MacMullan J, Seibyl J, Zubal IG. Sensitivity and specificity of quantitative difference SPECT analysis in seizure localization. *J Nucl Med*. 1999;40:730–736.
- Avery RA, Zubal IG, Stokking R, et al. Decreased cerebral blood flow during seizures with ictal SPECT injections. *Epilepsy Res*. 2000;40:53–61.
- McKinnon BT. Factors influencing expiration dates. In: Buchanan EC, McKinnon BT, Scheckelhoff DJ, Schneider PJ, eds. *Principles of Sterile Product Preparation*. Bethesda, MD: American Society of Health-System Pharmacists; 1995:95–103.
- Canadian Society of Hospital Pharmacists. Guidelines for Preparation of Sterile Products in Pharmacies. *CSHP Official Publications 2000*. 2000: 77–86.
- DuPont Pharmaceuticals Company [package insert]. Neurolite® kit for the preparation of technetium Tc99m bicisate for injection. Billerica, MA: DuPont Pharmaceuticals Company; September 1998.
- Amersham Healthcare [package insert]. Ceretec® kit for the preparation of technetium-99m exametazime injection. Arlington Heights, IL: Amersham Healthcare; February 1999.
- Ballinger JR, Gulenchyn KY, Reid RH. Radiopharmaceutical factors in the variable quality of [^{99m}Tc]HM-PAO images of the brain. *J Nucl Med*. 1990;31:118–122.
- Kowaluk EA, Roberts MS, Polack AE. Interactions between drugs and intravenous delivery systems. *Am J Hosp Pharm*. 1982;39:460–467.
- Moretti JL, Caglar M, Weinmann P. Cerebral perfusion imaging tracers for SPECT: which one to choose? *J Nucl Med*. 1995;36:359–363.



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