ORIGINAL ARTICLE

In-house preparation of pentavalent ^{99m}Tc labeled dimercaptosuccinic acid [^{99m}Tc(V)-DMSA], its quality control and clinical applications

Hameed Ullah[1*], Syed Qaiser Shah[2], Aakif Ullah Khan[3], Shakil Ahmad[1], Muhammad Rauf Khattak[1]

ABSTRACT

Background: A convenient method was developed for the preparation of pentavalent ^{99m}Tc dimercaptosuccinic acid using a commercially available meso-2,3-dimercaptosuccinic acid (DMSA) kit. The effects of various parameters, including incubation time, pH, and concentration of sodium bicarbonate on radiochemical purity (RCP) yield were evaluated. To determine tri- and pentavalent fractions, RCP yield of the labeled DMSA was measured using different solvent systems.

Methods: The RCP yield was studied using a thin-layer chromatography on silica gel as the stationary phase and n-butanol/acetic acid/distilled water (3:2:2) as the mobile phase.

Results: It was observed that hydrolyzed $^{99m}TcO_2$ and ^{99m}Tc(III)-DMSA remained at the origin (R_f = 0–0.1), and ^{99m}Tc(V)-DMSA at (R_f = 0.5–0.6), while free pertechnetate, $^{99m}TcO^-_4$ moved to the solvent front (R_f = 0.9–1). However, in acetone, ^{99m}Tc(III)-DMSA, ^{99m}Tc(V)-DMSA, and hydrolyzed $^{99m}TcO_2$ stayed at the origin (R_f = 0–0.1), while free pertechnetate, $^{99m}TcO^-_4$ moved to the solvent front (R_f = 0.9–1). When developed with 5% glycine, $^{99m}TcO_2$ stayed at the origin and the other species migrated to the solvent front. The labeling efficiency of the ^{99m}Tc(V)-DMSA was 96.6± 0.5% and was stable for 24 hours at room temperature.

Conclusion: Higher labeling yield, easy preparation with low cost, and good stability make ^{99m}Tc(V)-DMSA, a promising radio drug for clinical use.

Keywords: Medullary thyroid cancer, ^{99m}Tc(V)-DMSA, radiochemical purity, thin-layer chromatography.

INTRODUCTION

Meso-2,3-dimercaptosuccinic acid (DMSA) is an organosulfur compound containing two sulfhydryl (−SH) groups and is one of the many kits labeled with Technetium-99m (^{99m}Tc) for various medical applications [1]. The labeling of this compound depends upon the labeling techniques and oxidation state of ^{99m}Tc. At acidic pH (2–3), it chelates with ^{99m}Tc and forms a trivalent complex ^{99m}Tc(III)-DMSA used as a renal imaging agent. However, in alkaline pH (8–9), it chelates with ^{99m}Tc in higher oxidation state (+5) and forms a pentavalent complex ^{99m}Tc(V)-DMSA [2,3]. The biological behavior of these radiopharmaceuticals is different from one another. ^{99m}Tc(V)-DMSA is a nonspecific, multifunctional imaging agent that commonly accumulates in medullary thyroid carcinoma (MTC) together with head and neck, brain, lung, liver, breast, and some other soft tissue tumors [4,5]. A variety of other radiopharmaceuticals used for the identification and visualizations of these tumors include iodinated (^{123}I) or (^{131}I) metaiodobenzylguanidine, pentavalent ^{99m}Tc dimercaptosuccinic acid [^{99m}Tc(V)-DMSA], thallium chloride, (^{201}Tl), ^{99m}Tc methoxyisobutylisonitril, ^{99m}Tc ethylenediaminediacetic acid (EDDA)/hydrazinonicotinyl-Tyr (3)-octreotide (HYNIC-TOC), ^{111}In diethylenetriaminepenta-acetic acid-octreotide, and ^{18}F-FDG [6–9]. But the choice of the agent to be used is determined on the basis of local expertise, availability, and cost [10]. ^{99m}Tc(V)-DMSA is the least expensive technique used in most centers. Usefulness of this agent in diagnosis, staging, and restaging for MTC has already been reported from our institute [11,12]. This ^{99m}Tc(V)-DMSA is taken up by the primary as well as recurrent MTC tumor and its metastasis, with sensitivity ranging from 19% to 88% [13,14].

The structural analysis by Nuclear magnetic resonance reveals that in acidic condition (pH 2–3), DMSA forms a hexacoordinated asymmetric bis-complex, in which one molecule is bound to ^{99m}Tc via two -S- bridges and one -O- bridge, while the other is bound via one -S- bridge and two -O- bridges, and one −SH remains free, as shown in Figure 1(a) and in alkaline medium at pentavalent state (with pH range 7.5–8.5), both sulfhydryl groups (−SH) of DMSA are bound with ^{99m}Tc and no free −SH group is left for protein binding. The chemical structure of the ^{99m}Tc(V)-DMSA complex possesses four negatively charged carboxylate groups, and a central anionic technetium

Correspondence to:
Hameed Ullah
*Institute of Radiotherapy and Nuclear Medicine (IRNUM), Peshawar, Pakistan.
Email: hameedullahirnum@gmail.com

Full list of author information is available at the end of the article.

Received: 18 May 2018
Accepted: 01 November 2018

oxobis (dithiolate) core as shown in Figure 1(b).

Different methods have been reported for the synthesis of ^{99m}Tc(V)-DMSA, wherein majority used sodium bicarbonate to increase the pH; while in another method, conversion of trivalent [^{99m}Tc(III)-DMSA] into pentavalent ^{99m}Tc(V)-DMSA has been made by bubbling with pure oxygen [1,3,15]. Furthermore, it is reported that it can be prepared by adding 7% $NaHCO_3$ to the commercial DMSA kit followed by the addition of $Na^{99m}TcO_4$ [1,2,16,17]. The reported radiochemical purity (RCP) in these methods were 90% and 83%, respectively. Further for the characterization of the labeled radiotracer, they have proposed thin-layer chromatography (TLC) method with two different solvent systems, which could not separate well, all the species, the desired complex, ^{99m}Tc(V)-DMSA from radiochemical impurities, ^{99m}Tc (III)-DMSA, hydrolyzed reduced technetium (HR-^{99m}Tc), and free pertechnetate, $^{99m}TcO^-_4$ present in the radiopharmaceutical. However, in our study, we developed a reliable method for the preparation of ^{99m}Tc(V)-DMSA with higher labeling yield of 96.6%. For RCP determination, we used the TLC method with three solvent systems that give proper identity, purity, and quantification of the complexes and impurities. The aim of this work was to develop a liquid DMSA (V) kit, from an already available Renal DMSA kit, optimizing different reaction conditions for good labeling yield and to design a specific method for characterization of DMSA complexes.

MATERIALS AND METHODS

Materials

All the chemicals used in this experiment were of analytical grade. Stannous chloride dihydrate, sodium bicarbonate, and TLC paper were purchased from Merck, Germany. All other chemicals and solvents were obtained from Sigma, (UK). Dimercaptosuccinic acid (DMSA kit) and ^{99}Mo/^{99m}Tc generator were purchased from Isotope Production Division (IPD), PINSTECH, Islamabad, Pakistan. Dose Calibrator CRC-15R from Capintec, Inc (USA), TLC Scanner from Bioscan,Inc, D.C, (USA).

Methods

Preparation of ^{99m}Tc(V)-DMSA

The commercial DMSA kit supplied by IPD, PINSTECH, Islamabad, Pakistan was used for this preparation. This kit contained 1 mg DMSA, 0.35 mg of $SnCl_2.2H_2O$, 0.35 mg of ascorbic acid, and 20 mg of mannitol. We mixed 1 ml of 0.167 N sodium bicarbonate solution to make the medium alkaline. The pH of the reaction mixture determined by pH indicator strips was in the range of 8.4–8.5. Then 20–30 mCi (740–1,110 MBq) of freshly eluted sodium pertechnetate ($Na^{99m}TcO_4$) was added to the vial. The vial was incubated for 15 minutes at room temperature for reaction to complete. The whole process was done under aseptic conditions.

Percent radiochemical purity (% RCP) yield

The RCP was performed by TLC on silica gel (TLC–SG) to determine the labeling yield besides impurities. The TLC strips were cut into 1.5 × 12 cm and were developed in three different solvent systems. Solvent system I, used for the determination of free pertechnetate, $^{99m}TcO^-_4$. Solvent system II, for separation of hydrolyzed $^{99m}TcO_2$, while a solvent system III, containing: n butanol/ acetic acid/distilled water in the ratio of 3:2:2 by volume was used to separate ^{99m}Tc(V)-DMSA from $^{99m}TcO^-_4$ and ^{99m}Tc(III)-DMSA. The developed strips were then analyzed for different fractions using TLC scanner (Bioscan, USA).

Effect of pH

The effect of pH on the radiolabeling yield was studied by carrying out the reaction at pH 1–14. It was observed that at pH (2–3) ^{99m}Tc labels with DMSA in +3 oxidation state (trivalent) used for renal imaging. However, at the basic condition with pH 7.1, the formation of pentavalent DMSA (V) starts and the labeling yield reached a maximum at pH 8.5 by the addition of sodium bicarbonate. Beyond this point, labeling efficiency again starts decreasing with increase in pH.

Effect of sodium bicarbonate

To study the effect of sodium bicarbonate on the rate of

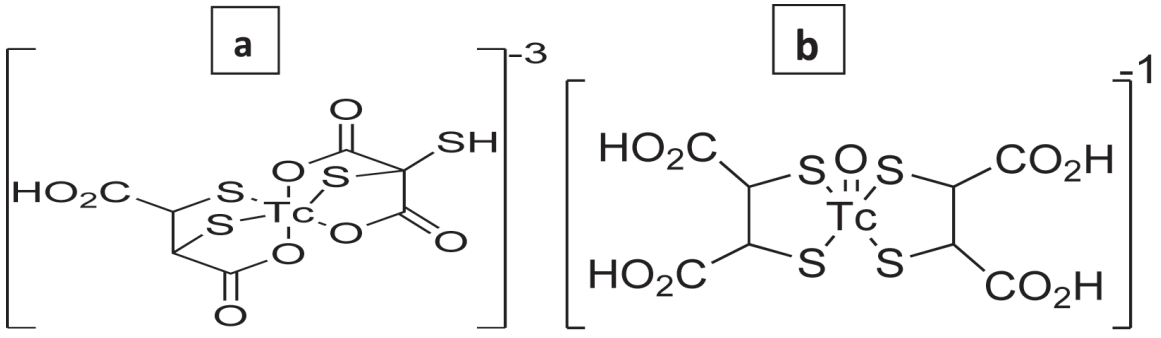

Figure 1. (a) Chemical structure of 99mTc (III) -DMSA) and (b) 99mTc (V)-DMSA.

formation of ^{99m}Tc(V)-DMSA, different concentrations of sodium bicarbonate (ranging from 0.05 to 1 N) were added to the reaction mixture for the elevation of pH. It was observed that radiolabeling yield was maximum when 1 ml of 0.167 N solution of sodium bicarbonate was used for pH adjustment, above and below this concentration, the desired labeling yield cannot be achieved.

RESULTS

Radiochemical quality control

Table 1 shows the range of R_f values for the different species found in the labeling of ^{99m}Tc (V)-DMSA. It is clear that all the species can be separated using the three solvent systems. In solvent system I, TLC-SG is used as a stationary phase and acetone is used as a mobile phase. In this system, ^{99m}Tc(III)-DMSA, ^{99m}Tc(V)-DMSA, and hydrolyzed, $^{99m}TcO_2$ remained at the origin (R_f = 0–0.1), while free pertechnetate, $^{99m}TcO^-_4$ moved to the solvent front. In solvent system II, hydrolyzed, $^{99m}TcO_2$ stayed at the origin but the other species [^{99m}Tc(III)-DMSA, ^{99m}Tc(V)-DMSA, and $^{99m}TcO^-_4$] migrated with the solvent front when 5% glycine was used as a solvent. A solvent system III, containing: n butanol/acetic acid/distilled water in the ratio of 3:2:2 by volume was used in order to separate ^{99m}Tc(V)-DMSA from $^{99m}TcO^-_4$ and ^{99m}Tc(III)-DMSA. In Figure 2, the R_f of $^{99m}TcO_2$ and ^{99m}Tc(III)-DMSA is shown at the origin (0–0.1), while a peak of ^{99m}Tc(V)-DMSA can be seen at R_f (0.5–0.6) and a small peak of free pertechnetate, $^{99m}TcO^-_4$ at solvent front (R_f = 0.9–1).

Incubation time

The effect of time on the labeling yield was studied by taking the aliquots in different reaction vials and measuring the rate of formation of ^{99m}Tc(V)-DMSA at various time intervals starting from 0, 5, 10, 15, 20, 30, and 60 minutes. Figure 3 showed that the duration of the reaction was important to achieve high labeling yield. The labeling observed after 5 minutes was 2.50% ± 0.40%, 12.45% ± 0.55% after 10 minutes, 96.6% ± 0.5% after 15 minutes, and almost 96.58% ± 0.50% after 20 minutes

Table 1. Rf values of DMSA complexes and impurities in different solvent systems.

System I		System II		System III	
R_f = 1 for $^{99m}TcO^-_4$ R_f = 0 for DMSA III, DMSA V & HR-Tc		R_f = 1 for $^{99m}TcO^-_4$ DMSA III, DMSA V & R_f = 0 for HR-Tc		R_f = 1 for $^{99m}TcO^-_4$ R_f = 0.5 for DMSA V R_f = 0 for HR-Tc & DMSA III	
St. Phase TLC-SG	Mob. Phase Acetone	St. Phase TLC-SG	Mob. Phase 5% Glycine	St. Phase TLC-SG	Mob. Phase nButanol/acetic acid/ Dist.H_2O

R_f, relative front; HR-Tc, hydrolyzed reduced technetium; Dist.H_2O, distilled water; St. Ph, stationary phase; Mob. Ph, mobile phase.

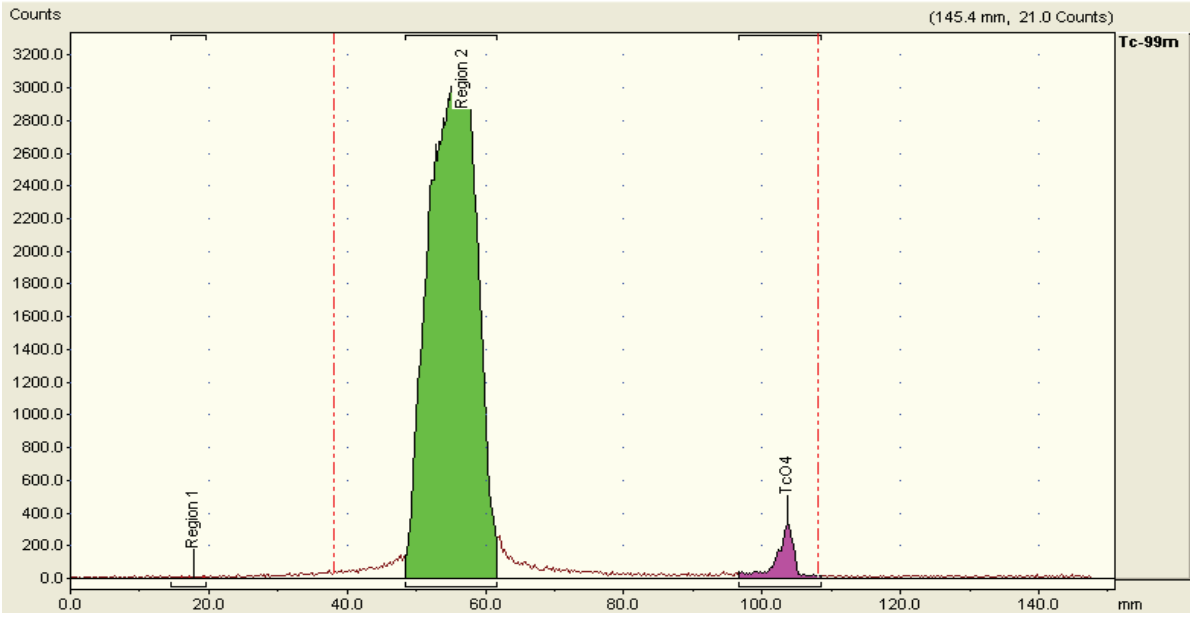

Figure 2. Peak of labeled 99mTc(V)-DMSA at Rf = 0.5–0.6 and free 99mTcO4 at Rf = 0.9–1.

that remains constant up to 60 minutes.

Effect of pH

Figure 4 showed the effect of pH on the radiolabeling yield of ^{99m}Tc(V)-DMSA. It was found that the maximum labeling efficiency was 96.6% ± 0.5% at pH 8.5, above and below this pH, the yield was lower. Further, it was observed that at pH 5, the labeling efficiency of ^{99m}Tc(V)-DMSA was 6.4% ± 0.4% as shown in Figure 5, hence pH 8.5 was selected as optimal in the final formulation.

Effect of sodium bicarbonate

Figure 6 showed that the rate of formation of ^{99m}Tc(V)-DMSA increases with increase in the concentration of sodium bicarbonate. It was found that the labeling efficiency reached to a maximum when 1 ml of 0.167 N sodium bicarbonate solution was used for pH adjustment, above this range further increase in concentration causes decrease in the radiolabeling yield.

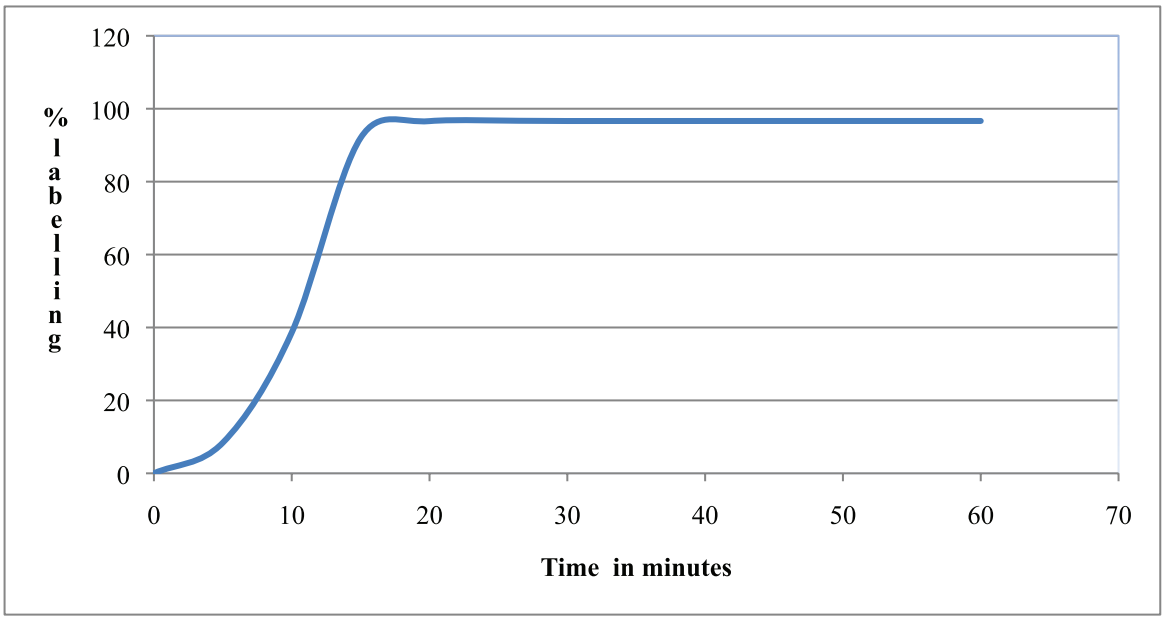

Figure 3. Effect of time on radiolabeling of 99mTc(V)-DMSA.

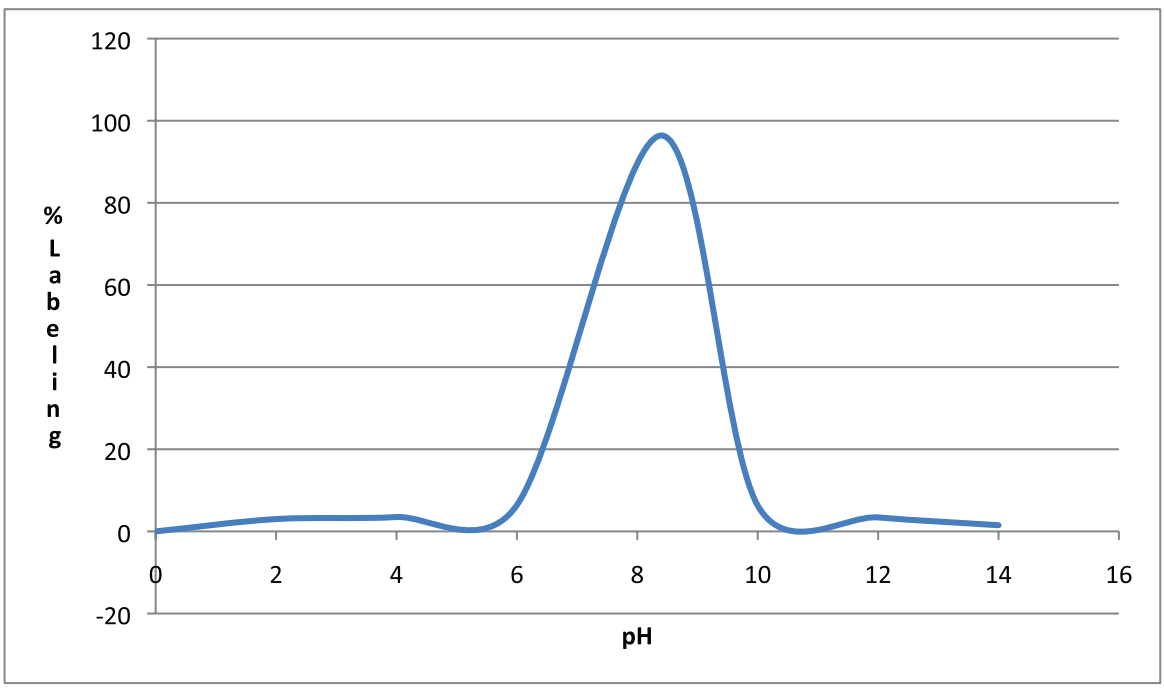

Figure 4. Effect of pH on the radiolabeling yield of 99mTc(V)-DMSA.

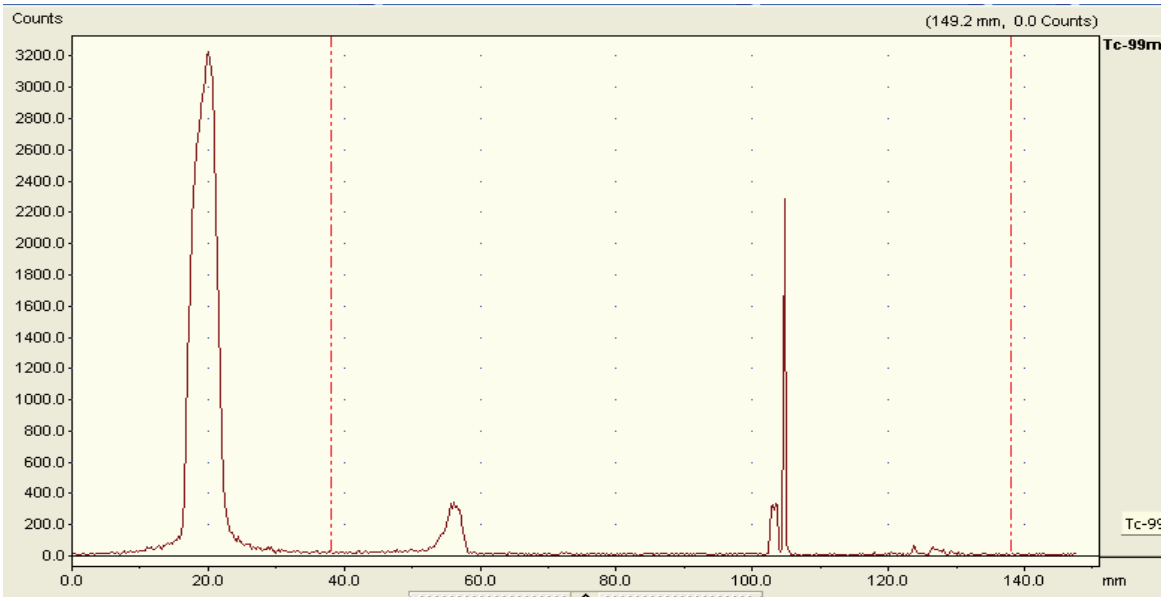

Figure 5. Chromatogram of DMSA labeling at pH 5 shows a maximum yield of 99mTc (III)-DMSA.

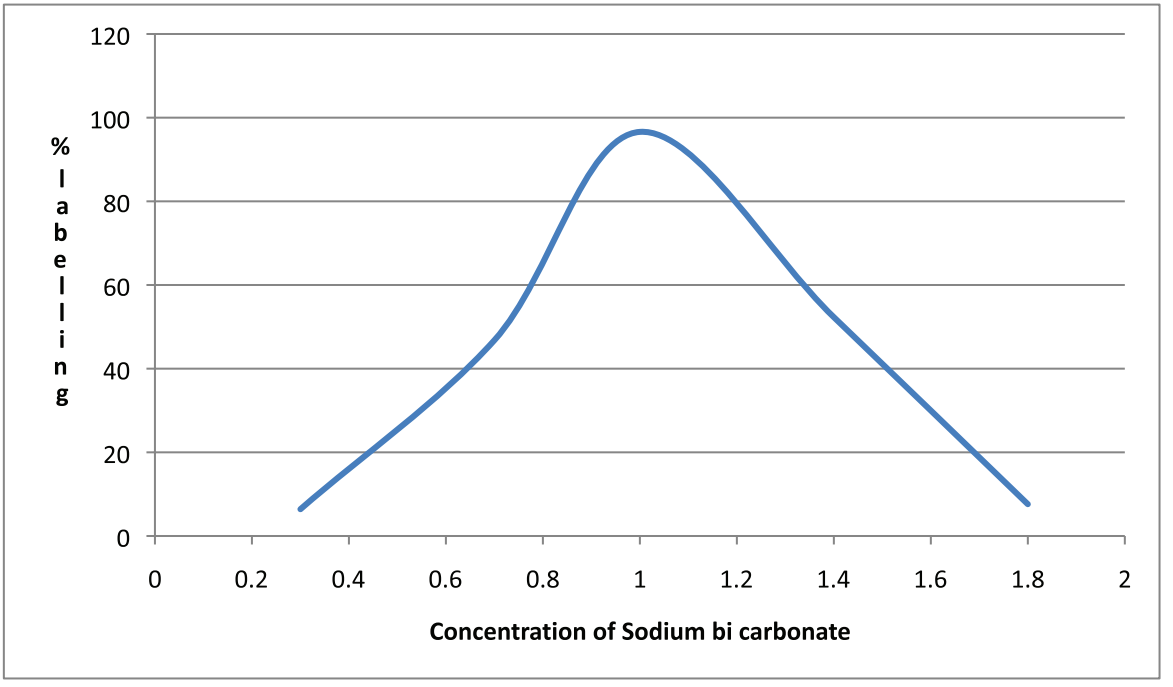

Figure 6. Effect of concentration of sodium bicarbonate on radiolabeling of 99mTc(V)-DMSA.

Stability in normal saline

TLC method with three mobile systems was used for evaluating the stability of ^{99m}Tc(V)-DMSA in saline at 37°C for up to 24 hours. In this investigation, 0.1 ml of the radiolabeled compound was incubated with 1 ml of normal saline. The strips were again developed in acetone and normal saline as the mobile phase. The developed strips were then analyzed for different fractions using TLC scanner. It was observed that the RCP of the complex was 96.2% ± 0.5% after 1 hour and still 92.3% ± 0.4% at 24 hours.

DISCUSSION

Pentavalent ^{99m}Tc dimercaptosuccinic acid [^{99m}Tc(V)-DMSA] is a well-established imaging tool most commonly used for the evaluation of MTC and other soft tissue tumors [2,18,19]. DMSA forms two different complexes with ^{99m}Tc at different pH ranges. At acidic pH (2–3), it chelates with ^{99m}Tc and forms ^{99m}Tc(III)-DMSA, but at basic condition (7.5–8.5), it forms ^{99m}Tc(V)-DMSA. In these DMSA complexes, the metal ^{99m}Tc can occur at +3 or +5 oxidation state [1,2]. The biological behavior of these complexes is different and hence shows altered

organ distribution [2]. Hence, the efficacy of this radiopharmaceutical agent depends upon the method of preparation used.

Radiopharmaceuticals quality control has a vital role in nuclear medicine imaging procedures [20]. The reliable outcome from these procedures depends upon the radiolabeling efficiency of the radiopharmaceuticals used [21]. Various chromatographic techniques are used for the determination of RCP depending upon the availability and sensitivity of the instrument, as well as the expertise of the professional, but none can be considered alone as universal to distinct possible forms of the complex. In our study, we used three solvent systems that enable the separation of the desirable product from undesirable impurities present in the ^{99m}Tc-DMSA.

The choice of a radiopharmaceutical agent depends upon its availability, financial implications, safety, sensitivity, and ability to detect greater lesions [10,12,18]. ^{99m}Tc (V)-DMSA has a high affinity for tumors, mainly used for the detection of MTC, brain, and some soft tissue tumors [4,22,25]. Moreover, ^{99m}Tc (V)-DMSA shows avid and persistent uptake in all the tumor deposits in the thoracic region but low uptake in the liver and spleen if compared with EDDA/HYNIC-Tyr^3 octreotide scintigraphy, hence the uptake in these regions will be clearly appreciated where it will be overshadowed in octreotide scan. It offers several advantages over other radiopharmaceuticals. It is safe, cheap, and readily available in each nuclear medicine facility and can be prepared as an in-house radiopharmaceutical with higher sensitivity from renal DMSA kit [12,15].

Although ^{99m}Tc(V)-DMSA is a promising imaging agent, the exact mechanism of its uptake in tumors is not well understood [23,24]. One study suggested the pH-sensitive character to be one of the factors influencing its accumulation in cancer cells [25]. In another study by Papantoniou and colleagues suggested that the uptake of ^{99m}Tc(V)-DMSA by breast tumors is related to proliferative activity, which is either directly related to tumor grade or to the mitotic activity and the cellular proliferation of breast tumors or lesions [26]. It is also suggested by several studies that the uptake of ^{99m}Tc-(V)-DMSA may be related to the structural similarity between the ^{99m}Tc(V)-DMSA core (phosphate like ion TcO_4^{-3}), and the PO_4^{3-} anion which is avidly taken up by some cancer cells [27]. Ohta et al. [28] have also postulated that ^{99m}Tc(V)-DMSA core resembles phosphate ion in its distribution pattern, and that is the mechanism by which the radiopharmaceutical accumulates in MTC. But some studies suggest a more cell-specific uptake of ^{99m}Tc(V)-DMSA than the phosphate localization [15,29]. Whatever the mechanism of uptake is, ^{99m}Tc(V)-DMSA seems to be a good radiopharmaceutical with the potentials to examine its utility as a nuclear medicine marker.

CONCLUSIONS

Locally formulated ^{99m}Tc(V)-DMSA is an economical radiopharmaceutical due to the convenient procedure, easy availability, low cost, good stability, and radiologically safe [30]. It can be used as an alternative imaging agent in centers where PET/CT is not available. The procedure employed for investigations of RCP of DMSA complexes is simple, convenient, and suitable for the identification of free pertechnetate, TcO_4, and hydrolyzed TcO_2 besides tri- or pentavalent DMSA complexes.

List of Abbreviations

EDDA	Ethylenediaminediacetic acid
FDG	Fluorodeoxyglucose
HYNIC-TOC	Hydrazinonicotinyl-Tyr (3)-octreotide
MTC	Medullary Thyroid Carcinoma
RCP	Radiochemical purity
^{99m}Tc(V)-DMSA	Technetium-99m pentavalent dimercaptosuccinic acid
TLC	Thin-layer chromatography

Funding

None.

Conflict of interests

The authors declare that there is no conflict of interest regarding the publication of this article.

Consent for publication

Written informed consent was taken from the participants of the study.

Ethical approval

Ethical Committee of the Institute approved this study for publication.

Author details

Hameed Ullah[1], Syed Qaiser Shah[2], Aakif Ullah Khan[3], Shakil Ahmad[1], Muhammad Rauf Khattak[1]

1. Institute of Radiotherapy and Nuclear Medicine (IRNUM), Peshawar, Pakistan
2. Institute of Chemical Sciences (ICS), University of Peshawar, KPK, Pakistan
3. Swat Institute of Nuclear Medicine, Oncology and Radiotherapy (SINOR), Swat, Pakistan

REFERENCES

1. Garnuszek P, Pawlak D, Maurin M, Jankovic D, Karczmarczyk U, Mikołajczak R. Comparison of chromatographic methods for quality control of DMSA complexes with ^{99m}Tc and 188Re at (III) and (V) oxidation states. Nucl Med Rev. 2012;15(2):95–100.
2. Shukla J, Mittal BR. Dimercaptosuccinic acid: a multifunctional cost effective agent for imaging and therapy. Indian J Nucl Med. 2015;30(4):295–302. https://doi.org/10.4103/0972-3919.164015
3. International Atomic Energy Agency. Technetium–99m radiopharmaceuticals: manufacture of kits. Technical Reports Series, Vienna, p. 85, 2008; https://doi.org/10.2172/4556052
4. Froberg AC, de Jong M, Nock BA, Breeman WA, Erion JL, Maina T, et al. Comparison of three radiolabelled peptide analogues for CCK–2 receptor scintigraphy in medullary thyroid carcinoma. Eur J Nucl Med Mol Imaging. 2009;36(8):1265–72. https://doi.org/10.1007/s00259-009-1098-9
5. Czepczyński R, Parisella MG, Kosowicz J, Mikołajczak R, Ziemnicka K, Gryczyńska M, et al. Somatostatin receptor scintigraphy using ^{99m}Tc–EDDA/HYNIC–TOC in patients with medullary thyroid carcinoma. Eur J Nucl Med Mol Imaging. 2007;34:1635–45. https://doi.org/10.1007/s00259–007–0479–1
6. Niafer M, Dabiri S, Bozorgi F, Niafer F, Gholami N. Metastatic medullary thyroid carcinoma: a case report. J Res Med Sci. 2011;16(4):568–73.

7. Subedi N, Prestwich R, Chowdhury F, Patel C, Scarsbrook A. Neuroendocrine tumours of the head and neck: anatomical, functional and molecular imaging and contemporary management. Cancer Imaging. 2013;13(3):407–22. https://doi.org/10.1102/1470-7330.2013.0034
8. Nicolini S, Ambrosini V, Fantini L, Fanti S. 68Ga DOTANOC PET/CT detects medullary thyroid cancer relapse at bone level. Clin Nucl Med. 2012;37:591–2. https://doi.org/10.1097/RLU.0b013e31824c5eab
9. Evangelia S, Maria P, Ioannis DE. The role of nuclear medicine in imaging and therapy of neuroendocrine tumors. Hospital Chronicles. 2012;7(2):81–90.
10. Khan AN, Syed GMS, Durr–e–Sabih, Siraj QH. Radionuclide liver imaging: Is there a role? Pak J Nucl Med. 2014;4:1–39.
11. Khan AU, Ahmad S, Manan A, Khan AA. Visualization of medullary carcinoma thyroid with locally formulated ^{99m}Tc (V)–DMSA and comparison with 131I–MIBG Scintigraphy. J Coll Physicians Surg Pak. 2001;11(6):394–6.
12. Khan AU, Hameedullah, Bahadur A, Khattak MR, Shah AS. Mediastinal spread of medullary thyroid carcinoma imaged by locally formulated ^{99m}Tc–DMSA(V). Pak J Nucl Med. 2013;1(3):73–7.
13 Howe TC, Padhy AK, Loke K, Magsombol B, Ng D, Goh A. Role of Tc–^{99m} DMSA (V) scanning and serum calcitonin monitoring in the management of medullary thyroid carcinoma. Singapore Med J. 2008;49:19–22.
14. Meijer JA, le Cessie S, van den Hout WB, Kievit J, Schoones JW, Romijn JA, et al. Calcitonin and carcinoembryonic antigen doubling times as prognostic factors in medullary thyroid carcinoma: a structured meta–analysis. Clin Endocrinol (Oxf). 2010;72:534–42. https://doi.org/10.1111/j.1365-2265.2009.03666.x
15. Al–Saeedi FJ, Mathew PM, Luqmani YA. Assessment of tracer ^{99m}Tc(V)–DMSA uptake as a measure of tumor cell proliferation in vitro. PLoS One. 2013;8(1):1–7. e54361. https://doi.org/10.1371/journal.pone.0054361
16. Adams BK, Fataar A, Byrne MJ, Levitt NS, Matley PJ. Pentavalent Technetium–99m (V)—DMSA uptake in a pheochromocytoma in a patient with sippleís syndromeî. J Nucl Med. 1990;31:106–8.
17. Hirano T, Tomiyoshi K, Zhang YJ, Ishida T, Inoue T, Endo K. Preparation and clinical evaluation of technetium–99m dimercaptosuccinic acid for tumour scintigraphyî. Eur J Nucl Med. 1994;21:82–5. https://doi.org/10.1007/BF00182308
18. Krishnamurthy A, Kumar RK, Ravishankaran P, Ramshankar V, Begum ASB, Rangarajan GK. Exploring the role of technitium–99m dimercaptosuccinyl acid (V) scan in medullary carcinoma thyroid patients with postoperative persistent hypercalcitoninemia in the era of positron emission tomography–computerized tomography. Indian J Nucl Med. 2014;29(3):146–50. https://doi.org/10.4103/0972-3919.136562
19. Kobayashi H, Sakahara H, Hosono M, Shirato M, Endo K, Kotoura Y, et al. Soft–tissue tumors: diagnosis with ^{99m}Tc(V) dimercaptosuccinic acid scintigraphy. Radiology. 1994;190(1):277–80. https://doi.org/10.1148/radiology.190.1.8259419
20. Sadeghpour H, Alavi M, Shahedi M, Entezarmahdi SM, Sakhteman A. Evaluation of radiochemical purities of some radiopharmaceuticals in Shiraz Namazi teaching hospital. Trends Pharm Sci. 2015;1(1):15–9.
21. Vallabhajosula S, Killeen RP, Osborne JR. Altered biodistribution of radiopharmaceuticals: role of radiochemical/pharmaceutical purity, physiological, and pharmacologic factors. Semin Nucl Med. 2010;40:220–41. https://doi.org/10.1053/j.semnuclmed.2010.02.004
22. Tsiouris S, Pirmettis I, Chatzipanagiotou T, Ptohis N, Papantoniou V. Pentavalent technetium–99m dimercaptosuccinic acid [^{99m}Tc–(V)DMSA] brain scintitomography—a plausible non–invasive depicter of glioblastoma proliferation and therapy response. J Neurooncol. 2007;85:291–5. https://doi.org/10.1007/s11060-007-9410-7
23. Al–Saeedi F. Role of ^{99m}Tc–(V)DMSA in detecting tumor cell proliferation. Anal Chem Insights. 2007;2:81–3. https://doi.org/10.4137/117739010700200001
24. Abdullah MM, Almohanadi S, Abulhuda F, Usmani S. Pentavalent [^{99m}Tc] DMSA and serum calcitonin in evaluation of patients with medullary carcinoma of the thyroid. AAMJ. 2008;6(3):116–27.
25. Horiuchi K, Saji H, Yokoyama A. ^{99m}Tc (V)–DMSA tumor localization mechanism: a pH sensitive Tc(V)– DMSA– enhanced target/nontarget ratio by glucose–mediated acidosis. Nucl Med Biol. 1998;25(6):549–55. https://doi.org/10.1016/S0969-8051(98)00012-2
26. Papantoniou V, Tsiouris S, Mainta E, Valotassiou V, Souvatzoglou M, Sotiropoulou M, et al. Imaging in situ breast carcinoma (with or without an invasive component) with technetium–99m pentavalent dimercaptosuccinic acid and technetium–99m 2–methoxy isobutyl isonitrile scintimammography. Breast Cancer Res. 2005;7(1):R33–45. https://doi.org/10.1186/bcr948
27. Denoyer D, Perek N, Le Jeune N, Frere D, Dubois F. Evidence that ^{99m}Tc–(V)–DMSA uptake is mediated by NaPi cotransporter type III in tumour cell lines. Eur J Nucl Med Mol Imaging. 2004;31(1):77–84. https://doi.org/10.1007/s00259-003-1334-7
28. Ohta H, Endo K, Fujita T, Konishi J, Torizuka K, Horiuchi K, et al. Clinical evaluation of tumour imaging using ^{99m}Tc(V) dimercaptosuccinic acid, a new tumour–seeking agent. Nucl Med Commun. 1988;9(2):105–16.
29. Denoyer D, Perek N, Le Jeune N, Cornillon J, Dubois F. Correlation between ^{99m}Tc–(V)–DMSA uptake and constitutive level of phosphorylated focal adhesion kinase in an in vitro model of cancer cell lines. Eur J Nucl Med Mol Imaging. 2005;32(7):820–7. https://doi.org/10.1007/s00259-005-1773-4
30. Skoura E, Rondogianni P, Alevizaki M, Tzanela M, Tsagarakis S, Piaditis G, et al. Role of [18F]FDG–PET/CT in the detection of occult recurrent medullary thyroid cancer. Nucl Med Commun. 2010;31(6):567–75. https://doi.org/10.1097/MNM.0b013e3283384587