Scandium-44: Benefits of a Long-Lived PET Radionuclide Available from the ^{44}Ti/^{44}Sc Generator System

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Abstract: ^{44}Ti/^{44}Sc radionuclide generators are of interest for molecular imaging. The 3.97 hours half-life of ^{44}Sc and its high positron branching of 94.27% may stimulate the application of ^{44}Sc-labeled PET radiopharmaceuticals. This review describes the current status of ^{44}Ti production, ^{44}Ti/^{44}Sc radionuclide generator development, post-processing of generator eluates towards medical application, identification of ligands adequate to Sc^{III} co-ordination chemistry, proof-of-principle labeling of ^{44}Sc-DOTA-octreotides, investigation of *in vitro* and *in vivo* parameters, and initial applications for molecular imaging – both in small animals and humans.

Keywords: Titanium-44, Scandium-44, Radionuclide generator, Post-processing, DOTA, Octreotide, Positron emission, PET.

INTRODUCTION

Radionuclide generator systems providing positron-emitting daughters of extended physical half-life are of new interest [1]. Table **1** shows the most interesting radionuclide generator systems providing positron emitting daughters. Recently, the ^{68}Ge/^{68}Ga radionuclide generator has shown significant potential for molecular imaging [1-3]. The high positron branching of 89% and the possibility of kit-type formulation of radiopharmaceuticals offer excellent prerequisites for the routine use of ^{68}Ga-labeled tracers in nuclear medicine using state-of-the-art positron emission tomography (PET) and PET/CT cameras. However, the physical half-life of ^{68}Ga ($t_{½}$ = 67.71 min) might limit the spectrum of clinical applications of ^{68}Ga-labeled radiodiagnostics. Furthermore, ^{68}Ga-labeled analogues of endoradiotherapeuticals of longer biological half-life (such as ^{90}Y- or ^{177}Lu-labeled peptides and proteins) cannot be directly used to determine individual radiation dosimetry.

In this context, the ^{44}Ti/^{44}Sc radionuclide generator is of interest. The 3.97 hours half-life of ^{44}Sc and its high positron branching of 94.27% may stimulate the application of ^{44}Sc-labeled PET radiopharmaceuticals. ^{44}Sc-labeled PET radiopharmaceuticals appear of interest for molecular imaging of longer lasting physiological processes.

A further scope is expected in the context of personalized patient treatment: Patients supposed to be treated by e.g. Me^{III}-based endoradiotherapeuticals (such as e.g. ^{177}Lu-DOTA-octreotides or other tumor targeting vectors) may receive an individual dose derived from pre-therapeutic ^{44}Sc-PET/CT measurements covering the relevant uptake kinetics. In a similar context, a particular application may lie in the radionuclide pair ^{44}Sc (diagnosis) and ^{47}Sc (therapy), representing a real *Theranostic* [2].

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In the 1960s and 1970s, several strategies were investigated to design a ^{44}Ti/^{44}Sc generator [4-7]. These studies have been typical radiochemical ones, using relatively low activities of ^{44}Ti and did not involve pharmaceutical aspects. The most recent studies on a ^{44}Ti/^{44}Sc generator described the concept and experimental parameters of a 5 mCi (185 MBq) generator system, utilizing the anion-exchange resin Bio-Rad AG 1-X8 (200-400 mesh, Cl^--form) (Filosofov *et al*., 2009) [8].

Finally, ^{44}Sc offers – in addition to the high-abundance positron emission – a high energy and high-abundant photon emission of 1157 keV / 98.95% [9]. While this may be considered when calculating radiation doses (cf. ^{124}I and others), it represents a unique nuclear decay parameter promising for 3-photon-coincidence imaging with potentially improved local resolution characteristics.

THE NUCLIDES: ^{44}Ti AND ^{44}Sc

An important characteristic of positron-emitting ^{44}Sc is its cyclotron-independent availability *via* the ^{44}Ti/^{44}Sc radionuclide generator system. The long-lived ^{44}Ti generates ^{44}Sc, which subsequently transforms to stable ^{44}Ca. Similar to the ^{68}Ge/^{68}Ga radionuclide generator, this generation of a radionuclide daughter with a physical half-life significantly shorter than its predecessor is among the few exceptions of the general tendencies within the β-decay processes. Nucleon binding energies of ^{44}Ti and ^{44}Sc do not differ significantly. Mass excess values referred to Δ=0 for ^{12}C for the proton-deficient arm of the 44 isobar are ^{44}V = -23859 keV, ^{44}Ti = -37548.3 keV, ^{44}Sc = -37815.8 keV, ^{44}Ca = -41469.1 keV [9], causing a delay in the ^{44}Ti → ^{44}Sc transformation kinetics.

^{44}Sc

According to most recent compilations [9], the positron branching of ^{44}Sc is 94.27% and its physical half-life is $t_{½}$ = 3.97 h ([10] gives $t_{½}$ = 3.927 h). There is a subsequent 1157 keV (98.95%) photon emission of ^{44}Sc. Important parameters

1874-4729/12 $58.00+.00

Table 1. Overview on Radionuclide Generators Relevant to PET [1]

Generator	Parent	Daughter			
	$t_{1/2}$	$t_{1/2}$	β^+_{branch} (%)	$E_{\beta}+$ (MeV)	Principal Application
^{82}Sr/^{82}**Rb**	25.55 d	1.273 min	95.43	1.479	Perfusion
^{140}Nd/^{140}**Pr**	3.37 d	3.39 min	51.0	1.067	Perfusion
^{118}Te/^{118}**Sb**	6.00 d	3.6 min	73.5	1.186	Perfusion
^{122}Xe/^{122}**I**	20.1 h	3.63 min	78.0	1.41	(Labeling)
^{128}Ba/^{128}**Cs**	2.43 d	3.62 min	68.9	1.27	Perfusion
^{134}Ce/^{134}**La**	3.16 d	6.45 min	64.0	1.206	Perfusion
^{62}Zn/^{62}**Cu**	9.26 h	9.673 min	97.430	1.314	Labeling; Perfusion
^{52}Fe/^{52m}**Mn**	8.275 h	21.1 min	95.0	1.17	Perfusion
^{68}Ge/^{68}**Ga**	270.95 d	67.71 min	89.14	0.8295	Perfusion →Labeling
^{110}Sn/^{110m}**In**	4.11 h	69.1 min	63.0	1.04	Labeling
^{44}Ti/^{44}**Sc**	60.0 a	3.97 h	94.27	0.632	Labeling
^{72}Se/^{72}**As**	8.40 d	26.0 h	87.8	1.17	Labeling

Table 2. Comparison of ^{68}Ga, ^{44}Sc and ^{89}Zr Mean ($\bar{E}_{\beta}$) and Maximum ($E_{\beta,max}$) Positron Energies [10] and Theoretical Specific Activities A_{spec} [9] Relative to ^{18}F and ^{15}O

Positron Emitter	Half-life	$\bar{E}_{\beta}$	$E_{\beta, max}$	A_{spec} (theory)
		(MeV)		(Ci / g)
^{68}Ga	67.7 min	0.829	1.899	$4.060 \cdot 10^7$
^{44}Sc	3.97 h	0.632	1.474	$1.814 \cdot 10^7$
^{89}Zr	78.41 h	0.396	0.902	$4.489 \cdot 10^5$
^{18}F	109.77 min	0.250	0.634	$9.516 \cdot 10^7$
^{15}O	2.04 min	0.735	1.732	$6.135 \cdot 10^9$

relevant to molecular imaging of the ^{44}Sc in comparison to other metallic positron emitters of interest for PET/CT, such as positron maximum and mean energies, as well as theoretical specific activities are summarized in Table **2** and compared with ^{18}F and ^{15}O. For the metallic positron emitters used in human studies, i.e. ^{68}Ga, ^{44}Sc and ^{89}Zr, mean kinetic energies of the positrons emitted range from about 400 to 800 keV. The value for ^{44}Sc is 0.632 MeV [10].

It should be noted, that these ^{44}Sc activities *per se* represent no-carrier-added (nca) situations. Supposedly the chemicals used in ^{44}Ti / ^{44}Sc radiochemical separations are free of contents of natural scandium salts, the ^{44}Sc is even carrier-free, thus reflecting the theoretical maximum specific activity of ^{44}Sc. As the theoretical specific activities follow the natural logarithm of the nuclide half-life, specific activities of ^{44}Sc are about a factor of 2 lower than those of ^{68}Ga.

^{44}Ti

The ^{44}Ti half-life varies in different studies from 39 to 66.6 years. Most recent studies revealed a half-life of 60 ± 1 years, cf. [11] and references therein. It transforms directly into the ground state of ^{44}Sc *via* electron capture, emitting low energetic photons at 67.9 keV and 78.3 keV.

^{44}Ti / ^{44}Sc RADIONUCLIDE GENERATOR

Either the parent (1) is longer-lived compared to the daughter nuclide (2), but not more than by a factor of about 100, i.e. $t_{1/2,1} < 100\ t_{1/2,2}$, or the parent is much longer-lived than the daughter ($t_{1/2,1} \gg t_{1/2,2}$, i.e. $\lambda_1 \ll \lambda_2$), The limiting case of radioactive equilibrium at relationships between decay constants of $\lambda_1 \ll \lambda_2$ is called secular equilibrium, i.e. the parent activity does not decrease measurably during many daughter half-lives. In the case of $t_{1/2,1} \gg t_{1/2,2}$ the maximum activity of the daughter occurs at the time, t, which is calculated by

$$t = \frac{1}{\lambda_2 - \lambda_1} \ln \frac{\lambda_2}{\lambda_1}.$$

The ^{44}Ti/^{44}Sc generator clearly represents a secular equilibrium system with a half-life ratio between parent and daughter of ca. 130 000 [1]. Consequently, 50% of saturation activity is generated every 3.97 hours, and identical ^{44}Sc batch activities very close to saturation may be eluted each day. Thus, ^{44}Sc activities derived from the generator every day correspond to nominal ^{44}Ti activities (supposing ^{44}Sc is separated quantitatively).

Absolute radioactivites of ^{44}Sc and ^{44}Ti are measured by γ-spectrometry using a high-purity germanium (HPGe) de-

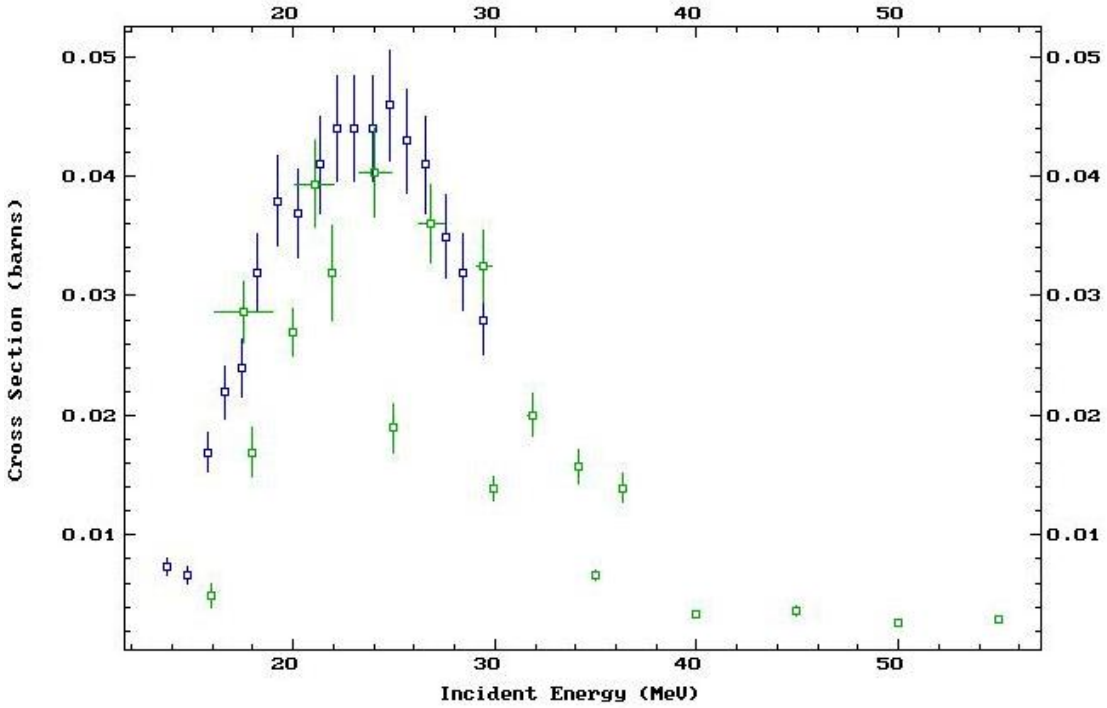

Fig. (1). Cross sections of the $^{45}Sc(p,2n)^{44}Ti$ Process [14].

tector. ^{44}Sc is quantitatively registered best *via* its 511 keV emission. For detection of $^{44}Ti(^{44}Sc)$, the 1157.0 (99.9%) and 1499.5 (0.91%) keV γ-emissions may be used at decay equilibrium. Measurements of ^{44}Ti radioactivity in this case should be performed not earlier than 120 h after ^{44}Sc measurements (more than 30 half-lives of ^{44}Sc). ^{44}Ti radioactivity itself is measured at 67.9 and 78.3 keV. Measurement of ^{44}Sc at high radioactivity is also accomplished in a dose calibrator. The Curie-meter ^{18}F-setting may be used with a multiplication factor of 0.7 to account for absolute ^{44}Sc activity.

^{44}Ti Production

Potential routine application of the promising radionuclide ^{44}Sc for PET/CT based molecular imaging may be best achieved *via* the $^{44}Ti/^{44}Sc$ radionuclide generator. It is worth mentioning, that of course there is the option of direct production of ^{44}Sc, *via* e.g. the $^{44}Ca(p,n)$ process. Nuclear cross sections of the latter process appear adequate to produce sufficient batch activities of ^{44}Sc. Radiochemical separation of nca ^{44}Sc from macroscopic amounts of the target material may be organized efficiently within a fraction of the physical half-life of ^{44}Sc. However, this aspect is not within the scope of this review.

$^{45}Sc(p,2n)^{44}Ti$ Nuclear Reaction

The most convenient approach towards ^{44}Sc would be a well-functioning $^{44}Ti/^{44}Sc$ radionuclide generator. However, not only the $^{44}Ti/^{44}Sc$ generator design represent challenges for basic radiochemistry, but also the high-yield production of the generator parent ^{44}Ti. ^{44}Ti is available in high concentration within supernovas in our universe; however, man-made production of this long-lived radionuclide needs sophisticated production routes *via* accelerated particles. There are two principal approaches, namely the direct production *via* mainly proton-induced nuclear reactions on natural scandium, spallation processes of type (p,xnyp) on targets such as vanadium, and Ti(n,xn) processes, for example. The latter approach was investigated recently *via* the $^{46}Ti(n,3n)^{44}Ti$ reaction [12] in order to evaluate the thermal release of ^{44}Sc from titanium metal [13].

$^{44}Ti/^{44}Sc$ radionuclide generators described in the literature so far are all based on the Sc(p,2n) production pathway. Cross sections are reproduced in Fig. (**1**), cf. [14]. Integral yields significantly depend on the proton energy. Maximum cross sections are at about 22-25 MeV. However, integral yields of ^{44}Ti increase at higher energy, and are about twice as high at 30 MeV proton energy [15].

The production rate limiting parameter is the long physical half-life of ^{44}Ti. Nuclear reaction saturation yields are achieved at $1-\exp^{(-\lambda \cdot t_{irr})}$ or $1-\exp^{(-\ln 2 \cdot t_{irr}/t_{1/2})}$, ($\lambda$ = nuclear decay constant, t_{irr} = period of irradiation, $t_{1/2}$ = physical half-life). Saturation is obtained at $1-\exp^{(-\ln 2 \cdot t_{irr}/t_{1/2})} = 0$, i.e. $t_{irr} / t_{1/2} \rightarrow \infty$. A 0.5 value of saturation is approached at $t_{irr} = t_{1/2}$, meaning an irradiation period of about 60 years. Realistic irradiation periods of e.g. 1 day or even 1 week offer saturation factors of 0.000032 and 0.000222 only, respectively. This can be compensated only in the case of high beam current irradiations, ideally approaching mA values of proton beam flux. Thus, the $^{45}Sc(p,2n)^{44}Ti$ process seems to be an effective nuclear reaction, however, cyclotrons of high proton flux are mandatory. A recent study estimated, that at 30 MeV proton energy and 1 mA proton beam intensity, a full one week irradiation would produce 560 MBq (15 mCi) of ^{44}Ti.

$^{44}Ti/^{44}Sc$ generators described in the literature [4-8] basically made use of relatively low radioactivity levels in the

Table 3. Distribution Coefficients of Ti^{IV} and Sc^{III} for Various HCl / $H_2C_2O_4$ Mixtures for Cation and Anion Exchange Resins [8]. Highlighted Concentrations and K_d-values are Relevant for Further Generator Handling

Concentration of solution, mol/L		K_d			
		AG 50-X8 (cation exchanger)		AG 1-X8 (anion exchanger)	
$H_2C_2O_4$	HCl	Ti^{IV}	Sc^{III}	Ti^{IV}	Sc^{III}
0.1	0	-	-	>1000	184
0.1	0.05	-	-	>1000	41
0.1	0.1	-	-	>1000	14
0.1	0.15	<< 1	12.0	>1000	5.1
0.1	0.20	<< 1	10.7	>1000	1.7
0.1	0.30	<< 1	7.0	370	0.2
0.1	0.50	<< 1	11.2	105	<< 1
0.1	0.75	~0.5	14.0	-	-
0.1	1.0	<< 1	8.1	17	<< 1
0.025	0	1.0	201	>1000	954
0.025	0.025	1.0	148	>1000	168
0.025	0.050	0.6	129	>1000	40.9
0.025	0.075	1.8	128	>1000	14.2
0.025	0.125	3.3	124	1050	2.68
0.025	0.175	3.1	120	410	0.3
0.025	0.250	2.9	119	290	<< 1
0.005	0	32	7619	>1000	2340
0.005	0.025	30.4	2378	>1000	67.2
0.005	0.0375	34.2	2242	>1000	24.0
0.005	0.05	33.6	2665	>1000	10.9
0.005	0.065	28.2	1872	>1000	4.0
0.005	0.08	33	1715	844	1.27
0.005	0.10	33	1646	688	0.71
0.005	0.125	25.6	1398	457	<< 1
0.005	0.25	-	-	46	<< 1
0.005	0.5	-	-	3.8	<< 1

micro-Curie range, cf. Table **3**. Although these investigations stimulated significant progress in basic radiochemistry, radiopharmaceutical or even medical studies with the correspondingly generated ^{44}Sc have not been possible so far. The latter became recently feasible, with a 5 mCi generator constructed at the Institute of Nuclear Chemistry, Johannes Gutenberg University Mainz, Germany [8] utilizing the ^{45}Sc(p,2n)^{44}Ti process. A metallic Sc target was developed to withstand high proton beam fluxes. Sc was melted in 150 to 220 μm thick layers onto copper backings of sophisticated structure for optimum heat transfer. Initial irradiations have been performed for a 150 μm Sc layer on 150 μA internal proton beam (E_p≈25 MeV) for 1 x 10 h and 1 x 5 h (2250 μAh), yielding 4.56 MBq ^{44}Ti corresponding to a production yield of 2 kBq/μAh. The developed target system was capable to withstand long-term irradiations at up to 200 μA. However, ^{65}Zn was co-produced at significant activities of 232 MBq. In order to reduce the ^{65}Zn contamination, new targets were developed with increased Sc layer thickness and intermediate layers of Ag. Finally, a 1.5 g Sc target was irradiated at 200 μA producing about 185 MBq (5 mCi) of ^{44}Ti.

Separation of ^{44}Ti and ^{44}Sc

^{44}Ti was separated from 1.5 g of massive scandium targets in multi-step procedures. In order to follow the separation of ^{44}Ti from the irradiated scandium target, and also for the determination of Sc^{III} distribution coefficients (see below), ^{46}Sc ($t_{½}$ = 83.79 d) was applied as a tracer. The irradiated scandium was dissolved in 18 mL of 2 M HCl transferred to cation exchange chromatography. A large column (H = 350 mm, S = 2 cm^2, V_o = 35 mL) of AG 50W-X8, 200-400 mesh (H^+-form) was eluted with 45 mL 1 M HCl, 30 mL 2 M HCl, 160 mL 3 M HCl, 200 mL 0.5 M oxalic acid

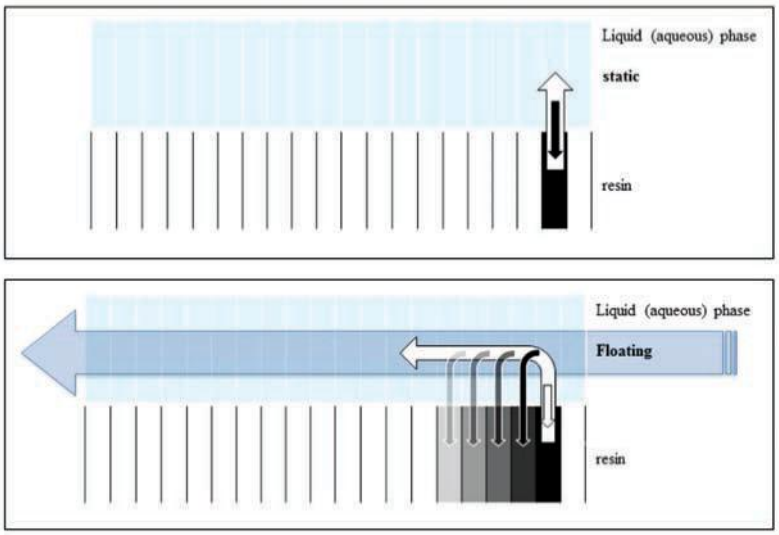

Fig. (2). Principal effects for solid Phase-based Ion Exchange Processes in the case of static conditions (Upper Type) and Dynamic Conditions (Lower Type). Parent Radionuclides Desorbed under Floating (Elution) Situation may Migrate and Re-adsorb at Resin Capacities Located Downstream. With many Subsequent Elutions and / or Elutions Performed at Larger Eluent Volumes, the Maximum of the Parent Radionuclide (Adsorption Zone) Moves Gradually.

($H_2C_2O_4$) consecutively. About 99.9% of the ^{44}Ti produced in the Sc(p,2n) nuclear reaction were finally isolated in a 48 mL fraction of 2 M HCl. A second chromatography provided a more complete separation with a separation factor of about 10^5, i.e. less than 10^{-3}% (about 15 μg) of the initial scandium still remaining in the ^{44}Ti fraction. In total, about 99.6% of the ^{44}Ti activities have been recovered following cation exchange purification of the no-carrier-added radionuclide from about 1.5 g of a macroscopic scandium target [8].

^{44}Ti/^{44}Sc RADIONUCLIDE GENERATOR DESIGN

For preparation of ^{44}Ti/^{44}Sc radionuclide generators, several radiochemical criteria are relevant, such as effective separation strategies providing high ^{44}Sc elution yields and low ^{44}Ti breakthrough. In addition, the type of Sc eluate should be carefully considered and ideally adopted to those solvent characteristics useful for subsequent labeling reactions (i.e. low volume, low pH, high purity etc.), cf. [1].

HCl / Oxalic Acid System

Classically, Sc^{III} is strongly adsorbed from oxalic acid solution, and its oxalate complex is selectively destroyed by the addition of hydrochloric acid. These properties can be used as the basis of a method for the anion exchange separation of Sc^{III} and Ti^{IV} in hydrochloric acid / oxalic acid mixtures [16]. Thus, the determination of distribution coefficients of Ti^{IV} and Sc^{III} on ion exchange resins is essential. Results of the K_d values obtained in [8] for the two different ion exchange resins and the various mixtures are shown in Table **3**. Accordingly, optimum conditions for efficient separations and for the design of generators could be to elute AG 1-X8 resins with 0.2 M HCl / 0.1 M $H_2C_2O_4$, 0.125 M HCl / 0.025 M $H_2C_2O_4$ or 0.06-0.08 M HCl / 0.005 M $H_2C_2O_4$ mixtures. To avoid the breakthrough of ^{44}Ti even at very high numbers of elutions it appeared to be favorable to use mixtures of lower concentration, namely 0.06-0.08 M HCl / 0.005 M $H_2C_2O_4$.

Consequently, a 5 mCi generator was prepared on a column made of PEEK (H = 150 mm, D = 3 mm, V_o = 0.55 mL) and filled with resin AG 1-X8 (200-400 mesh, Br^--form) [8]. Purified ^{44}Ti was dried and dissolved in 20 mL 0.1 M $H_2C_2O_4$. This solution was loaded to the generator and the generator was washed with 0.005 M $H_2C_2O_4$ / 0.07 M HCl mixture. The system achieved elution of 180 MBq ^{44}Sc in 20 mL of eluate solution (0.07 M HCl and 0.005 M $H_2C_2O_4$). The breakthrough of ^{44}Ti was 90 Bq. This corresponds to an excellent separation factor of $2 \cdot 10^6$.

^{44}Ti Breakthrough / Reverse-Type Elution Mode

In contrast to more common radionuclide generators utilized in daily nuclear medicine practice (such as e.g. the ^{99}Mo/^{99m}Tc system), the separation parameters should remain the same for a long period. While the ^{99}Mo/^{99m}Tc radionuclide generator is eluted for about one week only, the ^{44}Ti/^{44}Sc radionuclide generator is supposed to work for many years. Thus, high long-term stability becomes a relevant issue.

There are only a few reports on studies to develop ^{44}Ti/^{44}Sc radionuclide generators. Using 0.1 M $H_2C_2O_4$ / 0.2 M HCl mixture on Dowex-1 resin, 60-70% elution yield of ^{44}Sc in 30-50 mL was reported [4]. A solvent extraction technique with an organic phase of 1% 1-phenyl-3-methyl-4-capryl-pyrazolone-5 in methyl isobutyl led to >90% recovery of Sc in less than 10 mL with a Ti contamination of $< 10^{-6}$ [5]. Elution yields of 42-46% and decontamination factor of $5 \cdot 10^4$ were reported in studies with 0.01 M HCl as an eluent and ^{44}Ti being adsorbed on inorganic ZrO_2 as an analogue of Ti^{IV} [6], cf. also Table **3**.

In fact, typical medical radionuclide generators rely on solid phase-based ion exchange processes. Distribution volumes are determined in batch studies, i.e. under rather static conditions. However, real generator elutions reflect dynamic systems. Fig. (**2**) illustrates the principal effects of elutions performed many times and / or at larger eluent volumes. If the ^{99}Mo/^{99m}Tc radionuclide generator is eluted for about one

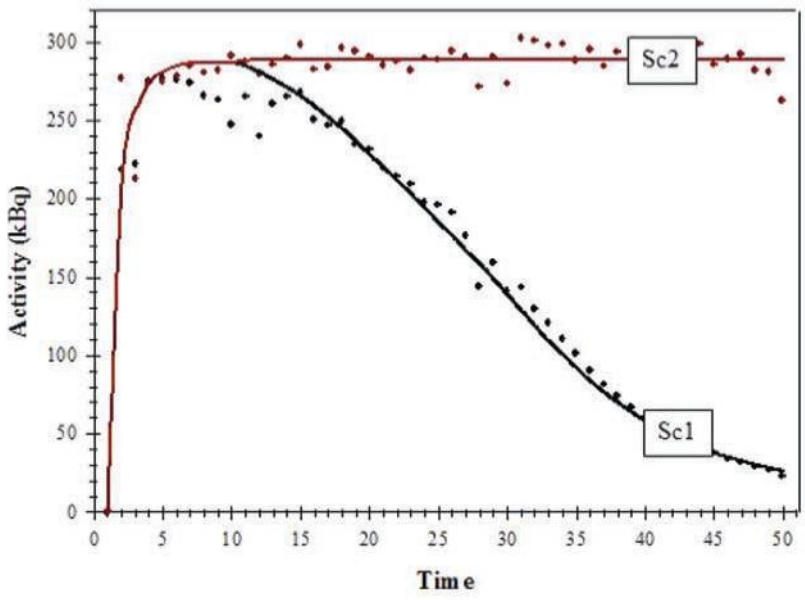

Fig. (3). Yield of ^{44}Sc for Increasing Number of Elutions for "direct" (Sc1) and "reverse" (Sc2) Elution Modes after 50 Elutions [8].

week with e.g. 5 elutions totally, each elution of 10 mL, the overall elution volume is 50 mL. In the case of the ^{44}Ti/^{44}Sc radionuclide generator, a one-elution volume is 20 mL. Supposed the generator is eluted every working day of only one year, which gives about 250 elutions, the overall elution volume is 5000 mL – a factor of 100 difference between the two different radionuclide generators. The difference becomes even more pronounced in the case of e.g. 5 years usage of the ^{44}Ti/^{44}Sc system. Interestingly, the ^{68}Ge/^{68}Ga radionuclide generator is similar. With its secular equilibrium characteristics like the ^{44}Ti/^{44}Sc system, and up to three elutions each day with e.g. 10 mL 0.1 M HCl, and e.g. 200 days of usage, these altogether 600 elutions make up a total elution volume of 6000 mL for the ^{68}Ge/^{68}Ga system.

In the context of long-term stability of ^{44}Ti/^{44}Sc generators, a "reverse" type of washing steps after each elution using 0.07 M HCl / 0.005 M $H_2C_2O_4$ mixtures thus appeared to be essential.

To demonstrate the effect, two smaller columns made of PEEK (diameter 3 mm, length 40 mm) were filled with AG 1-X8, 200-400 mesh, in Br^--form. Both generators were eluted using 10 mL of 0.1 M $H_2C_2O_4$ / 0.2 M HCl solutions. In this case, the somewhat lower K_d values for Ti^{IV}, cf. Table **3**, may allow following the eventual breakthrough of ^{44}Ti at a limited number of elutions already.

While generator G1 was eluted in a standard procedure, i.e. in a single direction ("direct"), the generator G2 was in addition "regenerated" after each elution using 0.1 M $H_2C_2O_4$ / 0.2 M HCl applied in alternating direction ("reverse"). Elution of both generators was carried out 3 times a week. Fig. (**3**) illustrates the yield of ^{44}Sc (Sc1 for G1, Sc2 for G2) obtained for the increasing number of elutions for both generator types G1 and G2. After about 15 elutions, the activity of ^{44}Sc eluted started to drop for G1, which is due to the increasing breakthrough of ^{44}Ti. In contrast, the "reverse" elution protocol applied to generator G2, showed a constant yield of ^{44}Sc elutions (Sc2) for the complete 50 elution runs applied. In conclusion, this type of generator design guarantees an irrelevant breakthrough of ^{44}Ti and an almost constant level of ^{44}Sc elution.

This elution profile corresponds with the breakthrough of ^{44}Ti as shown in Fig. (**4**). The "direct" elution strategy of pilot generator G1 results in an increasing breakthrough of ^{44}Ti, which results in a 50% desorption of ^{44}Ti after about 30 elutions, and an almost complete release of ^{44}Ti after 50 elutions. In contrast, the breakthrough of ^{44}Ti in the case of the "reverse" type elution scheme is negligible for the first 10 elutions, and is increasing only slightly in the following 40 elutions. The maximum breakthrough of ^{44}Ti is about 0.2%.

Improvement (Regeneration) of ^{44}Ti Distribution Profiles on the Column

"Reverse" elution modes obviously provide high retention of ^{44}Ti on the column. However, the distribution of ^{44}Ti is still changing with increasing elutions. Thus, a possibility to improve the distribution profile by "reverse" elution with different composition of $H_2C_2O_4$ / HCl solutions was tested.

Elution of the generator G2 was carried out 3 times a week as described earlier. After 50 elutions, the ^{44}Ti distribution profile was analyzed using μPET registration. Subsequently, the generator was washed using 4 mL of 0.1 M $H_2C_2O_4$ and 2 mL of 0.1 M $H_2C_2O_4$ / 1 M HCl mixture and again 4 mL of 0.1 M $H_2C_2O_4$ consecutively in "reverse" direction. This operation was repeated 5 times. ^{44}Ti distribution profiles on the column of the pilot generator G2 after 50 elutions (a) and after regeneration with using 0.1 M $H_2C_2O_4$ and 0.1 M $H_2C_2O_4$ / 1 M HCl mixture solutions (b) are compared graphically in Fig. (**5**) as analyzed by μPET.

The coronar distribution image of ^{44}Ti(^{44}Sc) on the pilot generator G2, as analyzed by μPET imaging, is illustrated in Fig. (**6**) for the "reverse" generator type G2 after 50 elutions before and after the "regeneration" procedure. The zone of ^{44}Ti became narrower (Fig. **6b**).

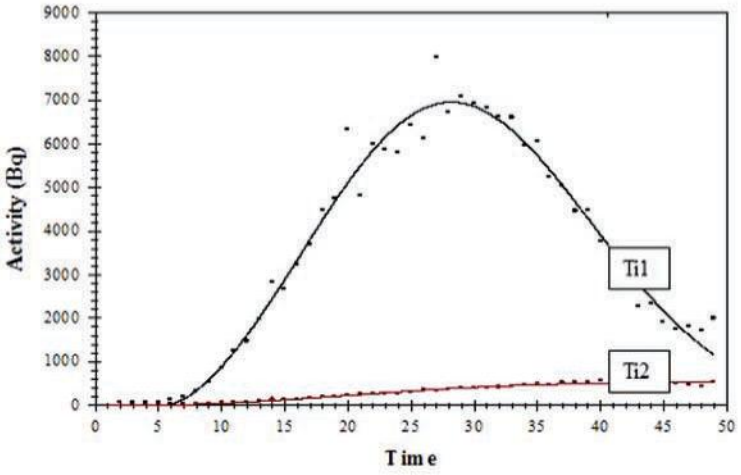

Fig. (4). Breakthrough of ^{44}Ti for Increasing Number of Elutions for "Direct" (Ti1) and "Reverse" (Ti2) Elution Modes after 50 Elutions [8].

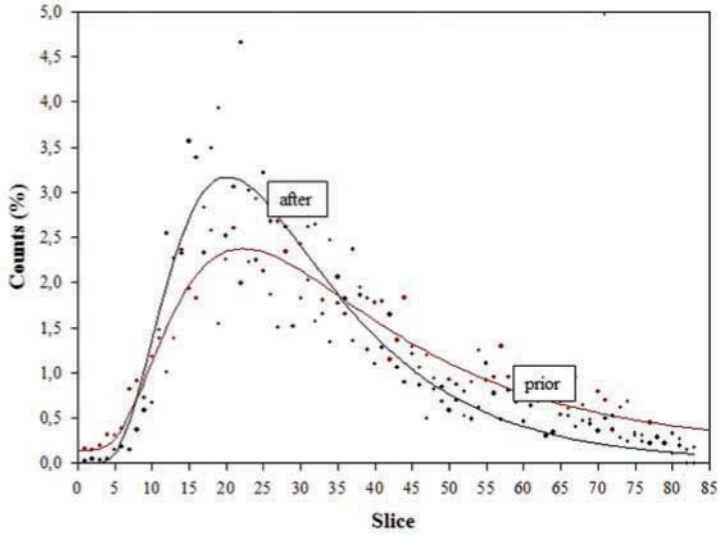

Fig. (5). Distribution Profile in Percent for ^{44}Ti Prior and after "Regeneration" as Measured by μPET [8]. Each Slice unit is about 0.5 mm.

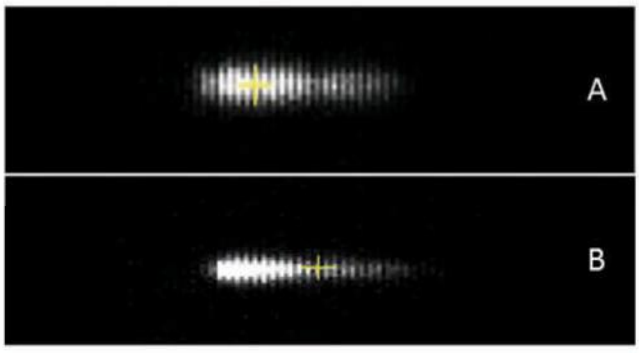

Fig. (6a, b). Distribution images for ^{44}Ti Prior (**A**) and after (**B**) "Regeneration" as Measured by μPET [8]. Quantitative Data on the Annihilation of ^{44}Ti/^{44}Sc of the Generator Column. The Improvement of the Distribution is Obvious.

Preparation of the "Reverse" 5 mCi Prototype ^{44}Ti / ^{44}Sc Radionuclide Generator

Typical ^{44}Ti / ^{44}Sc radionuclide generator elution profiles of ^{44}Sc are shown in Fig. (**7**) for the initial experiments (mean data for the elutions 4, 5, 6 and 7) as well as a typical result for an elution performed after one year (elution number 54). The corresponding activities of ^{44}Ti in the individual fractions are given in Fig. (**8**) and indicate an improvement of breakthrough with increasing number of elutions. Aliquots 4-7 contain 85±2% of the total ^{44}Sc activity. In all aliquots the content of ^{44}Ti is less than 10^{-4}% (150 Bq).

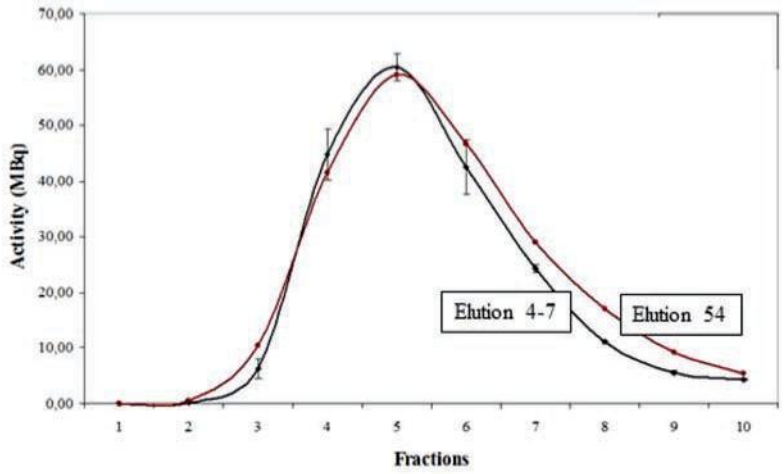

Fig. (7). Elution Profile of ^{44}Sc (Mean of Elutions 4-7) and 54 (after 1 year). Curie-meter measurements. Each fraction contains 2 mL [8].

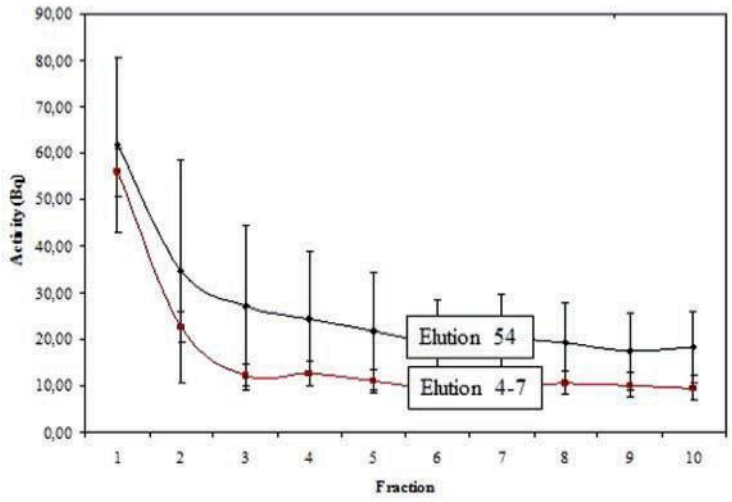

Fig. (8). Breakthrough of ^{44}Ti (Mean for the Elutions 4-7) and 54 (after 1 year). γ-spectroscopy. Each fraction contains 2 mL [7].

Final Apparative Scheme of the "Reverse" ^{44}Ti/^{44}Sc-Generator

A modular system of the 5 mCi generator in Fig. (**9**) presents the central generator column in a horizontal position (II). Two reservoirs for the eluate solutions are connected to the inlet (I) and the outlet (III) position of the generator column. The reservoirs II and III are connected to an air pressure *via* filter F to avoid contaminations of eluate composition with metals from the air. Transfer of eluates from the reservoirs through the generator is achieved by air pressure (elution of ^{44}Sc) or vacuum ("reverse" elution) using manually the empty syringe S. All parts of the prototype ^{44}Ti/^{44}Sc-generator are connected *via* tubing and 3-way valves. The generator works in a "reverse" scheme of elutions. The initial elution is organized by transferring 20 mL of the eluate solution of reservoir (I) through the generator into the ^{44}Sc vial (IV). After each elution the generator is eluted with the same eluate composition in a reverse way using reservoir (III). While the eluate in reservoir (III) is refreshed routinely, the eluate in bottle (I) can be used for next elution of the generator. Eluted ^{44}Sc in collecting vial (IV) can be used for further experiments. The scheme guarantees for safe handling, as it represents an inherently closed system with respect to ^{44}Ti.

Using optimum K_d values of Ti^{IV} and Sc^{III} for $HCl/H_2C_2O_4$ mixtures, i.e. 0.2 M HCl / 0.1 M $H_2C_2O_4$ and 0.07 M HCl / 0.005 M $H_2C_2O_4$, and after several years of regular elution of 5 mCi ^{44}Ti/^{44}Sc radionuclide generator, the yield of ^{44}Sc and ^{44}Ti is stable and the breakthrough of ^{44}Ti is very low. The system achieves elution of 97% (180 MBq) ^{44}Sc in 20 mL of eluate solution. The breakthrough of ^{44}Ti is $5 \cdot 10^{-5}$% (90 Bq). This corresponds to an excellent separation factor of $2 \cdot 10^{6}$. Compared to other ^{44}Ti/^{44}Sc radionuclide generators, higher elution yields of ^{44}Sc in less volume and a higher separation factor were achieved, also higher ^{44}Ti activity compared to only kBq in other studies (Table **4**). Recently, the thermal release of ^{44}Sc from titanium metal [12] was investigated. Although the nuclear reaction route ^{46}Ti(n,3n)^{44}Ti produces ^{44}Ti, absolute ^{44}Ti production yields have not been mentioned and were most probably rather at keV level only. Nevertheless, the thermal release of scandium isotopes is quantitative in vacuum at temperatures at > 1400°C and within 1 hour heating period. Whether this approach is realistic in regard to the half-life of ^{44}Sc and in the

Table 4. Comparison of Different ^{44}Ti/^{44}Sc Radionuclide Generators

Year of Studies	Activity (MBq)	Yield of Sc (%)	Volume (mL)	Separation Factor
1967 [4]	-	60-70	30-50	$2 \cdot 10^4$
			50	10^3 (after 40 elution)
1979 [5]	-	-	-	-
1973 [6]	0.037	42-46	30	$5 \cdot 10^4$
2009 [8]	185	97	20	$2 \cdot 10^6$

case large batch activities of ^{44}Sc are needed remains to be demonstrated.

POST-ELUTION PROCESSING OF ^{44}Ti/^{44}Sc GENERATOR-DERIVED ^{44}Sc FOR MEDICAL APPLICATION

The 5 mCi ^{44}Ti/^{44}Sc radionuclide generator design provides [8] stable high-purity elution of significant activities of ^{44}Sc of 180 MBq per elution. In the context of medical applications of ^{44}Sc eluted from the present generator system, its absolute activities are sufficient for initial studies. By fractionating the ^{44}Sc eluate, it is possible to obtain approximately 85% of the available activity in a volume of 8 mL of the eluate 0.005 M $H_2C_2O_4$ / 0.07 M HCl. Even this volume appears still too large and the content of hydrochloric acid too high for labeling of e.g. nanomoles of peptides for application in nuclear medicine. Therefore, an efficient post-elution processing with concentration of ^{44}Sc eluate on the cation-exchange resin was developed, paralleling the approach of post-processing of ^{68}Ge/^{68}Ga generators [17, 18]. The concept consists in (i) identifying an absorber material positioned at the initial generator outlet to online fix the ^{44}Sc, and (ii) to utilize a small volume of a solution to quantitatively elute ^{44}Sc for subsequent labeling purposes.

^{44}Sc Adsorption on Line

The first step of pre-concentration studies utilized cation-exchange resins to adsorb ^{44}Sc from the generator eluate of 0.005 M $H_2C_2O_4$ / 0.07 M HCl composition. The results of optimization studies concerning the capabilities of different cation-exchange resins are presented in detail in [19]. The lowest retention of ^{44}Sc (42%) was observed when Chelex 100 (200-400 mesh, Na^+-form) resin, containing iminodiacetate ions coupled to a styrene divinylbenzene support, was utilized. Use of the strong cation exchange resin AG 50W-X4 (200-400 mesh, H^+-form) with sulfonate groups on the styrene divinylbenzene matrix increased the retention of ^{44}Sc on the columns up to 51%. Utilization of the AG 50W-X8 (200-400 mesh, H^+-form) based on the same matrix with the same functional groups like AG 50W-X4 (200-400mesh, H^+-form), but differing in the cross-linkages value, resulted in 89% retention of ^{44}Sc on the resin. Therefore, this resin was chosen for subsequent studies.

^{44}Sc Desorption

Several solutions were tested systematically for the elution of ^{44}Sc from the cation exchange column. While application of only 1 mL 0.1 M diammonium oxalate gave the best ^{44}Sc recovery (95%), however labeling of DOTATOC with ^{44}Sc as eluted by 1 mL of 0.1 M diammonium oxalate and added to 4 mL HEPES buffer (pH = 4.0) containing the peptide failed. Further studies were focused on ammonium acetate solutions, despite the fact that the recovery of ^{44}Sc by 2 mL of the utilized mixture was slightly lower than with oxalates [20, 21]. High recovery of ^{44}Sc (~90%) was obtained, when 3 mL of a 0.25 M ammonium acetate buffer acidified to pH = 4.0 by addition of acetic acid was used.

Combined Protocol: Elution of the ^{44}Ti/^{44}Sc Generator and Post-Processing

The built up of an on-line module system of generator post-processing is illustrated in Fig. (**9**). In addition to the generator itself, the miniaturized chromatography column (V) that was prepared from two 3-way valves (I and II), filled with 53 mg of Bio-Rad AG 50W-X8 (200-400 mesh, H^+-form) resin is connected to the ^{44}Ti/^{44}Sc generator *via* tubing. The 20 mL of a 0.005 M $H_2C_2O_4$ / 0.07 M HCl solution passes the ^{44}Ti/^{44}Sc generator with a flow rate of 1 mL/min by using syringe (S) and the eluted ^{44}Sc adsorbs on-line on the small cation exchange column. The ^{44}Sc retains on the column, whereas the generator eluate continues to the waste vial (valve IV). By using a standard single-use syringe the column with cation-exchange resin is washed by 2–4 mL of H_2O to remove the remaining traces of the initial eluate solution, which are collected in the waste vial as well. Finally, 5 mL air is blown through the column.

In the following step, the 3 mL of 0.25 M ammonium acetate buffer, pH = 4.0, are slowly (0.7 mL/min) pressed through the column with a 2 minute break for 1 mL. Finally, air is passed through the column (V) to remove traces of the ammonium acetate buffer solution remaining in the dead volume of the column. ^{44}Sc is collected in an 11 mL glass reacting vial (VII). Reconditioning of the cation exchange cartridge is performed by washing with 1 mL of 4 M HCl and finally by 1 mL H_2O. In the first elution step, >98% of cationic ^{44}Sc retains on the column, whereas most of the ^{44}Ti content of the initial generator eluate (~80%) passes the column and is transferred to the waste. Application of 3 mL 0.25 M ammonium acetate (pH = 4.0) recovered ~90% of ^{44}Sc. The obtained ^{44}Sc solution in the acetate buffer was ready for labeling chemistry. The amount of ^{44}Ti in the final ^{44}Sc fraction was less than 7 Bq. The initial breakthrough of 5×10^{-5}% was thus further reduced by a factor of 10. This represents an approach to further remove the amount of co-eluted ^{44}Ti breakthrough. The final content of ^{44}Ti in the 140-160 MBq ^{44}Sc fraction ready for labeling is thus 7 Bq,

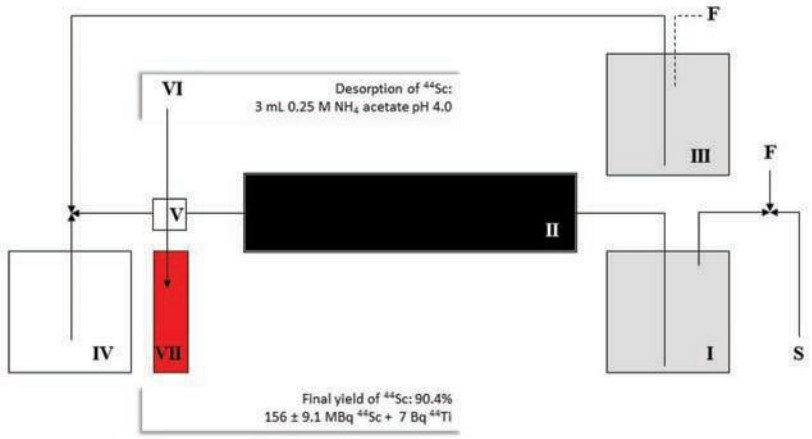

Fig. (9). Scheme of the Post-elution Processing of ^{44}Sc-eluates.

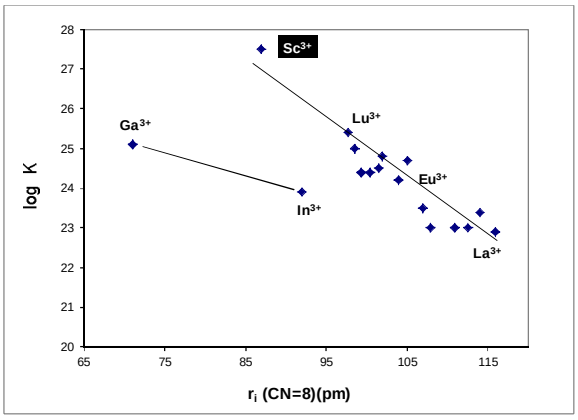

Fig. (10a). Me^{III}-DOTA Complex Formation Data Presented as logK Values Depending on the Ionic Radii of Various Trivalent Metals [23].

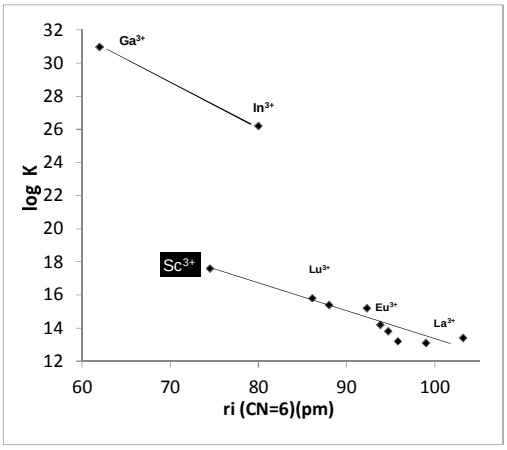

Fig. (10b). Me^{III}-NOTA Complex Formation Data Presented as logK Values Depending on the Ionic Radii of Various Trivalent Metals [23].

representing an astonishingly very low contamination of around $<2 \times 10^{-7}$.

SYNTHESES OF ^{44}Sc RADIOPHARMACEUTICALS

The post-elution processing of volumes and impurities is easily compatible with the synthesis of ^{44}Sc-labeled compounds. A chemically and radiochemically highly pure ^{44}Sc fraction of very high radioactivity concentration of around 50 MBq/mL is obtained, representing 150 MBq overall activity. This for the first time allows for systematic follow-up research towards medical application of ^{44}Sc. $^{44}Sc^{III}$, in principle, is a metallic cation suitable for complexation with many chelators conjugated to peptides or other molecular targeting vectors. However, a key question is to identify the most adequate chelator.

Me^{III}-DOTA Complex Formation

The thermodynamic stability constants (logK) of the Me^{III}-DOTA complexes formed (DOTA = 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid) are several orders of magnitude higher than in the case of open-chain analogue chelators, like EDTA (ethylenediaminetetraacetic acid) or DTPA (diethylene triamine pentaacetic acid). For example, the stability constants of Gd^{III}-DOTA and Gd^{III}-DTPA are 25.8 and 22.1, respectively. For Sc^{III}, the stability constants for DOTA and DTPA are logK = 27.0 and 20.99, respectively [24-26]. Recently, Sc^{III} complex formation constants have been compared with other non-radioactive and radioactive Me^{III}-metals, which work for medical imaging, such as Gd for Magnetic Resonance Tomography (MRT), ^{111}In and ^{68}Ga for PET, ^{90}Y and ^{177}Lu for therapy. Those are DOTA and eventually NOTA and their derivatives. Fig. (**10**) summarizes complex formation data obtained for NOTA and DOTA and presents logK values depending on the ionic radii of various trivalent metals [23]. Sc^{III}-DOTA complexes show very high thermodynamic stability of logK = 27.5 and are thus even more stable than Lu^{III} or Ga^{III} analogues. For NOTA (1,4,7-triazacyclononane-1,4,7-tetraacetic acid), the trend is opposite: Sc-NOTA got a logK of 14.8 only. Supposed the kinetic stability is similarly high for Sc-DOTA and Sc-NOTA complexes (as known for Ga-analogues), DOTA is the macrocyclic ligand of choice for ^{44}Sc-labeled radiopharmaceuticals rather than NOTA. However, systematic research may reveal other ligand structures more adequate.

Me^{III}-DOTA-peptide Synthesis and Radiopharmaceutical Stability

As a proof-of-principle, the potential of ^{44}Sc for labeling DOTA-conjugated peptides was investigated. DOTA-D-

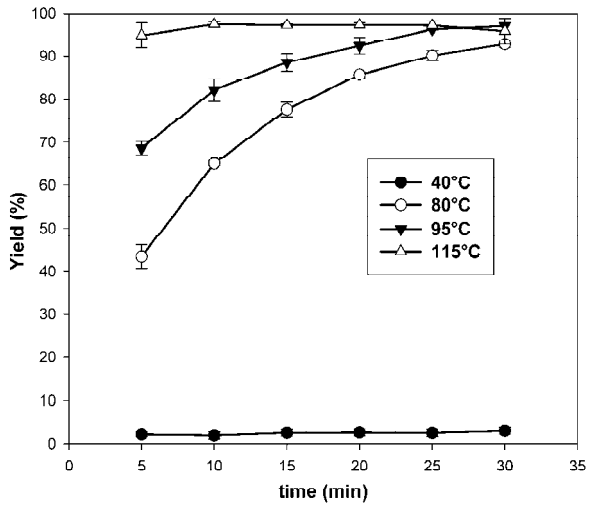

Fig. (11). Formation of ^{44}Sc-DOTA-TOC at 21 nmol DOTA-TOC Concentration, 150 MBq ^{44}Sc, pH 4.0 as a Function of Heating at Different Temperatures [25].

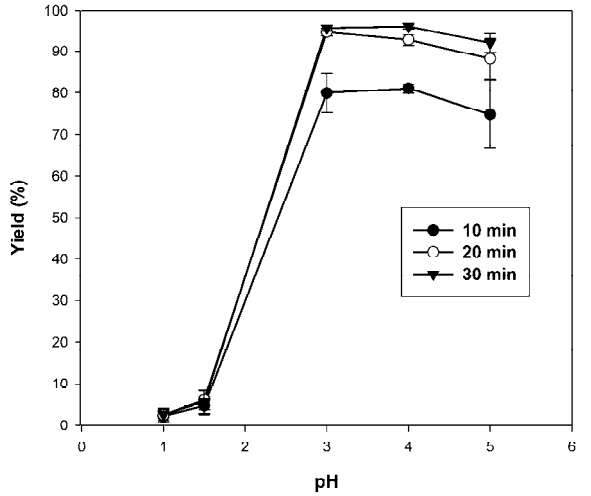

Fig. (12). Formation of ^{44}Sc-DOTA-TOC as a Function of pH (21 nmol of DOTA-TOC, T = 95ºC) [25].

Phe^1-Tyr^3-octreotide (DOTA-TOC) and DOTA-D-Phe^1-Tyr^3-octreotate (DOTA-TATE) were used as model molecules to study and optimize the labeling procedure [25]. Reaction parameters such as buffer conditions, concentration of peptide, pH range, reaction temperature and time were optimized. Using 21 nmol of DOTA-TOC in 3 mL of post-processed ^{44}Sc eluate in ammonium acetate buffer pH = 4.0 provided labeling yields >98% within 25 minutes of heating in an oil-bath at 95ºC. This time can be reduced to only 3 minutes by applying microwave-supported heating. The influence of temperature on formation of ^{44}Sc-DOTA-TOC conjugate is shown in Fig. (**11**) for 21 nmol DOTA-TOC concentration. Variable radiochemical yields were obtained if ~14 nmol of DOTA-TOC were used. At 7 nmol of DOTA-TOC the labeling yield was less than 20%.

Labeling of DOTA and DOTA-conjugated derivatives with medically useful metallic radionuclides is generally performed at pH = 2–6. For ^{44}Sc the highest reaction yield was obtained, at pH between 3 and 5 (Fig. **12**). This study confirmed, that the previously developed method of post-elution processing of ^{44}Sc generator eluate gave not only the best results of radionuclide desorption from miniaturized column, but also that the obtained 2–3 mL ^{44}Sc fraction in acetate buffer (pH = 4.0) is simultaneously adequate for direct labeling of DOTA-conjugated molecular tracers without a necessity of further changing its pH [25].

Purification of ^{44}Sc-DOTA-octreotides

In both cases of ^{44}Sc-DOTA-TOC formations, i.e. conventional and microwave heating, the radiochemical purity was >98% and additional purification of the product was not necessary. However, in case of lower yields or studies that require removing of acetate ions, the purification can easily be performed on a reverse-phase (RP) C-18 column. The solution of ^{44}Sc-DOTA-TOC was passed through an equilibrated small C-18 cartridge and conjugate was recovered in 400 μL fraction of pure ethanol with ~94% efficacy and containing <0.9% free ^{44}Sc. Thus, a radiochemically pure product, not depending on the initial radiolabeling yield, is guaranteed. The ethanol from final fraction can be either evaporated by flushing a stream of nitrogen at room temperature for 10–15 minutes or directly added to 0.9% NaCl. Following sterilization by filtration through a 0.22 μm membrane, this ^{44}Sc-DOTA-peptide solution is available for biological or medical studies.

Stability of ^{44}Sc-DOTA-octreotide

Different metals cations present in the reaction mixture may compete strongly with medically useful radionuclides while labeling with DOTA or DOTA-conjugated derivatives. Amounts of some metallic cations can cause transmetallation of radionuclide-conjugates and finally induce a release of the free radionuclide into solution. Therefore, the stability of ^{44}Sc-DOTA-TOC was analyzed in presence of relevant metal cations. The addition of Fe^{3+}, Cu^{2+}, Ca^{2+} and Mg^{2+} did not induce any removal of ^{44}Sc from the conjugate. The ^{44}Sc-DOTA-TOC was stable even after 25 h incubation at 37ºC with metal cations at rather high concentration levels of 10^{-2} M [25]. Similarly, the stability of ^{44}Sc-DOTA-TOC in the presence of other complexing agents, like EDTA and DTPA was investigated to check if ligand-ligand substitution occurs. This was not the case; the experimental studies confirmed high stability of ^{44}Sc-DOTA-TOC even after 25 h incubation at 37ºC with EDTA or DTPA ligands at molar ratios of competing ligand to DOTA-peptide equal to 1:1, 10:1 and 100:1 [25]. ^{44}Sc-DOTA-TOC in pure ethanol fraction at room temperature and 37ºC is stable for up to 7 h. Stability of ^{44}Sc-DOTA-TOC was analyzed also in 0.9% NaCl and in PBS (pH = 7.4), indicating high stability of the formed conjugate even after 22 h incubation at 37ºC [25].

BIOLOGICAL EVALUATION OF ^{44}Sc-LABELLED RADIOPHARMACEUTICALS *IN VITRO*

Binding Affinities

^{44}Sc-DOTA-peptides are supposed to target transmembrane G-protein coupled tumor receptors. Although the targeting molecule (the peptide) and the bifunctional chelator (DOTA) are identical, the choice of the trivalent metal is known to influence binding affinities. For ^{44}Sc-analogues,

Table 5. Binding Affinities of Me^{III}-DOTA-complexes of Octreotide and Bombesin Derivatives DOTA-TOC and DOTA-BN[2-14]NH_2, Respectively

Me^{III}	IC_{50} (nM)		
	DOTA-TOC		DOTA-BN[2-14]NH_2
	Displacement of ^{125}I-[Tyr^{11}]-SST-14, AR42J cells [24]		Displacement of ^{125}I-[Tyr^4]-BN, PC-3 cells [25]
	Tyr^{11}-SST-14	Me^{III}-DOTA-TATE	Me^{III}-DOTA-BN[2-14]NH_2
-	0.67±0.12	0.64±0.22	1.78 ± 0.12
Ga		0.20±0.20	0.85 ± 0.06
Sc		0.70±0.18	6.49 ± 0.13
Lu			1.34 ± 0.11
Y			1.99 ± 0.06

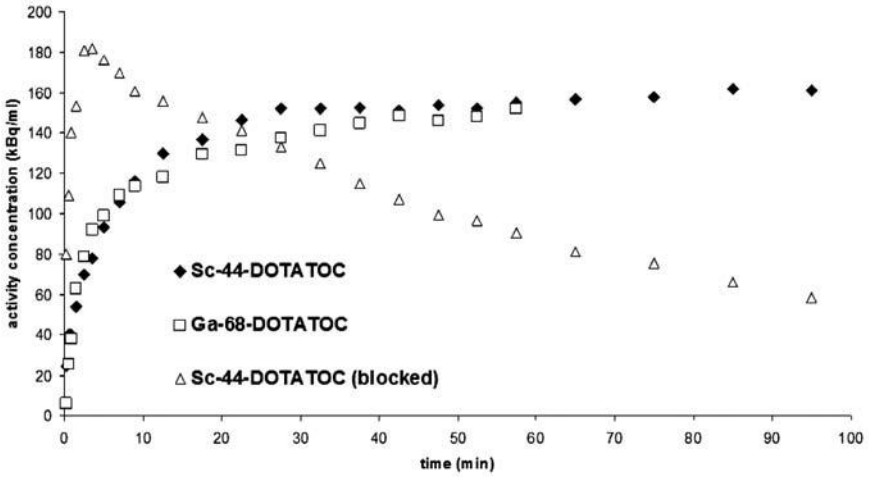

Fig. (13). Tumor Uptake Kinetics of ^{44}Sc-DOTA-TOC in Tumor Bearing Mice Compared to Blockade and ^{68}Ga-DOTA-TOC [28].

data have been determined for octreotide and bombesin derivates so far [26, 27]. For octreotide targeting vectors, IC_{50} values of Sc-DOTA-TOC and Ga-DOTA-TATE are 0.7 nM and 0.2 nM. These values are quite similar, and the Sc-analogue parallels the IC_{50} value of the lead compound Tyr^{11}-SST-14. The *in vitro* receptor affinity of Sc-DOTA-BN[2-14]NH_2 is IC_{50} = 6.49 ± 0.13 nM vs. IC_{50} = 0.85 ± 0.13 for Ga-DOTA-BN[2-14]NH_2. ^{44}Sc analogues seem to be somewhat lower in terms of binding affinities, but stay within the same order of magnitude with known derivatives. Both internalization and efflux of ^{68}Ga- and ^{44}Sc-DOTA-BN[2-14]NH_2 was investigated in the same cell line. The internalization rate of ^{44}Sc-DOTA-BN[2-14]NH_2 was faster than for the ^{68}Ga-labelled analogue while the percentage of internalization at the end of incubation was at a comparable level. ^{68}Ga-DOTA-BN[2-14]NH_2 was externalized faster than ^{44}Sc-DOTA-BN[2-14]NH_2 [27].

BIOLOGICAL EVALUATION OF ^{44}Sc-LABELLED RADIOPHARMACEUTICALS *IN VIVO*

Organ uptake *ex vivo* and uptake kinetics *in vivo* of ^{44}Sc tracers have been investigated for DOTA-octreotide and DOTA-bombesin derivatives as well as for DOTA-puromycin derivatives [27, 28, 29].

^{44}Sc-DOTA-TOC

Tumor uptake kinetics was measured in Nude mice with subcutaneous xenograph (AR42J cells). 3-7 MBq of ^{44}Sc-DOTA-TOC were injected and distributions with and without excess cold DOTA-TOC were analyzed by small animal imaging (Focus 120, dynamic and static imaging, reconstruction: OSEM, without attenuation correction, cross calibration with phantom measurements, organ activity determination: manual VOI (PMOD 2.95)). Results shown in Fig. (**13**) indicate specific binding of ^{44}Sc-DOTA-TOC and similar uptake kinetics for ^{68}Ga- and ^{44}Sc-DOTA-TOC. Fig. (**14**) presents small animal PET images for the kidney and tumor region, clearly demonstrating prolonged visualization of the experimental tumors even at 17 hours post injection [28].

^{44}Sc-DOTA-BN[2-14]NH_2

Biodistribution of ^{68}Ga- and ^{44}Sc-DOTA-BN[2-14]NH_2 was investigated in healthy rats. Small animal PET images were assessed in male Copenhagen rats bearing the androgen-independent Dunning R-3327-AT-1 prostatic cancer tumor. The biodistribution studies of both ^{44}Sc-DOTA-BN[2-14]NH_2 and ^{68}Ga-DOTA-BN[2-14]NH_2 in normal rats revealed a specific uptake in target organs and tissues and excretion mainly through urinary tract. µPET images of rat

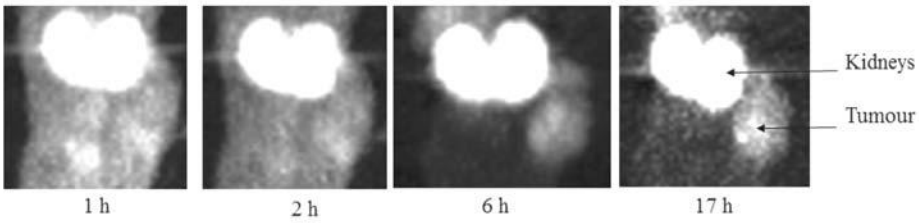

Fig. (14). Small Animal PET Images f of ^{44}Sc-DOTA-TOC in Tumor Bearing Mice for Kidney and Tumor Region, Clearly Demonstrating Prolonged Visualization of the Experimental Tumors even at 17 hours post Injection [28].

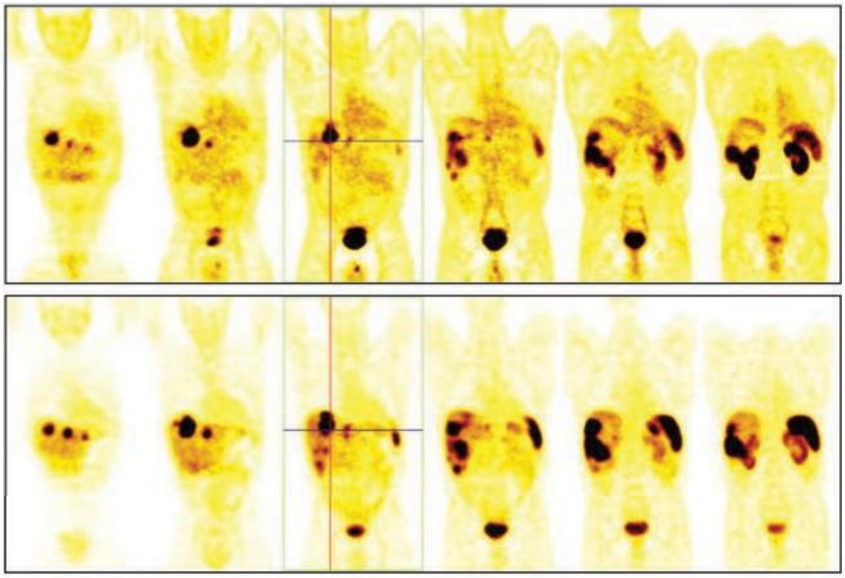

Fig. (15). ^{44}Sc-DOTA-TOC PET/CT imaging (37 MBq Injected) of Somatostatin Receptor Positive Liver Metastases (Department of Nuclear Medicine/PET Center, Zentralklinik Bad Berka, Germany) Comparing ^{44}Sc-DOTA-TOC, 40 min p.i (upper row) vs. ^{68}Ga-DOTA-TATE 90 min. p.i. (lower row) [2].

prostate carcinomas demonstrated that both tracers were accumulated in the tissue with both tracer showing similar distribution patterns. Also the uptake kinetics showed no difference between the ^{44}Sc- and the ^{68}Ga-labeled compound [27].

^{44}Sc-DOTA-puromycin

Puromycin an antibiotic that inhibits protein synthesis by competitive incorporation against an aminoacyl-tRNA on the ribosome A-site. DOTA-Puromycin was synthesized using a puromycin-tethered controlled-pore glas. ^{44}Sc-DOTA-Puromycin was applied to μPET-studies. 20-25 MBq of ^{44}Sc-DOTA-puromycin was administered to tumor bearing rats *via* tail vein and animals were scanned for 1 hour dynamically. Specificity of the imaging was further validated by dissecting the animals after the measurement and *in vitro* blocking experiments using puromycin or cycloheximide to block protein synthesis. μPET images of tumor bearing rats showed significant tumor uptake of ^{44}Sc-DOTA-puromycin and clearly visible tumor outlines. Obtained data from biodistribution was similar to prior biodistribution- and protein-incorporation-studies. In both blocking experiments cellular uptake of ^{44}Sc-DOTA-puromycin could be totally blocked by blocking protein synthesis. This demonstrates direct correlation between cellular uptake of ^{44}Sc-DOTA-puromycin and protein synthesis.

INITIAL HUMAN APPLICATION OF ^{44}Sc-LABELED RADIOPHARMACEUTICALS

As a proof-of-principle, ^{44}Sc-DOTA-TOC was synthesized and somatostatin receptor localization was verified in patient studies at the Department of Nuclear Medicine/PET Center, Zentralklinik Bad Berka, Germany [2]. ^{44}Sc-DOTA-TOC PET/CT imaging of somatostatin receptor positive liver metastases in a patient was clearly successful, even at low activity (37 MBq injected). ^{44}Sc-DOTA-TOC PET at early time-points (40 min p.i.) is comparable to ^{68}Ga-DOTA-TATE (90 min. p.i.) in the same patient, c. Fig. (**15**). The most impressive result is depicted in Fig. (**16**), demonstrating accurate PET imaging at late time-points of 18 hours post injection. The perspective is to measure the uptake kinetics of e.g. ^{44}Sc-DOTA-TOC in metastatic neuroendocrine tumors in order to estimate the optimum radiation dose the individual patient will receive in a subsequent therapeutic application of biologically and chemically analogous compounds, e.g. ^{90}Y- DOTA-TOC or ^{177}Lu-DOTA-TOC.

SUMMARY

^{44}Ti production itself is effective using the ^{nat}Sc(p,2n) reaction. ^{44}Ti/^{44}Sc radionuclide generators based on solid phase-based anion exchange chromatography and dilute hy-

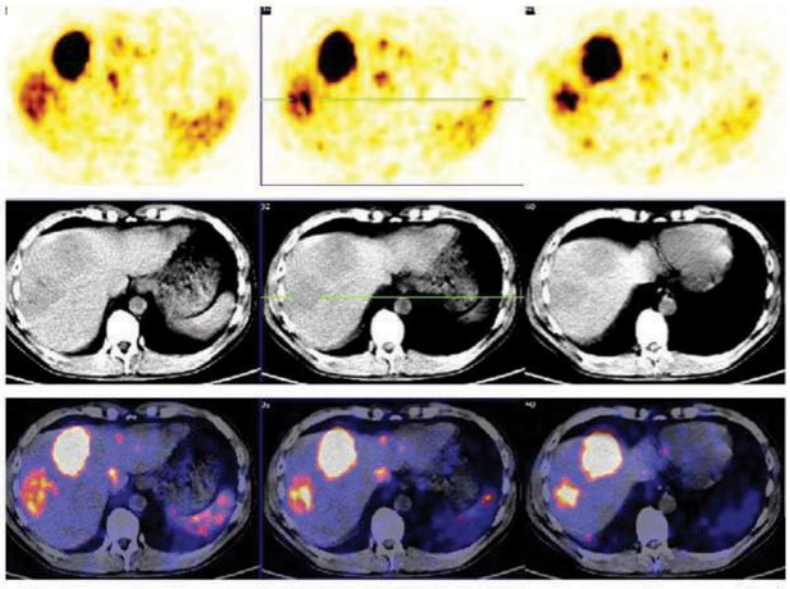

Fig. (16). PET/CT Imaging of Somatostatin Receptor Positive Liver Metastases 18 h after Administration of 37 MBq of ^{44}Sc-DOTA-TOC (Department of Nuclear Medicine/PET Center, Zentralklinik Bad Berka, Germany). ^{44}Sc-DOTA-TOC PET (upper row), CT (middle row), PET / CT fusion (lower row) [2].

drochloric acid / oxalic acid mixtures meet the challenges of robust generator performances. A protocol of reverse generator elution seems to be mandatory to guarantee long-term stability in terms of parent breakthrough. A ca. 186 MBq ^{44}Ti/^{44}Sc generator is in operation since four years and 170 MBq ^{44}Sc are obtained with < 10 Bq of ^{44}Ti.

On-line post-processing based on cation-exchanger purification enables application of the isolated ^{44}Sc fraction for radiopharmaceutical labeling strategies. About 140-160 MBq activity of generator-derived ^{44}Sc-fractions are ready for labeling. Concerning the design of ^{44}Sc-labeled radiopharmaceuticals, the macrocyclic chelators DOTA was identified as promising (bifunctional) ligand with thermodynamic stability constant, even superior to ^{68}Ga.

^{44}Sc-DOTA-conjugated tumor targeting vectors investigated so far are stable *in vitro* and *in vivo*, and reveal pharmacological parameters adequate to long-term (up to one day) molecular imaging. Initial human studies hold promise for a variety of important diagnostic and therapeutic directions.

Areas of future interest are, (i) molecular imaging of a variety of ^{44}Sc-labeled tracers using PET/CT, (ii) potential application of diagnostic ^{44}Sc tracers matching therapeutic applications of analogue compounds labeled with e.g. ^{90}Y or ^{177}Lu, but also with the β^- emitter ^{47}Sc, and (iii) investigating new options of ^{44}Sc molecular imaging by using new PET/3G camera based on β^+/γ decay of radionuclide [30].

CONFLICT OF INTEREST

Declared none.

ACKNOWLEDGEMENTS

The author wishes to thank A. F. Novgorodov, Yu. G. Alenitzky and A. V. Skripnik (Joint Institute of Nuclear Research, DLNP, Dubna, Russian Federation) for helpful discussions. Special acknowledgement goes to VG Kaplun, AG Suzikov and IA Eliseev (Second Central Scientific Research Institute, "V.G. Khlopin Radium Institute", Russian Federation) and D.V. Efremov (Scientific Research Institute of Electrophysical Equipment, Russian Federation), for providing the irradiated Sc target.

Significant developments of the generator, its post-processing and ^{44}Sc-labelling chemistry are due to the great scientific contributions by N. S. Loktionova, M. Fellner and E. Eppard (Institute of Nuclear Chemistry, University of Mainz, Germany), D. V. Filosofov (Joint Institute of Nuclear Research, DLNP, Dubna, Russian Federation), and M. Pruszyński (Institute of Nuclear Chemistry and Technology, Warszawa, Poland).

Special thanks to A. Majkowska-Pilip (Institute of Nuclear Chemistry and Technology, Warszawa, Poland), for complex formations studies, E. Koumarianou and R. Mikolajczak (IAE Radioisotope Centre POLATOM, 05-400 Swierk-Otwock, Poland), S. Eigner and K. Eigner Henke (Radiopharmacy, Nuclear Physics Institute of the Academy of Science of the Czech Republic), O. Thews (now University Halle, Germany), M. Miederer and H.-G. Buchholz (Department of Nuclear Medicine, University Mainz, Germany) for biological evaluation and finally R. P. Baum (Department of Nuclear Medicine/PET Center, Zentralklinik Bad Berka, Germany) for the human application of ^{44}Sc-labeled radiopharmaceuticals.

Financial support by COST actions D38 and BM0607 as well as the DFG is acknowledged (DFG RO 985 / 18).

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Received: December 09, 2011 Revised: March 23, 2012 Accepted: April 05, 2012