CHAPTER 4

DEVELOPMENT OF $^{68}$Ge/$^{68}$Ga GENERATORS

“The important thing in science is not so much to obtain new facts as to discover new ways of thinking about them.”

WILLIAM LAWRENCE BRAGG
4.1 Introduction

4.1.1 Gallium-68 ($^{68}$Ga): An excellent radioisotope for Positron Emission Tomography

$^{68}$Ga is an excellent positron emitting radioisotope suitable for clinical positron emission tomography (PET) applications in Nuclear Medicine [19,142]. It emits positrons with 89% positron branching accompanied by 1.077 keV photon emission of low (3.22%) abundance [15]. The relatively short half-life of $^{68}$Ga ($t_{1/2} = 67.71$ min) permits PET application with suitable $^{68}$Ga radiotracers, while maintaining an acceptable radiation dose to the patient. Moreover, the half-life of $^{68}$Ga matches the pharmacokinetics of many peptides and other small molecules due to rapid diffusion, localization at the target and fast blood clearance [143]. The efficacies of $^{68}$Ga-based tracers are comparable to that of $^{18}$F-based agents and have stimulated researchers to investigate the potential of $^{68}$Ga based PET imaging agents [19,142,144]. Apart from being a PET radionuclide that enables imaging with better resolution, $^{68}$Ga$^{3+}$ has more amenable chemistry attributes for labeling than $^{99m}$Tc, a versatile SPECT radionuclide as well as $^{18}$F, the most widely used PET radionuclide. Numerous $^{68}$Ga based radiopharmaceuticals have been found useful in clinical studies [19,145-153]. $^{68}$Ga$^{3+}$ is stable and forms complexes with the cyclic ligand DOTA with high affinity and is thus suitable for preparation of high specific activity $^{68}$Ga-labeled peptides or other biomolecules conjugated to DOTA [19,145]. $^{68}$Ga-labelling of DOTA-coupled peptides can be performed in a very short time, allowing excellent imaging of neuroendocrine and neuroectodermal tumors [19,143,145]. Particularly, DOTA-TOC (DOTA-D-Phe$^1$-Tyr$^3$-octreotide) labeled with $^{68}$Ga have shown high binding affinity for human somatostatin receptors and possess excellent tumor imaging capabilities [152,153].

4.1.2 Availability of $^{68}$Ga from $^{68}$Ge/$^{68}$Ga generator

The $^{68}$Ge/$^{68}$Ga generator system is an excellent source to avail ready-to-use $^{68}$Ga for clinical positron emission tomography (PET) applications, allowing PET imaging at facilities
without an on-site cyclotron [19,142,154]. The cyclotron independent availability of $^{68}$Ga from a $^{68}$Ge/$^{68}$Ga generator at a reasonable cost makes it an attractive and realistic option for countries with limited or no cyclotron facilities. The relatively long-lived $^{68}$Ge ($t_{1/2}$ 270.95 d, electron capture (EC) 100%) produces short-lived $^{68}$Ga ($t_{1/2}$ 67.71 min), which subsequently decays to stable $^{68}$Zn [15,19]. The simplified decay scheme of $^{68}$Ge is illustrated in Fig. 4.1.

![Fig. 4.1: Simplified decay scheme of $^{68}$Ge (energy levels not drawn to scale)](image)

The long half-life of the parent radionuclide $^{68}$Ge ensures the cost-effective availability of $^{68}$Ga within the PET facility for long periods of time. The parent radioisotope, $^{68}$Ge can be produced in a small cyclotron through various reactions, such as, $^{69}$Ga (p,2n), $^{69}$Ga (d,3n), $^{66}$Zn ($\alpha$,2n) etc. involving varied targets and charged particles [19,155]. The processes amenable for the production of $^{68}$Ge are listed in Table 4.1. It can be seen from the table that though the yield of the $^{69}$Ga (p,2n) reaction using Ga$_2$O$_3$ film is almost comparable to that of Ge (p,xn) reaction, higher energy protons are required for the latter reaction. The Ge (p,xn) reaction is not preferred because the specific activity of $^{68}$Ge produced is significantly lower than that from $^{69}$Ga (p,2n) reaction. Moreover, radioarsenic isotopes are also formed in the Ge (p,xn) processes. The yields for the $^{69}$Ga (d,3n), Zn ($\alpha$,xn) and $^{66}$Zn ($\alpha$,2n) reactions are much smaller than for $^{69}$Ga (p,2n) and Ge (p,xn) reactions.
Table 4.1: The relevant nuclear reactions yielding $^{68}$Ge

<table>
<thead>
<tr>
<th>Nuclear reaction</th>
<th>Target form</th>
<th>Projectile</th>
<th>Yield</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Chemical</td>
<td>Physical</td>
<td>Thickness</td>
<td>Energy (MeV)</td>
</tr>
<tr>
<td>$^{69}$Ga (p,2n)</td>
<td>nat.$^{68}$Ga$_4$Ni</td>
<td>disk</td>
<td>3 mm</td>
<td>19.5-0</td>
</tr>
<tr>
<td>$^{69}$Ga (p,2n)</td>
<td>nat.$^{68}$Ga$_2$O$_3$</td>
<td>film</td>
<td>-</td>
<td>55-13</td>
</tr>
<tr>
<td>$^{69}$Ga (p,2n)</td>
<td>Ga</td>
<td>solid</td>
<td>-</td>
<td>22</td>
</tr>
<tr>
<td>nat.$^{66}$Ge (p,xn)</td>
<td>Ge</td>
<td>foil</td>
<td>107 mg cm$^{-2}$</td>
<td>64-28</td>
</tr>
<tr>
<td>$^{69}$Ga (d,3n)</td>
<td>nat.$^{68}$Ga$_2$O$_3$</td>
<td>disk</td>
<td>0.5 mm</td>
<td>30</td>
</tr>
<tr>
<td>nat.$^{66}$Zn ($\alpha$,xn)</td>
<td>Zn</td>
<td>foil</td>
<td>13 mg cm$^{-2}$</td>
<td>40-20</td>
</tr>
<tr>
<td>$^{66}$Zn ($\alpha$,2n)</td>
<td>Zn</td>
<td>film</td>
<td>-</td>
<td>40-20</td>
</tr>
</tbody>
</table>

'nat.' indicates natural target
Therefore, out of these, the $^{69}\text{Ga}$ (p,2n) reaction is most suitable for routine production of carrier free $^{68}\text{Ge}$ using a compact medical cyclotron [19,155], owing to the energetics of the reaction as well as the purity of the product that can be obtained.

Though the $^{68}\text{Ge}/^{68}\text{Ga}$ radionuclide generators have been the object of development and investigation for almost 50 years [19,154], their proper and relevant clinical use has started only recently [144,163,164], due to lack of proper sorbents, as one of the reasons. Undoubtedly, the major impetus for the development of $^{68}\text{Ge}/^{68}\text{Ga}$ generator stems from the recognized potential of PET technique and $^{68}\text{Ga}$-based radiopharmaceuticals, which demand instant, in-house availability of $^{68}\text{Ga}$ suitable for clinical use [165-167]. Several $^{68}\text{Ge}/^{68}\text{Ga}$ generator systems have been proposed over the past 50 years in an attempt to provide a reliable source of the positron-emitter $^{68}\text{Ga}$, that can readily be converted into radiopharmaceuticals for PET [19,155,168-182]. Of these systems, the column chromatographic $^{68}\text{Ge}/^{68}\text{Ga}$ generator has emerged as an effective, efficient, and the most popular generator system, owing to its simplicity and convenience to use in a hospital radiopharmacy.

In the early attempts towards the preparation of column chromatographic $^{68}\text{Ge}/^{68}\text{Ga}$ generators, inorganic oxides such as $\text{Al}_2\text{O}_3$ or $\text{ZrO}_2$ were used as sorbent materials [19,168]. For the preparation of these generators, carrier-free $^{68}\text{Ge}$ was absorbed onto the column and $^{68}\text{Ga}$ could be eluted with EDTA solution. The elution yields of such generators were appreciably high (~70-80%). Since these early generator systems provided $^{68}\text{Ga}$ in a chelated form, destruction of the EDTA complex was necessary, which rendered the preparation of the radiopharmaceutical tedious, time-consuming, and with a reduced overall yield. Thus, further development was focused on generator systems yielding $^{68}\text{Ga}^{3+}$ in its hydrated ionic form. Though there are several reports on the preparation of $^{68}\text{Ge}/^{68}\text{Ga}$ generators using metal oxides or hydroxides (like $\text{Al}_2\text{O}_3$, $\text{Fe(OH)}_3$, $\text{SiO}_2$, $\text{Sb}_2\text{O}_5$, $\text{SnO}_2$ and $\text{TiO}_2$) as sorbents [19,168-
from which $^{68}$Ga could be eluted in an ionic form, their clinical use failed because of the presence of these metal oxides or the corresponding metal ions in the $^{68}$Ga eluate \cite{19,168}. A very low breakthrough of the column packing material and the parent radioisotope were reported for a CeO$_2$ based $^{68}$Ge/$^{68}$Ga generator \cite{177}. However, this approach was not taken further as the yield of $^{68}$Ga from the CeO$_2$ based generator was low (~56%) \cite{177}. Alternatively, a macroporous styrene-divinylbenzene copolymer with N-methylglucamine groups was used as the sorbent matrix for the preparation of $^{68}$Ge/$^{68}$Ga generator. $^{68}$Ga was eluted from this generator with the low affinity chelator, sodium citrate, in good yields (~80%), and the $^{68}$Ge breakthrough was <0.0004% \cite{181,182}. Another complementary approach was the use of a pyrogallol-formaldehyde resin with high affinity for Ge (IV), where $^{68}$Ga was obtained as $^{68}$GaCl$_4^-$ using 5.5 M HCl as eluent \cite{173}. The [$^{68}$GaCl$_4^-$] complex was then adsorbed on a small anion exchange column to remove low levels of $^{68}$Ge breakthrough (<1 ppm). Elution with small volume of water resulted in the decomposition of the chloro complexes, and concentrated solutions of $^{68}$Ga$^{3+}$ in 0.5 M HCl were finally obtained. However, the performance of both these organic sorbents for the preparation of $^{68}$Ge/$^{68}$Ga generator has been demonstrated only with tracer level of activity (~370 kBq of $^{68}$Ge/$^{68}$Ga) \cite{173,181,182}. Therefore, further investigations on the radiation stability and performance of the organic sorbents at higher level of activity are needed before clinical applications are undertaken.

Nowadays, the most commonly available commercial $^{68}$Ge/$^{68}$Ga generator systems are based on ‘modified’ TiO$_2$ or SnO$_2$ sorbents \cite{19,167,183}, from which ionic $^{68}$Ga$^{3+}$ can be availed in 0.1-1 N HCl medium. The major limitation of these commercially available generators is that the $^{68}$Ga obtained from the primary column is not optimally suited for the routine synthesis of $^{68}$Ga-labeled radiopharmaceuticals \cite{19,167,183}. The $^{68}$Ga-eluates from most of the commercial generators have low specific volume of $^{68}$Ga and may contain
different trace elements owing to the solubility of metal oxide sorbent. The presence of these competing metal ions in the eluate is a major obstacle in the complexation chemistry of $^{68}$Ga [19,166,167,183] and therefore necessitates the inclusion of multiple post-elution processing steps [19,165-167,184,185]. Moreover, there is a drastic decrease in the elution yield of $^{68}$Ga and increase in the $^{68}$Ge breakthrough with passage of time or increasing number of elutions [19,166].

In view of the above described drawbacks, development of alternate sorbents with high sorption capacity and selectivity for $^{68}$Ge along with appreciable radiation resistance and chemical stability in acidic medium, is of considerable importance and deserves a serious consideration. Use of such sorbents would not only facilitate the elution of $^{68}$Ga with high radioactive concentration and avoid the need of additional concentration step, but also render $^{68}$Ga of acceptable radionuclidic and chemical purity.

**4.1.3 The present work**

The availability of a reliable, simple-to-handle $^{68}$Ge/$^{68}$Ga generator would facilitate more research on new diagnostic radiopharmaceuticals with $^{68}$Ga. In this chapter, the attention is focused on exploring the potential of nanomaterials as promising sorbents for the preparation of $^{68}$Ge/$^{68}$Ga generators. The potential of nanomaterial based sorbents in the preparation of clinically useful $^{99}$Mo/$^{99m}$Tc generators was described in the earlier chapter. It is expected that use of such materials can minimize the number of steps involved in obtaining clinical grade $^{68}$Ga from $^{68}$Ge. Several favorable characteristics, such as high surface area, availability of reactive surface sites and pore structure make nanoparticles excellent sorbent for generator preparation. This chapter describes the development of two novel $^{68}$Ge/$^{68}$Ga generators using tetragonal nano-zirconia (t-ZrO$_2$) and nano-ceria-polyacrylonitrile composite (CeO$_2$-PAN) as the sorbent matrices. The feasibility of each of these methods, in terms performance of the generators with respect to $^{68}$Ga-elution yield, low $^{68}$Ge
breakthrough, high radioactive concentration of the $^{68}$Ga solution and adequate purity of the $^{68}$Ga for preparation of radiopharmaceuticals, has been demonstrated and evaluated.

4.2 Materials

Reagents such as hydrochloric acid, ammonium hydroxide, etc. were of analytical grade and were procured from S.D. Fine Chemicals, India. High performance liquid chromatography (HPLC) grade water and zirconyl chloride ($\text{ZrOCl}_2\cdot8\text{H}_2\text{O}, +99.9\%$) were purchased from E. Merck, Germany. Cerium (III) nitrate ($+99.9\%$) was obtained from BDH, India. Analytical grade ($+99.999\%$) $\text{GeO}_2$ was procured from Aldrich, England. DOTA-TATE ($\text{DOTA-D-Phe}^1\text{-Tyr}^3\text{-octreotate, DOTA}=1,4,7,10\text{-tetraazacyclododecane-1,4,7,10-tetraacetic acid}$) was obtained from Pi Chem, Austria. Paper chromatography (PC) strips (3 MM, 20 mm width) were purchased from Whatman International Limited, England. $^{68}$Ge in HCl medium was obtained from Atom Hightech Company Limited, China, through an IAEA coordinated research project.

HPGe detector coupled with a multichannel analyzer (MCA) (Canberra Eurisys, France) with a 1.5 keV resolution at 1333 keV and range from 1.8 keV to 2 MeV was used for analysis of $^{68}$Ga. The efficiency of this instrument was estimated using a standard $^{152}$Eu source. Gamma activity of $^{68}$Ga was routinely assayed using a NaI (Tl) scintillation counter (400-600 keV). The chemical analysis for the determination of trace level of metal contaminations was done using Inductively Coupled Plasma-Atomic Emission Spectroscopy (ICP-ES JY-238, Emission Horiba Group, France). The UV-Visible spectrometry of the $^{68}$Ga samples were carried out using JASCO V-530, UV/Vis Spectrophotometer. The complexation yields of $^{68}$Ga labeled compounds were studied using HPLC technique. The HPLC instrument (JASCO PU 1580, Japan) was equipped with a NaI (Tl) detector as well as a UV-Visible spectrometer.
4.3 Separation of $^{68}$Ga from $^{68}$Ge

4.3.1 Synthesis of t-ZrO$_2$ and CeO$_2$-PAN composite

The synthesis of t-ZrO$_2$ and CeO$_2$-PAN was carried out as per the procedure described in Chapter 2. Owing to the simple and reliable synthesis procedure using commercially available chemicals, large scale preparation of these sorbents could be achieved in the laboratory. It was therefore considered worthy to establish the suitability of these materials for the preparation of $^{68}$Ge/$^{68}$Ga generators. The materials synthesized were porous with sufficiently large surface area and hence, could be used as a solid phase support in column chromatography operation. The materials demonstrated high resistance toward chemical attack and precluded the presence of Zr and Ce ion impurities in the eluate that could possibly interfere in complexation chemistry of Ga$^{3+}$ ions. Subsequent investigations were directed towards optimization of experimental conditions necessary for the effective separation of $^{68}$Ga from $^{68}$Ge.

4.3.2 Determination of the distribution ratios ($K_d$) of the $^{68}$Ge and $^{68}$Ga ions

In order to explore the potential of t-ZrO$_2$ and CeO$_2$-PAN for the separation of $^{68}$Ga from $^{68}$Ge, distribution ratios ($K_d$) of Ge and Ga ions were determined at different concentrations of HCl solutions. The $K_d$ values of the Ge and Ga ions were determined by batch equilibration method. A stoppered conical flask containing 200 mg of the respective sorbents, immersed in 20 mL of HCl solution spiked with 37 kBq of the $^{68}$Ge/$^{68}$Ga radiotracer, was shaken for 1 h at room temperature (25 °C). Subsequently, the supernatant solution was filtered through Whatman filter paper (No. 50). Since $^{68}$Ge decays solely by electron capture to $^{68}$Ga, the activity of $^{68}$Ge could not be directly estimated by $\gamma$-ray spectrometry. For determination of the activity of $^{68}$Ge, the filtrate was allowed to decay for 24 h, so that all the $^{68}$Ga activity would decay except the amount which grows from and is in equilibrium with the $^{68}$Ge. This $^{68}$Ga activity would correspond to the $^{68}$Ge activity and hence
measured in a well type NaI (Tl) counter using appropriate window settings (400-600 keV). On the other hand, for quantification of $^{68}$Ga, it was measured immediately after filtration. The $K_d$ values were calculated by using the following expression:

$$K_d = \frac{(A_i - A_{eq})V}{A_{eq}m}$$

where, $A_i$ is the initial total radioactivity of 1 mL the solution, $A_{eq}$ is the unadsorbed activity in 1 mL of the solution at equilibrium, $V$ is the solution volume (mL) and $m$ is the mass (g) of the adsorbent. All equilibration experiments were carried out in triplicate. The results obtained with t-ZrO$_2$ and CeO$_2$-PAN are summarized in Table 4.2 and Table 4.3 respectively.

### Table 4.2: Distribution ratios ($K_d$) of $^{68}$Ge and $^{68}$Ga ions in t-ZrO$_2$

<table>
<thead>
<tr>
<th>Conc. of HCl (M)</th>
<th>$K_d$ $^{68}$Ge</th>
<th>$K_d$ $^{68}$Ga</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.001</td>
<td>12598 ± 120</td>
<td>8 ± 2</td>
</tr>
<tr>
<td>0.01</td>
<td>12743 ± 88</td>
<td>0.10 ± 0.05</td>
</tr>
<tr>
<td>0.05</td>
<td>6645 ± 46</td>
<td>0.2 ± 0.1</td>
</tr>
<tr>
<td>0.1</td>
<td>6333 ± 52</td>
<td>0.10 ± 0.06</td>
</tr>
<tr>
<td>0.5</td>
<td>2131 ± 100</td>
<td>0.5 ± 0.1</td>
</tr>
<tr>
<td>1</td>
<td>1204 ± 84</td>
<td>0.7 ± 0.2</td>
</tr>
<tr>
<td>2</td>
<td>931 ± 50</td>
<td>0.6 ± 0.1</td>
</tr>
<tr>
<td>3</td>
<td>907 ± 53</td>
<td>0.4 ± 0.2</td>
</tr>
<tr>
<td>4</td>
<td>614 ± 45</td>
<td>0.4 ± 0.1</td>
</tr>
<tr>
<td>5</td>
<td>412 ± 61</td>
<td>0.6 ± 0.2</td>
</tr>
</tbody>
</table>

(*n = 3, ‘±’ indicates standard deviation*)
Table 4.3: Distribution ratios (K\textsubscript{d}) of \textsuperscript{68}Ge and \textsuperscript{68}Ga ions in CeO\textsubscript{2}-PAN

<table>
<thead>
<tr>
<th>Conc. of HCl (M)</th>
<th>K\textsubscript{d} \textsuperscript{68}Ge</th>
<th>K\textsubscript{d} \textsuperscript{68}Ga</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.001</td>
<td>5233 ± 144</td>
<td>113 ± 8</td>
</tr>
<tr>
<td>0.01</td>
<td>5152 ± 119</td>
<td>0.12 ± 0.07</td>
</tr>
<tr>
<td>0.05</td>
<td>4766 ± 68</td>
<td>0.2 ± 0.1</td>
</tr>
<tr>
<td>0.1</td>
<td>3033 ± 162</td>
<td>0.3 ± 0.1</td>
</tr>
<tr>
<td>0.5</td>
<td>2109 ± 113</td>
<td>0.5 ± 0.2</td>
</tr>
<tr>
<td>1</td>
<td>1654 ± 121</td>
<td>0.6 ± 0.2</td>
</tr>
<tr>
<td>2</td>
<td>1636 ± 111</td>
<td>0.7 ± 0.1</td>
</tr>
<tr>
<td>3</td>
<td>837 ± 56</td>
<td>0.6 ± 0.3</td>
</tr>
<tr>
<td>4</td>
<td>414 ± 59</td>
<td>0.7 ± 0.2</td>
</tr>
<tr>
<td>5</td>
<td>342 ± 77</td>
<td>0.7 ± 0.1</td>
</tr>
</tbody>
</table>

(n = 3, ‘±’ indicates standard deviation)

It can be seen from the tables that for both the sorbents, under all examined concentrations, Ge ions had significantly high K\textsubscript{d} values compared to Ga ions. Further, it can be noticed that the K\textsubscript{d} values for Ge ions decreased gradually with increase in concentration of HCl. On the other hand, \textsuperscript{68}Ga\textsuperscript{3+} ions had distinctly lower K\textsubscript{d} values under acidic conditions and could thus be effectively separated from \textsuperscript{68}Ge. Owing to the very high K\textsubscript{d} values of Ge ions in 0.01 M HCl and very low K\textsubscript{d} values of Ga\textsuperscript{3+} under the same conditions, it was decided to use 0.01 M HCl solution for elution of Ga\textsuperscript{3+}. Quantitative retention of \textsuperscript{68}Ge was achieved and \textsuperscript{68}Ga could be easily eluted out using 0.01 M HCl solution.

Nano zirconia and nano ceria particles can be considered to consist of discrete metal oxide clusters covered by surface hydroxyl groups. The interaction of nanosized metal oxide particles with aqueous solutions results in the hydroxylation of surface sites and this imparts a
pH-dependent surface charge which is primarily responsible for the uptake of metal ions. This phenomenon was studied by determination of the zeta potential of these materials at different pH environments. The zeta potential of t-ZrO$_2$ (Fig. 2.12) was positive up to pH 4, while in case of CeO$_2$-PAN (Fig. 2.16), the zeta potential values were positive up to pH 6. On further increase in pH, the zeta potential of these nanomaterials became zero (IEP) and subsequently became increasingly negative with further rise in pH. In dilute acid solutions (pH 2-3), the principal germanium compounds probably are [GeO(OH)$_3$]$^-$, [GeO$_2$(OH)$_2$]$^{2-}$ and [[Ge(OH)$_4$]$_6$(OH)$_3$]$^{3-}$ which are negatively charged [186,187]. The strong affinity of the sorbents for $^{68}$Ge ions at pH ~2 is probably due to the electrostatic attraction of negatively charged germanium ions for the positively charged surface of the t-ZrO$_2$ and CeO$_2$-PAN. In the same medium, Ga exists as Ga$^{3+}$ ions and hence a nearly complete elution could be achieved due to electrostatic repulsion of the positively charged Ga$^{3+}$ ion from the positively charged surface of the nanomaterials.

However, on increasing the acidity of the solution, the $K_d$ values of Ga ions increased slightly. This might be due to the tendency of formation of negatively charged [GaCl$_4$]$^-$ complex at higher acidity which is probably retained on the positively charged surface of the nanomaterials. Therefore, 0.01 N HCl solution was chosen for elution of $^{68}$Ga in all subsequent experiments.

4.3.3 Determination of the time of equilibration

In order to study the time dependence of sorption of $^{68}$Ge onto t-ZrO$_2$ and CeO$_2$-PAN, the $K_d$ of $^{68}$Ge ions was determined in 0.01 M HCl as a function of time. The attainment of equilibrium was indicated by the constant $K_d$ value after a certain period of time and the results for t-ZrO$_2$ and CeO$_2$-PAN are shown in Fig. 4.2 and Fig. 4.3, respectively. It could be inferred from the figures that the equilibrium was attained within 50-60 minutes, when t-ZrO$_2$
was used as the sorbent and within 25-30 minutes when CeO$_2$-PAN was used. Therefore, this contact period was maintained in all subsequent batch equilibration experiments.

![Graph showing variation in $K_d$ values of $^{68}$Ge ions with time using t-ZrO$_2$ as sorbent](image1)

**Fig. 4.2:** Variation in $K_d$ values of $^{68}$Ge ions with time using t-ZrO$_2$ as sorbent

![Graph showing variation in $K_d$ values of $^{68}$Ge ions with time using CeO$_2$-PAN as sorbent](image2)

**Fig. 4.3:** Variation in $K_d$ values of $^{68}$Ge ions with time using CeO$_2$-PAN as sorbent
4.3.4 Determination of the sorption capacity of t-ZrO$_2$ and CeO$_2$-PAN

The sorption capacity of the sorbents were evaluated both under static as well as dynamic conditions. The solution of non-radioactive Ge(IV) was prepared by dissolving GeO$_2$ in 0.1 N sodium hydroxide. The concentration and the initial pH of the Ge solution were adjusted with 0.1 N HCl. HPGe detector coupled with a multichannel analyzer was used for analysis of $^{68}$Ga and $^{68}$Ge and also for their quantitative determination.

4.3.4.1 Static sorption capacity

The static sorption capacities of t-ZrO$_2$ and CeO$_2$-PAN for Ge ions were determined by batch equilibration method. An accurately weighed amount of sorbent (~200 mg) was taken in a stoppered glass conical flask and equilibrated with 20 mL of the Ge solution (5 mg of Ge mL$^{-1}$) spiked with ~370 kBq (10 µCi) of $^{68}$Ge, for 1 h at pH 2. Subsequently, the contents were filtered and the filtrate was allowed to decay for 24 h. The activity of the decayed $^{68}$Ge solution was measured using a NaI(Tl) detector and compared with that of the standard $^{68}$Ge solution. The sorption capacity was calculated using the following expression:

$$\text{Capacity} = \frac{(A_o - A_e)V.C_o}{A_o m}$$

where $A_o$ and $A_e$ represented the radioactivity of $^{68}$Ge in 1 mL of supernatant solution before and after sorption, respectively, $C_o$ was the total Ge content (5 mg) in 1 mL of solution before sorption, $V$ was the volume of solution and $m$ was the mass (g) of the sorbent. The batch equilibration studies indicated that the static sorption capacities of t-ZrO$_2$ and CeO$_2$-PAN were 135 ± 10 mg Ge g$^{-1}$ and 40 ± 5 mg g$^{-1}$, respectively, in 0.01 M HCl solution (n = 10).

4.3.4.2 Determination of breakthrough pattern and dynamic sorption capacity

In order to evaluate the sorption capacity of the nanomaterials for Ge under dynamic conditions, a borosilicate glass column of dimension 8 cm × 0.6 cm (i.d.) with a sintered disc ($G_0$) at the bottom was packed with a known amount of the respective sorbent. After the column was conditioned with 0.01 M HCl, Ge solution (5 mg Ge mL$^{-1}$), spiked with
$^{68}$Ge/$^{68}$Ga radiotracer, was allowed to pass through the column at a flow-rate of ~0.25 mL min$^{-1}$. 1 mL of this solution was kept as reference. The effluent was collected in fractions of 1 mL aliquots and allowed to decay for 1 day. The $^{68}$Ge activity in the reference ($C_0$) and effluent fractions ($C$) were determined by measuring the 511 keV $\gamma$-ray peak in a HPGe detector. The ratio of the count rate ‘$C$’ of each 1 mL effluent fraction to the count rate ‘$C_0$’ of 1 mL of the original feed Ge solution was taken as the parameter to follow the sorption pattern. The breakthrough curves developed for t-ZrO$_2$ and CeO$_2$-PAN are illustrated in Fig. 4.4 and Fig. 4.5, respectively. It can be seen from Fig. 4.4 that for t-ZrO$_2$, the breakthrough point was reached after (76±3) mg of Ge (n=10), was quantitatively retained by 1 g of the sorbent in the column.

Fig. 4.4: Breakthrough profile of Ge ions on passing Ge (5 mg Ge mL$^{-1}$) solution through a 500 mg t-ZrO$_2$ column at a flow rate of 0.25 mL min$^{-1}$
Fig. 4.5 shows that the breakthrough capacity of CeO$_2$-PAN was (20±2) mg of Ge per g of the sorbent (n = 10). These results reflect that even a small column containing 200 mg of t-ZrO$_2$ or CeO$_2$-PAN is adequate for the preparation of a 37 GBq (1 Ci) $^{68}$Ge/$^{68}$Ga generator.

**Fig. 4.5: Breakthrough profile of Ge ions on passing Ge (5 mg Ge mL$^{-1}$) solution through a 1 g CeO$_2$-PAN column at a flow rate of 0.25 mL min$^{-1}$**

It must be mentioned here that the concentration of Ge ions (5 mg Ge mL$^{-1}$) used in these experiments, are far higher than the ‘real’ generator situation. However, it still serves the purpose and provides information on the sorption capacities of both the sorbent materials for Ge ions, which are appreciably high. This concentration of Ge ions was chosen to indicate the excellent capacity of the sorbent, much more than adequate to adsorb Ge ions expected to be present in a clinical scale $^{68}$Ge/$^{68}$Ga generator. Since $^{68}$Ge is available in no-carrier added form, the amount of Ge present in 37 GBq (1 Ci) of $^{68}$Ge solution is only 0.14 mg. On passing Ge solutions, containing 0.14 mg of Ge (equivalent to 37 GBq of $^{68}$Ge) through two columns, each containing only 200 mg of t-ZrO$_2$ or CeO$_2$-PAN, it was found that Ge ions were quantitatively taken up by the respective sorbents.
4.4 Development of $^{68}\text{Ge}/^{68}\text{Ga}$ generators

The separation processes could be demonstrated by developing two 740 MBq (20 mCi) $^{68}\text{Ge}/^{68}\text{Ga}$ generators using t-ZrO$_2$ and CeO$_2$-PAN as sorbent matrices, respectively. For the preparation of each generator, a borosilicate glass column of dimension 8 cm × 0.6 cm (i.d.) with a sintered disc ($G_0$) at the bottom was packed with 1 g of the respective sorbent and kept in a lead shielded container. It was pre-conditioned with 0.01 M HCl solution. A schematic diagram of the $^{68}\text{Ge}/^{68}\text{Ga}$ generator system is shown in Fig. 4.6. All the operations were carried out in the closed cyclic system using connecting tubes. Input/output connections were made with standard teflon tubings of 1 mm inner diameter and connectors. The generator column, connectors and connection tubings were integrated within a small portable lead shielded unit throughout experimental use for radioprotection purpose. Only the elution vial and output vial were accessible externally. A disposable 0.22 µm membrane filter was attached to the generator column output by teflon tubing.

Fig. 4.6: Schematic diagram of the $^{68}\text{Ge}/^{68}\text{Ga}$ generator system
The $^{68}$Ge/$^{68}$Ga solution containing 740 MBq (20 mCi) of $^{68}$Ge at pH 2 was percolated into each column maintaining a flow rate of 0.25 mL min$^{-1}$. In case of both t-ZrO$_2$ as well as CeO$_2$-PAN based generators, about 740 MBq (20 mCi) of $^{68}$Ge activity at pH 2, were quantitatively retained by the sorbents. The $^{68}$Ge-loaded generator columns were then washed with 100 mL of 0.01 M HCl solution. On washing with 0.01 N HCl solution, only <0.5% of $^{68}$Ge came out, from each generator column.

4.5 Choice of the eluent for the elution of $^{68}$Ga from the $^{68}$Ge/$^{68}$Ga generators

For the success of a $^{68}$Ge/$^{68}$Ga generator in clinical PET, it is essential to obtain $^{68}$Ga in an uncomplexed chemical form, maximize the elution yield of $^{68}$Ga, minimize the $^{68}$Ge breakthrough and keep the elution volume to a minimum. These requirements place severe constraints on the eluent that can be used, and the choice is rather limited. Gallium exists exclusively in +3 oxidation state in aqueous acidic solutions. It hydrolyzes nearly completely at neutral pH [188,189] and readily forms highly insoluble amorphous Ga(OH)$_3$, while Ga ions at hydrochloric acid concentrations $>$5.5 M form the negatively charged [GaCl$_4$]$^{-}$ complex [185]. In view of these limitations, it was decided to work under acidic conditions and use 0.01 M HCl as eluent, based on effectiveness of this solution in desorbing $^{68}$Ga$^{3+}$ from the sorbents while maintaining the level of $^{68}$Ge impurity in the eluate to a permissible level. The decay of $^{68}$Ge to $^{68}$Ga is not accompanied by any serious disruption of chemical bonds. As these $^{68}$Ge ions start transforming into $^{68}$Ga ions, which do not form negative complexes at 0.01 M HCl, they are not retained by the sorbent matrices and hence an easy displacement of $^{68}$Ga$^{3+}$ ions is expected.

4.6 Elution profiles of $^{68}$Ge/$^{68}$Ga generators

In order to optimize the minimum volume of eluent required for the elution of $^{68}$Ga with maximum yield and radioactive concentration, the elution profiles of the generators were studied by collecting the $^{68}$Ga eluates as 0.5 mL aliquots, and the activity of each
fraction was determined by measuring the 511 keV γ-ray peak in a HPGe detector. The $^{68}$Ga-elution profiles of t-ZrO$_2$ and CeO$_2$-PAN based generators are illustrated in Fig. 4.7 and Fig. 4.8, respectively.

![Elution profile of the t-ZrO$_2$ based $^{68}$Ge/$^{68}$Ga generator](image)

**Fig. 4.7: Elution profile of the t-ZrO$_2$ based $^{68}$Ge/$^{68}$Ga generator**

It can be seen from the figures that the elution profiles of both the generators are quite sharp in nature. In the case of t-ZrO$_2$ based generator, <2% of the $^{68}$Ga was eluted in the first 1 mL fraction and >90% of the $^{68}$Ga activity was eluted in the subsequent 2 mL of eluate. Therefore, for the regular elutions of the t-ZrO$_2$ based generator, the first 1 mL fraction containing negligible amount of $^{68}$Ga was discarded and the majority of $^{68}$Ga was available in 2 mL of 0.01 N HCl solution with appreciable radioactive concentration. Similarly, in the case of the CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generator, <3% of the $^{68}$Ga was eluted in the first 1.5 mL fractions and >90% of the $^{68}$Ga activity was obtained in the subsequent 2 mL of eluent. Therefore, for the regular elutions of the CeO$_2$-PAN based generator, the first 1.5 mL fraction containing negligible amount of $^{68}$Ga was discarded and the majority of $^{68}$Ga in the
subsequent in 2 mL of 0.01 N HCl solution was used for the preparation of radiopharmaceuticals.

![Diagram](image.png)

**Fig. 4.8: Elution profile of the CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generator**

**4.7 Quality control of $^{68}$Ga eluate**

**4.7.1 Radionuclidic purity**

In order to utilize $^{68}$Ga obtained from $^{68}$Ge/$^{68}$Ga generators for clinical applications, special attention is necessary to ensure that the level of $^{68}$Ge radionuclidic impurity present in $^{68}$Ga is always below the permissible limit. This is important since, with the passage of time, even trace amounts of the long-lived $^{68}$Ge impurity would reach appreciable proportions as a radionuclidic impurity. When small quantities (MBq/mCi levels) of radiopharmaceuticals labeled with short-lived $^{68}$Ga are administered to patients for diagnostic purposes, even fractional percentages of the long-lived parent ($^{68}$Ge), if present, will add to the radiation dose to the patients and are therefore undesirable. The radionuclidic purity of the $^{68}$Ga eluted from both the generators was assessed by measuring the half-life of $^{68}$Ga and by $\gamma$-ray spectrometry.
of the decayed $^{68}$Ga samples using a calibrated HPGe detector coupled to a multi-channel analyzer.

4.7.1.1 Decay pattern of $^{68}$Ga

The decay pattern of $^{68}$Ga was monitored by following the half-life of $^{68}$Ga using a NaI (Tl) counter (window 400-600 keV). The decay was followed for nearly 6 h (~6 half-lives of $^{68}$Ga). The decay profile of a typical $^{68}$Ga sample is shown in Fig. 4.9. The absence of any deviation at the lower end of the straight line decay curve confirmed that the $^{68}$Ga samples were pure and contained negligible quantities of long-lived $^{68}$Ge. The results were almost similar in case of both the generators. The half-life of $^{68}$Ga as calculated using the decay profile was $(66.7 \pm 0.5)$ min ($n = 5$) which was close to the 67.71 min half-life reported for $^{68}$Ga.

![Decay profile of $^{68}$Ga](image)

**Fig 4.9: Decay profile of $^{68}$Ga**

4.7.1.2 $\gamma$-ray spectrometry

Since $^{68}$Ge decays solely by electron capture to $^{68}$Ga, the amount of $^{68}$Ge contamin-
ation in $^{68}$Ga eluate could not be directly estimated by $\gamma$-ray spectrometry. The $^{68}$Ge contamination level in $^{68}$Ga was quantified by allowing the separated $^{68}$Ga samples to decay for 24 h and then measuring the 511 keV $\gamma$-ray peak, corresponding to emission from $^{68}$Ga daughter. This in turn corresponds to the level of $^{68}$Ge contaminant, which exists in secular equilibrium with $^{68}$Ga. The amount of $^{68}$Ge impurities in $^{68}$Ga eluates from both the generators were always <20 Bq ($<10^{-5}$% of the total $^{68}$Ga activity) in all the elutions over the period of 1 year.

The radionuclidic purities of $^{68}$Ga obtained from the t-ZrO$_2$ and CeO$_2$-PAN based generators were comparable to that obtained from commercial generators [19,166,167]. However, it must be noted that the eluate ($^{68}$Ga) from commercial generators were subjected to multiple purification steps [2,166,167,183] to obtain clinical grade $^{68}$Ga, whereas, the $^{68}$Ge/$^{68}$Ga generators described here provided $^{68}$Ga of similar purity in a single step. This proved that $^{68}$Ga was availed from the nanomaterials based generators with high radionuclidic purity and was hence suitable for radiopharmaceutical applications.

4.7.2 Chemical purity

The $^{68}$Ga eluted from the generators might be associated with other metal ions (like Zr, Ce, Cu, Pb, Co, Cr, Cd, Ni, Fe, Mn and Zn ions) as chemical impurities [19,167]. The presence of Zr and Ce ions in the $^{68}$Ga eluates could be a possibility due to dissolution of t-ZrO$_2$ and CeO$_2$-PAN in HCl medium, respectively. The other metal ion impurities like Cu, Pb, Co, Cr, Cd, Ni, Fe, Mn ions could be introduced through the raw materials used and are noted to be occasionally present in the $^{65}$Ge solution [19,167]. Additionally, $^{68}$Ge may contain significant amounts of Zn ions as a decay product of $^{68}$Ga. [19,167]. These metal ions might be eluted along with $^{68}$Ga in HCl medium. The presence of these chemical impurities in $^{68}$Ga might interfere in the complexation of $^{68}$Ga with various ligands and biomolecules. Thus it is necessary to estimate the concentrations of these metal ions in the $^{68}$Ga as a quality assurance
step. In order to determine the presence of chemical impurities in the $^{68}$Ga eluate, the $^{68}$Ga samples were allowed to decay for 7 days. The trace levels of the metal ion contamination in the decayed samples were determined by ICP-AES analysis. The calibration curves for these ions were obtained by using standard solutions having known concentration of these ions.

The level of Zr ions in the $^{68}$Ga eluate from the t-ZrO$_2$ based $^{68}$Ge/$^{68}$Ga generator, as analyzed by ICP-AES analysis, was found to be as low as (0.05±0.01) ppm [(0.05±0.01) µg mL$^{-1}$] (n=5). Similarly, in the $^{68}$Ga eluate from the CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generator, the level of Ce ions was (0.08 ± 0.03) ppm [(0.08±0.03) µg mL$^{-1}$] (n = 5) The level of Zr and Ce ions impurity in the $^{68}$Ga eluates were analyzed by random selection of $^{68}$Ga samples over a period of 1 year. It was found that the level of Zr and Ce ions in the $^{68}$Ga eluates from the respective generators was consistently low over this prolonged period of time. It is clear from the result that both t-ZrO$_2$ and CeO$_2$-PAN are stable to both radiation as well as chemical degradation on repeated elution over a prolonged period of 1 year and do not lead to the dissolution of the column matrices, unlike the conventional sorbents [19,166,167].

In the $^{68}$Ga eluted from both the generators, Pb and Cr ions were below the detectable limits of the ICP-AES system used and the amounts of Co, Cu, Cd, Ni, Fe ions etc. were <5 ng mL$^{-1}$. Owing to the unavailability of instrumental facility to analyze radioactive samples, elemental analysis of Zn in the freshly eluted $^{68}$Ga samples could not be carried out. The presence of Zn ions below appreciable amounts which could interfere in the complexation chemistry of $^{68}$Ga could be indirectly tested by investigating the labeling efficacy of $^{68}$Ga, described in the next section.

It was also essential to scrutinize the $^{68}$Ga eluate from the CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generator for the presence of organic residues, possibly due to the radiolytic degradation of the polyacrylonitrile matrix. Trace amounts organic residue from the polyacrylonitrile binding matrix was assayed by UV-Visible spectrometry using the decayed $^{68}$Ga samples. It
is reported that PAN shows weak absorption at \( \lambda_{\text{max}} \) of 278 nm in the UV-Visible spectra, which corresponds to the n–\( \pi^* \) transition of nitrile-groups [190]. From the UV-Visible spectra of the decayed \(^{68}\)Ga samples it could be inferred that PAN residue was not present in the \(^{68}\)Ga eluate as no absorption was observed at this wavelength.

4.8 Labeling efficacy of \(^{68}\)Ga

In order to evaluate the suitability of \(^{68}\)Ga for biomedical applications, it was used for the preparation of \(^{68}\)Ga-DOTA-TATE. This is also an indirect test to ascertain the chemical purity, as high chemical purity is essential to achieve a good complexation yield of the radiolabeled agent. For radiolabeling of DOTA-TATE with \(^{68}\)Ga, 20 \( \mu \)L of DOTA-TATE solution (1 \( \mu \)g \( \mu \)L\(^{-1} \)) in HPLC grade water was mixed with 980 \( \mu \)L of 0.01 M ammonium acetate buffer (pH \( \sim \)5) and 1 mL of \(^{68}\)Ga eluate (~296 MBq, 8 mCi in 0.01 N HCl medium) was added to it. The pH of the resulting mixture was found to be ~4-4.5, and was carefully adjusted to ~4 (if required) by addition of 0.1 N HCl. The resulting mixture was incubated at 90 \( ^\circ \)C for 15 min. The extent of complexation achieved was determined by paper chromatography (PC) using 50% acetonitrile in water (v/v) as the eluting solvent as well as by high performance liquid chromatography (HPLC). HPLC was carried out using a dual-pump HPLC unit with a C-18 reversed phase HiQ-Sil (5 \( \mu \)m, 25 \( \times \) 0.46 cm) column. The elution was monitored by measuring the 511 keV \( \gamma \)-ray of \(^{68}\)Ga using NaI (Tl) detector (window 400-600 keV). The mobile phase consisted of water (A) and acetonitrile (B) mixtures with 0.1% trifluoroacetic acid and the following gradient elution technique was adopted for the separation: 0-4 min 95% A, 4-15 min 95% to 5% A, 15-20 min 5% A, 20-25 min 5% A to 95% A, 25-30 min 95% A. The flow rate was maintained at 1 mL min\(^{-1} \). The paper chromatography (PC) patterns of the \(^{68}\)Ga-DOTA-TATE and uncomplexed \(^{68}\)Ga\(^{3+} \) as \(^{68}\)GaCl\(_3\) are shown in Fig. 4.10.
Fig. 4.10: Paper chromatographic patterns of (a) $^{68}\text{Ga}$-DOTA-TATE and (b) $^{68}\text{GaCl}_3$ in 50% acetonitrile in water
From the PC pattern, it can be seen that $^{68}$Ga-DOTA-TATE moved towards the solvent front ($R_f = 0.8-0.9$) (Fig 4.10a), while under identical conditions unlabeled $^{68}$Ga$^{3+}$ remained at the point of application ($R_f = 0$) (Fig. 4.10b). Further, it was observed that as low as 20 µg of DOTA-TATE (13.9 nmol) was sufficient for labeling ~296 MBq (8 mCi) of $^{68}$Ga with >99% complexation yield. The complexation yield of $^{68}$Ga-DOTA-TATE was validated by HPLC studies. A typical HPLC pattern of $^{68}$Ga-DOTA-TATE is shown in Fig. 4.11.

![Fig. 4.11: HPLC pattern of $^{68}$Ga-DOTA-TATE](image)

The specific activity of $^{68}$Ga-DOTA-TATE was ~21.3 MBq nmol$^{-1}$ and it was obtained with >99% radiochemical purity. The high radiochemical purity of $^{68}$Ga-DOTA-TATE was comparable to that of $^{68}$Ga-DOTA-peptides prepared by the reported methods, adopting multiple purification steps [19,166,167,185]. The present findings amply suggest that traces of Zn ions, if present, in the eluate do not interfere significantly in the complexation chemistry of $^{68}$Ga. The small volume of the $^{68}$Ga eluate from the t-ZrO$_2$ and CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generators, with appropriate radioactive concentration and the availability of $^{68}$Ga$^{3+}$ ions in an uncomplexed and highly pure form, facilitate the direct use of $^{68}$Ga for labeling of biomolecules without the inclusion of post-elution processing procedures.

4.9 Elution performance of the generators over a period of 1 year

The $^{68}$Ge/$^{68}$Ga generators were eluted on every working day over a period of 1 year.
Over this period of time, the generators were eluted for more than 260 times. The performance of the t-ZrO$_2$ and CeO$_2$-PAN based generators with respect to the radiochemical yield of $^{68}$Ga and the $^{68}$Ge breakthrough is illustrated in Fig. 4.12 and Fig. 4.13, respectively. The figures show that for the both the generators, the radiochemical yield of $^{68}$Ga was always $>80\%$ and the level of $^{68}$Ge impurity present in $^{68}$Ga was $<10^{-5}\%$ over this period of 1 year.

**Fig. 4.12: Performance of the t-ZrO$_2$ based $^{68}$Ge/$^{68}$Ga generator over a period of 1 y, data points shown at 10 d intervals**

A major advantage of the t-ZrO$_2$ and CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generators is the consistency in their performance which is far superior to that of the widely used SnO$_2$ and TiO$_2$ based $^{68}$Ge/$^{68}$Ga generators, which have been reported to show degrading performance on repeated elution, over a prolonged period of time [19,166,170]. It is reported by Asti et al [166], that the amount of $^{68}$Ge breakthrough increased with time ($\sim15\%$ increase per month), ranging from $1.1\times10^{-2}\%$ to $2.6\times10^{-2}\%$ of the $^{68}$Ga activity within the 7 months of evaluation.
Moreover, the elution yields of $^{68}$Ga from these generators decreased from 82% to 69% on repeated elution (100 times) over the period of 7 months [166].

![Graph](image)

**Fig. 4.13: Performance of the CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generator over a period of 1 y, data points shown at 10 d intervals**

Though the consistency of the elution yield (>80%) and purity of $^{68}$Ga are good indications of the radiation stability of the sorbents, the effect of radiation on the performance of t-ZrO$_2$ and CeO$_2$-PAN loaded with clinically useful amount of $^{68}$Ge (~1.85 GBq or 50 mCi) is yet to be demonstrated. However, it is well reported that nanocrystalline metal oxides exhibit enhanced radiation stability compared to the bulk materials [68]. For both the generators, the sorbent matrices were stable to radiation, over a prolonged period of time, and did not lead to bleeding of Zr or Ce ions in the $^{68}$Ga eluate, unlike the commercially available TiO$_2$ or SnO$_2$ based $^{68}$Ge/$^{68}$Ga generators [19,166,167,183]. However, this needs to be ascertained by preparing generators of much higher activity.
4.10 Simulated study for the separation of Ga from a Ge carrier-added solution, equivalent to ~3.7 GBq (100 mCi) of $^{68}$Ge

The performance of t-ZrO$_2$ and CeO$_2$-PAN as column matrices for $^{68}$Ge/$^{68}$Ga generators of higher level of activity, was investigated by using inactive Ge carrier-added solution simulated to represent ~3.7 GBq (100 mCi) of $^{68}$Ge. The simulated solution was prepared by dissolving 21 µg of GeO$_2$ (equivalent to 3.7 GBq of no-carrier-added $^{68}$Ge) in 0.1 M NaOH. The resultant solution was evaporated to dryness and then reconstituted with 0.01 M HCl solution. The above solution was spiked with an equilibrium mixture of $^{68}$Ge/$^{68}$Ga containing 185 MBq (5 mCi) of $^{68}$Ge. The pH of the solution was adjusted to ~2 by adding 1 N HCl. The mixture was loaded in a borosilicate glass column [8 cm × 0.6 cm (i.d.)] containing 1 g of the respective sorbent, adopting the procedure outlined above. $^{68}$Ga was eluted with 3 mL of 0.01 N HCl solution under the same conditions as in the previous studies. The efficiency of $^{68}$Ga elution and the $^{68}$Ge breakthrough were determined, for the generators developed using t-ZrO$_2$ and CeO$_2$-PAN sorbents. For both the sorbent materials, the recovery of $^{68}$Ga from Ge/Ga mixture simulated to represent 3.7 GBq (100 mCi) of $^{68}$Ge, was as good when lower amounts of $^{68}$Ge were used. The overall yields of $^{68}$Ga in the simulated experiments were always >80% and the $^{68}$Ge breakthrough was <10$^{-5}$%. It must be mentioned here that although the simulated experiments gives a fair idea about the sorption capacity and separation efficacy of both the sorbents at higher level of activity, the effect of radiation dose over a prolonged period of time on the performance of the material is yet to be demonstrated.

4.11 Advantages of t-ZrO$_2$ and CeO$_2$-PAN based $^{68}$Ge/$^{68}$Ga generators

The present study clearly suggests that both t-ZrO$_2$ and CeO$_2$-PAN are very effective sorbents for the preparation of $^{68}$Ge/$^{68}$Ga generator for clinical applications. The major benefits in the use of these nanomaterial based sorbents in the preparation of $^{68}$Ge/$^{68}$Ga generators are: (1) high capacity of the sorbents owing to small size and high specific surface
area (2) rapid packing due to the high density of the sorbents that settles in a few minutes, (3) rigidity which allows the use of high flow rates, (4) enhanced chromatographic efficiency due to large surface to volume ratio and (5) negligible $^{68}$Ge bleeding due to the stable chemical link of the $^{68}$Ge species to the matrix and (6) chemical and radiation stability of the sorbent matrices (7) direct usability of the $^{68}$Ga eluate for the preparation of radiopharmaceuticals.

Several modifications can be incorporated in these new generator systems developed and reported here. This includes scaling up to higher activity level (up to 3.7 GBq, 100 mCi), use of extremely high purity reagents to avoid metal ion contamination, elution of $^{68}$Ga under sterile conditions and automation of the entire process. It is my goal to carry out all these developments in near future to improve this generator into an easily adaptable system for hospital radiopharmacies.

### 4.12 Conclusions

In the present study, a rapid, simple, reliable and chemically efficient chromatography method has been established using t-ZrO$_2$ and CeO$_2$-PAN as sorbent matrices, to avail $^{68}$Ga with acceptable radioactive concentration, yield and purity from $^{68}$Ge. The efficacy of these new generation sorbent materials could be demonstrated by developing two 740 MBq (20 mCi) $^{68}$Ge/$^{68}$Ga generators, which are still giving consistently good performance after repeated elutions over a period of 1 year. $^{68}$Ga could be regularly eluted from the generator with acceptable radioactive concentration with substantially high yield and purity. The efficacy of $^{68}$Ga for the preparation of radiopharmaceuticals for PET imaging could be confirmed by radiolabeling DOTA-TATE with very high complexation yield. The results presented here are promising and the generator systems are amenable for automation. These generators may be very useful for countries where commercial sources of PET radioisotopes are not readily available or too expensive.