SAMARIUM-153 LABELLED MICROPARTICLES FOR LIVER TUMOUR TARGETED THERAPY WITH IMAGING FUNCTIONALITY

N.A.A. Hashikin[1], C.H. Yeong[1], B.J.J. Abdullah[1], K.H. Ng[1], L.Y. Chung[2], R. Dahalan[3], and A.C. Perkins[4]

1 Department of Biomedical Imaging and University of Malaya Research Imaging Centre, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur, Malaysia

2 Department of Pharmacy, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur, Malaysia

3 Medical Technology Division, Malaysian Nuclear Agency, 43000 Kajang, Bangi, Malaysia.

4 Radiological and Imaging Sciences and Nottingham Digestive Diseases Biomedical Research Unit, University of Nottingham, Nottingham, NG7 2UH, United Kingdom.

Abstract— **Samarium-153 (^{153}Sm) are widely used in radiation synovectomy and pain management for patients with bone metastases. However, its therapeutic application has not been fully explored. ^{153}Sm has been proven to be useful for imaging purposes. This provides a beneficial alternative for therapy with pure beta emitter especially for liver radioembolization with Yttrium-90 (^{90}Y). This study aimed to develop an alternative radioembolic agent using ^{153}Sm and biocompatible resin microparticles for liver cancer therapy. The ion-exchange resin; Amberlite IR-120 H^+ commercially available in large beads were crushed and sieved to 20 – 40 µm and labelled with $^{152}SmCl_3$ salt prior to neutron activation. Administered activity of 3 GBq ^{153}Sm was aimed based on the standard activity used by the ^{90}Y SIR-Spheres. 6 hours irradiation in 1.494 x 10^{12} $n.cm^{-2}.s^{-1}$ flux produced 3.1 $GBq.g^{-1}$ immediately after activation. Characterization of the microparticles, gamma spectroscopy, and in-vitro radiolabelling studies were carried out and compared to a commercially available resin readily made in 20 – 40 µm, Fractogel EMD SO_3^- (S). ^{153}Sm-Amberlite microparticles possess a superior and suitable characteristics for liver radioembolization with added imaging capabilities.**

Keywords— **Samarium-153 (^{153}Sm), liver cancer, neutron activation, radioactive microspheres, radioembolization.**

I. Introduction

Liver radioembolization is a non-physiological targeted therapy where radiolabeled embolic particles are percutaneously delivered directly to the tumour. Liver malignancies i.e. hepatocellular carcinoma (HCC) is the third most common cause of death from cancer worldwide (1). HCC is often diagnosed at the later stages when curative approaches are no longer feasible (2). Radioembolization using ^{90}Y-microparticles are now increasingly used as palliative treatment for HCC. This approach may prolong patients' survival and provide possibilities for curative intents by down-staging the tumours for possible resection or transplantation (3-7).

Currently two commercially available radioembolic agents; glass (TheraSphere®, Nordion, Canada) and resin (SIR-Spheres, SIRTex, Australia) microparticles, both labelled with ^{90}Y. ^{90}Y is produced by high-purity separation from Strontium-90 (^{90}Sr), which is a nuclear fission product of Uranium-235 (^{235}U) fuel in a nuclear reactor. The complexity of ^{90}Y production resulted in high cost ^{90}Y-microparticles. Since ^{90}Y is a pure beta emitter, the distribution of ^{90}Y-microparticles after each procedure is difficult to be verified. Technetium-99m macro-aggregated albumin (^{99m}Tc-MAA) has been used prior to therapy for lung shunting quantification providing brief information of source distribution within the liver and lungs. However, this pretreatment planning method did not accurately reflect the intrahepatic distribution of ^{90}Y-microparticles. This is due to resolution and partitioning dissimilarities between ^{99m}Tc and ^{90}Y images, as a result of different physical characteristics and number of particles infused (8, 9). Bremsstrahlung imaging may be used, but with very poor spatial resolution.

Radionuclides with both therapeutic beta and diagnostic range of gamma energies would be ideal for "theranostics" (therapy plus diagnostic) treatment. Ideal therapeutic radionuclide has optimum physical half-life, suitable linear energy transfer (LET) and range in tissue, high ratio of non-penetrating to penetrating radiation, short lived or stable daughter, good and selective concentration with prolonged retention in tumour and minimum uptake by normal tissue (10). Neutron activation is preferred in radionuclide production due to wide availability of reactors and relatively simpler process. ^{153}Sm is potentially suitable as alternative to ^{90}Y. The imaging properties of ^{153}Sm has been proven feasible in a gastrointestinal scintigraphy by Yeong, Abdullah (11). Most important microparticles' feature is size range of 20 – 40 µm. Microparticles with resistivity to physical heat and body chemicals, near plasma density, biocompatible and easily labelled with radionuclides are highly preferred.

II. Materials & Methods

A. Preparation of ^{152}Sm-labelled microparticles

A commercially available ion-exchange resin; Amberlite IR-120 H^+ (620 – 830 µm) was obtained from Fluka GmbH (Buchs, Switzerland). Samarium (III) chloride hexahydrate ($^{152}SmCl_3 \cdot 6H_2O$) with assay purity ≥99 % was obtained from Aldrich Chemical Co. (Wisconsin, USA). The Amberlite IR-120 resin was oven dried at 70°C for 12 h. The dried resin was ground using a grinding planetary ball mill machine (XQM-(2-6) L, ChangSha LangFeng Metallic Material Ltd., China) at 200 rpm for approximately 5 h. The resin powder was subsequently sieved using a mechanical sieve shaker (AS 200 Analytical Sieve Shaker, Retsch GmbH, Haan, Germany) attached with 20 and 40 µm wire mesh stainless steel test sieves (Endecotts Ltd., London, UK). Another commercial ion-exchange resin, Fractogel EMD SO_3^- (S) (Merck Millipore, Massachusetts, USA) suspended in 20 % ethanol and 150 mmol.l^{-1} NaCl, ready made in 20 – 40 µm was used for functional comparison. Using Büchner funnel filtration, Fractogel resin was thoroughly washed with distilled water to eliminate the ethanol and NaCl. 1 g of $SmCl_3.6H_2O$ was dissolved in 10 ml distilled water. 5 g of washed Fractogel resin was poured into the $SmCl_3$ solution and stirred for 5 min to allow binding of the Sm^{3+} ions to the resin. The ^{152}Sm-Fractogel resin was washed by flushing distilled water through the resin to remove unbound Sm^{3+} ions. These steps were repeated for ^{152}Sm-Amberlite resin. Finally, both formulations were oven dried at 70°C for 12 h.

B. Characterisation of ^{152}Sm- microparticles

Fourier transform infrared (FTIR) spectroscopy (600 – 4000 cm^{-1} range) was carried out (Nicolet 6700, Thermo Fisher Scientific Inc., Massachusetts, USA) on the Amberlite resin. FTIR spectra of $SmCl_3$ salt, fresh Amberlite resin beads, resin after grinding and sieving, resin after labelling, and resin after 6 h neutron activation were compared. Field emission scanning electron microscopy (FESEM) and energy dispersive X-ray (EDX) spectroscopy were carried out on both ^{152}Sm-resins using a FESEM system (Quanta FEG 250, FEI, Oregon, USA). The particle density, ρ_s of both ^{152}Sm-microparticles were measured using a helium gas pycnometer (AccuPvc II 1340, Micromeritics Ins. Corp., Georgia, USA) at standard room temperature of 25°C. The ρ_s was incorporated into following equation to obtain particle concentration, PC (particles.ml^{-1}) for each ^{152}Sm-microparticles in 0.9 % saline solution:

$$PC = \frac{6\rho_f}{\pi D_p^3 \left(\rho_f + \frac{\rho_s}{C} - \rho_s\right)} \quad (1)$$

where,

C : mass fraction (% w/w)
D_p : mean diameter of the particles (cm)
ρ_f : density of the solvent (g.cm^{-3})
ρ_s : particle density (g.cm^{-3})

C. Neutron activation

Both ^{152}Sm-microparticles were neutron activated in Malaysian Nuclear Agency (MNA), Selangor, Malaysia. The TRIGA PUSPATI Reactor (RTP) (Triga Mark II, General Atomics, California, USA) is a pool type with solid enriched uranium (20 % weight, 235U). The samples were sealed in individual polyethylene vial and placed into polyethylene ampoule. Two neutron activation methods (Table 1); Pneumatic Transfer System (PTS) and Rotary Specimen Rack (RR), were studied to achieve 3 GBq of ^{153}Sm. The irradiation time, t can be estimated using the formula:

$$A_t = \sigma_{act}\, \varphi N(1-e^{(-\lambda t)}) \quad (2)$$

where,

A_t : activity (Bq)
σ_{act} : thermal neutron activation cross-section (barns)
φ : neutron flux (n.cm^{-2}.s^{-1})
N : number of parent atoms = (m / w) x θ x 6.023 x 10^{23};
m : mass of element in the sample
w : atomic weight of element
θ : isotopic abundance
λ : decay constant (s^{-1}) = 0.693/ $t_{1/2}$
t : irradiation time (s)

Table 1. Neutron activation protocols to achieve ^{153}Sm activity of 3 GBq.

Method	PTS	RR
Thermal neutron flux, θ_{th} (n.cm^{-2}.s^{-1})	4.813 x 10^{12}	1.494 x 10^{12}
Irradiation time	5 minutes	6 hours
Location in the reactor	Near to the core	Peripheral to the core
Sample entrance and exit	Automatic	Manual

D. Gamma spectroscopy

After 48 h of cooling, gamma spectroscopy was carried out for each sample to determine presence of long-lived radionuclide impurities. Hyper-pure germanium detector (Canberra, Meriden, USA) and gamma spectrum analysis software (GenieTM 2000 Ver. 3.2, Canberra, Meriden, USA) were used. Each sample was counted for 5 min at a distance so that detection yield do not exceed 20 %.

E. Optimum formulation and radiolabelling efficiency

1 g of $SmCl_3.6H_2O$ was labelled to 1, 2, 3, 4, 5, and 6 g of each resin to determine optimum formulation with best labelling efficiency. All samples were activated via PTS for 5 min. Each sample was equally separated into three 10 ml test tubes followed by addition of 10 ml distilled water. The samples were mixed using a roller mixer (Movil-Rod, J.P. Selecta, Barcelona, Spain) at 50 rpm for 1 h. Next, the samples were centrifuged at 1200 rpm for 5 min. 1 ml of supernatant was pipetted from each tube and transferred into gamma assay tubes. These steps were repeated until a total of 8 ml supernatants were obtained from each sample within 48 h. All supernatant samples were assayed using gamma scintillation counter (2470 Wizard2, PerkinElmer Inc., Massachusetts, USA). All steps were repeated in human blood plasma. Labelling efficiency of each formulation was calculated using equation previously used (12):

$$\text{Retained activity (\%)} = (A_{sus}-A_{sup})/A_{sus} \times 100\ \%$$

where,

A_{sus}: Activity of suspension before supernatant extraction

A_{sup}: Activity of supernatant

III. Results

In Figure 1, the functional groups (1000 – 1200 cm^{-1}) of the resin were still present despite harsh physical process during sample preparation. No major differences between peaks in spectra shown in Figure 1 (b) – (e).

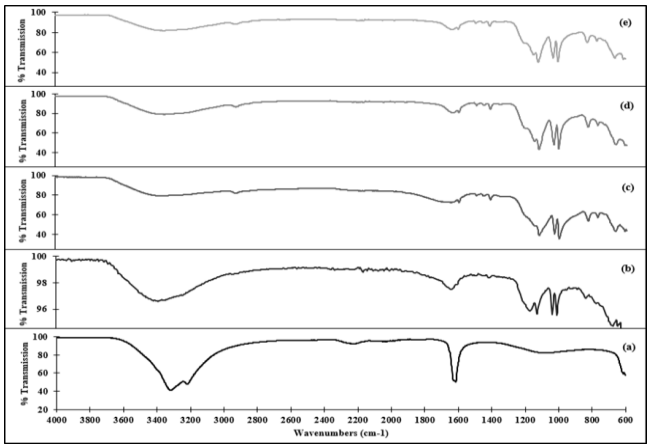

Figure 1. (a) $SmCl_3.6H_2O$ salt. (b) Fresh Amberlite IR-120 H^+ beads. (c) Amberlite IR-120 H^+ ground and sieved to size 20 – 40 µm. (d) Amberlite microparticles labelled with $SmCl_3.6H_2O$ salt. (e) ^{153}Sm-Amberlite microparticles after 6 h neutron activation.

In Figure 2 (a), ^{152}Sm-Amberlite microparticles were observed to be irregular in shapes, however the size are in the acceptable range of 20 – 40 µm. EDX spectra of both resins showed that they comprised mostly of C, O, Sm and S. However, for ^{152}Sm-Fractogel, Cl was also found present in a significant amount (1.66 % atomic).

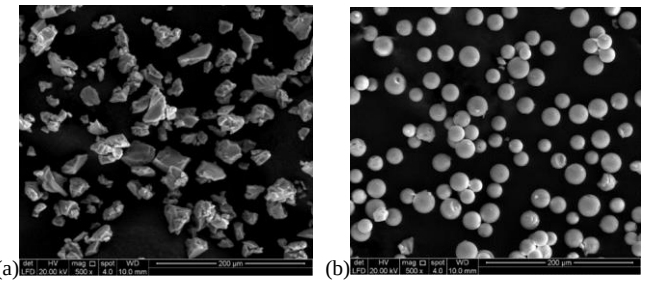

Figure 2. FESEM images of (a) ^{152}Sm-Amberlite and (b) ^{152}Sm-Fractogel microparticles.

The particle density of ^{152}Sm-Amberlite and ^{152}Sm-Fractogel is 2.538 ± 0.012 and 2.283 ± 0.002 $g.cm^{-3}$ respectively. These correspond to 27.7 and 30.7 million microparticles respectively. The specific activity per 1 g of ^{153}Sm-resins immediately after 5 min activation via PTS was 0.148 ± 0.004 GBq. This correspond to only 0.072 GBq after 48 h. The specific activity achieved via RR method immediately after 6 h activation was 3.104 ± 0.029 GBq. The corresponded activity of 1.513 GBq after 48 h was closer to the initial target of 3 GBq. Hence, specific activity per microparticle for ^{153}Sm-Amberlite and ^{153}Sm-Fractogel were 55 Bq and 49 Bq respectively.

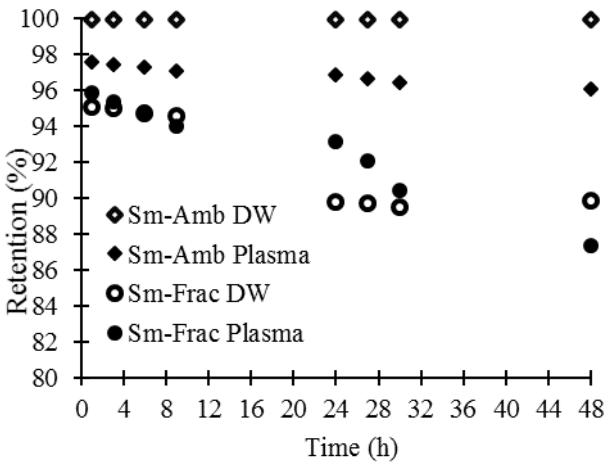

Figure 3. Percentage retention of ^{153}Sm in both resin suspended in distilled water (DW) and blood plasma over 48 h.

The most dominant photopeak observed in both samples was the 103.1 ± 0.2 keV, associated with principle gamma energy of ^{153}Sm. In the ^{153}Sm-Fractogel samples, two other peaks were consistently observed; 1368.4 ± 0.2 and 2753.1 ± 0.2 keV, associated with ^{24}Na. No significant impurities were observed in ^{153}Sm-Amberlite samples.

^{153}Sm-Amberlite shows significantly better labelling efficiency with 8.42 ± 0.86 % higher compared to ^{153}Sm-

Fractogel. The optimum formulations determined for both ^{153}Sm-Amberlite and ^{153}Sm-Fractogel was 1:3 and 1:4 respectively. ^{153}Sm-Amberlite showed better retention over 48 h in both distilled water and blood plasma (Figure 3).

IV. Discussion

Ion exchange resins were chosen due to its relatively easy labelling and commercial availability. Resins are generally chemically inert hence results in minimal radionuclide leaching. Due to its insoluble characteristic, it is not absorbed by the body thus, are extremely safe to be use in medicinal products with limited side effects (13). Amberlite IR-120 H^+ was chosen due to its excellent labelling efficiency as reported in an earlier study (12).

Chlorine (Cl) in the Fractogel resin may be activated into radioactive chlorine (^{38}Cl) during neutron activation. The presence of Cl in the Fractogel resin is due to the NaCl suspension in its commercial packing. From gamma spectroscopy, only ^{24}Na we found in the samples because ^{38}Cl may already been fully decay because of its short half-life. However, if the concentration is higher, ^{38}Cl may still be present. This issue may be overcome with more thorough resin washing during preparation.

The particle densities of the ^{152}Sm-microparticles developed in this study was in between the density of the commercial SIR-Sphere (1.6 g.cm^{-3}) and TheraSphere (3.2 g.cm^{-3}) particles. Since ^{153}Sm-Amberlite are slightly dense, this resulted in lower number of microparticles per gram which eventually contribute to higher specific activity per microparticles compared to ^{153}Sm-Fractogel.

Despite being non-spherical as a result of grinding, ^{153}Sm-Amberlite possess much better functional quality in all aspects compared to ^{153}Sm-Fractogel. The labelling efficiency and retention of ^{153}Sm-Amberlite showed that shape irregularity may not be a huge problem since ^{153}Sm are still mostly intact and the capacity of binding is significantly higher than the other resin.

V. Conclusion

We have prepared 20 – 40 µm microparticles using ion exchange resin labelled with ^{153}Sm produced via neutron activation. It is easy to prepare and does not involve unnecessary radiation exposure during the labelling process. Amberlite IR-120 resin was chosen rather than Fractogel EMD SO^{3-} because of its excellent labelling efficiency with strong retention of ^{153}Sm tested over 48 h, no radioactive impurities produced from neutron activation, and lower production cost. ^{153}Sm-microparticles has the potential to be an optimal option as an alternative to ^{90}Y-microparticles, with added advantage of gamma radiation for imaging of source distribution. Dosimetric studies to estimate total ^{153}Sm activity needed to deliver equivalent tumour dose and therapeutic response from 3 GBq ^{90}Y shall be carried out. Further animal studies for in-vivo distribution, biochemical stability and labelling efficacy should also be carried out prior to clinical studies.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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Use macro [author address] to enter the address of the corresponding author:

Author: C.H. Yeong
Institute: Department of Biomedical Imaging and University of Malaya Research Imaging Center, Faculty of Medicine
Street: University of Malaya, 50603
City: Kuala Lumpur
Country: Malaysia
Email: chyeong@um.edu.my