# Separation Of Radioisotope $^{113m}$In Using Column Chromatography Based on Silica Gel Matrix

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**ABSTRACT**

Radioisotope indium-113m ($^{113m}$In) with half-life, $T_{1/2}$ = 1.7 hours and gamma energy, Eγ = 391 keV is suitable and meets the criteria as radiotracer in industry. $^{113m}$In radioisotope was obtained from tin-113 decay ($^{113}$Sn, $T_{1/2}$ = 115 days) of $^{112}$Sn (n,γ) $^{113}$Sn neutron activation in nuclear reactor. The process of separation of radioisotope $^{113m}$In using column chromatography method based on silica gel matrix using 0.05 M HCI solution. Radionuclide and radiochemical purity tests were performed using the gamma-spectrometry method and paper chromatography. The final product specification in the form of $^{113m}InCl_3$ is clear solution, pH 2, obtained yield of 81.83%, radionuclide purity of 90.22%, radiochemical purity of 91.61 ± 0,29% and stable for 3 days at room temperature.

**Keywords**: $^{113\,m}$In radioisotopes, silica gel, chromatography, radiotracer, industry

## INTRODUCTION

The rapid advance in radiotracer investigation has aroused widespread interest in the use of short-lived gamma-emitting radionuclides, especially for many industrial and nuclear medicine applications that require radioactivity for several minutes to several days. By using the short-lived radionuclide tracers, we could conduct measurement repetitions in short interval of time without having to worry about the remaining activities from the previous test. One of the disadvantages of using short half-life gamma emitter radionuclides as the industrial and nuclear medicine tracer is practical problems that only be used at near location to the nuclear reactor facility, because the short half-life limits the transport problem at the delivery distance. Therefore, timely availability in investigation location is the main obstacles that limit the use of this tracer in the industry and nuclear medicine.

To ensure a continuous supply of short-lived radionuclides, radioisotope generators (RG) are an effective strategy for solving practical problems. RG thus facilitates the activities in remote locations and also enables fast service delivery to handles urgent problems. Provision RG (in industry) would be cheaper than medical (nuclear medicine) radionuclide generators since there is no necessary to comply with strict regulatory requirements of radiopharmaceuticals.

Many couples of parent and daughter radionuclides have potential to be used in RG system, in this research, $^{113}$Sn was selected to be evaluated on the basis of half-lives, types of radiation and energy as well as ease of production. Parent radionuclide, $^{113}$Sn decay 98.2 percent to $^{113m}$In as a daughter radionuclide which possesses 391.69 keV of gamma energy and a half-life at 1.7 hours, likely to be used as RG $^{113}$Sn/$^{113m}$In in the nuclear medicine and industry. While the half-life of the parent is 115 days, this generator offers about 1 year of application (International Atomic Energy Agency, 2013; Mostafa, A. El-Sadek, El-Said, & El-Amir, 2009; Tárkányi et al., 2011).

Considering that $^{113}$Sn radioisotope is neutron reaction products from $^{112}$Sn (n, γ) which has natural isotope abundances in the amount of 0.97 % and a very small cross-section of 1 barn, it is necessary to use $^{112}$Sn enriched isotope (enrichment more than 95%). $^{113}$Sn radioisotope can be produced at low flux research reactors. Separation process $^{113m}$In via elution using diluted hydrochloride acid (theoretical: 70% of the activity $^{113m}$In and escaped $^{113}$Sn < 0.01% of $^{113m}$In activity (Allan, Ali, Hanafi, & El-Azony, 2010). The chemical form of $^{113m}$In in RG $^{113}$Sn/$^{113m}$In eluate is $In^{3+}$ at low chloride concentration. While higher chloride concentrations, it formed complex chlorides such as $InCl^{2+}$, and even $InCl^{2+}$ on a stronger chloride concentration converted to

anionic form $InCl^{4-}$ (Abdel-Halim, 2002; Narita et al., 2014).

The utilization of radioisotopes $^{113m}$In that has a medium half-life (1.7 hours), low gamma energy (391 keV) and excess chemical properties of various concentrations of chloride, $^{113m}$In become the main choice in the application of nuclear techniques for industrial/ hydrology field. One of the activities in hydrology is to reduce the speed of sedimentation (silt/sediment) in boat harbor by knowing where it came from and the direction of movement of these sediments (International Atomic Energy Agency, 2011). Besides that, $^{113m}$In has the potential utilization in various industrial fields as a tracer for aqueous phase, organic or solid. To understand the behavior of each stage of tracing process is important to investigate the performance of the system, indium-113m is possible for labeling complex compounds that stable in these phases (Toncheva, Gavazov, Lekova, Stojnova, & Dimitrov, 2011). Radioisotope $^{113}$Sn was obtained by neutron activation or irradiation of the $^{112}$Sn in a nuclear reactor. Separation process of Indium-113m from RG $^{113}$Sn/$^{113m}$In by elution with HCl 0.2 N to obtain $^{113m}In^{3+}$ ions, ($^{113m}InCl_3$). How to mastery $^{113m}InCl_3$ radioisotope production from nature tin metal target using column chromatographic separation methods based on silica gel matrix is the purpose of this research.

## EXPERIMENTAL SECTION

### Reagents and Instrumentation.

Materials used in this study are tin (Sn) metal from E. Merck, hydrochloric acid, silica gel 60 (from 0.063 to 0.200 mm) from E. Merck Catalog No. 107 734 (70-230 mesh ASTM), hydrogen peroxide ($H_2O_2$ 30%), distilled water, ammonium molybdate (1%), phosphate buffer ($Na_2HPO_4$), activated carbon (0.3 to 0.5 mm), universal pH indicator paper, 90% ethanol, 85% acetone, Whatman 1chromatography paper (E. Merck), Whatman 31ET chromatography paper (E. Merck), and Whatman 3MM chromatography paper. While the equipments used are cutlery quartz, analytical balance METTLER toledo AL 204, micropipette (Eppendorf), centrifuge, heater (Thermolyne) brand Nouva II, spectrometer γ single channel (SCA) ORTEC models 402 A, spectrometer-γ multi-channel (MCA) with a detector HPGe Canberra DSA-1000, oven Heraeus T5050, nuclear reactors GA Siwabessy BATAN, hot cells, glove boxes, container, inner and outer aluminum capsule (nuclear grade) , quartz glass, glass columns, syringes (Terumo syringe), glass vial, set of paper chromatography system including chamber, beakers, measuring cups, measuring pipette, pasteur pipette, tweezers, spatula, and gloves (Sensi gloves).

### Irradiation and chemical process of $^{112}$Sn target.

1000 mg Sn metal target containing quartz glass was placed in the aluminum nuclear grade inner capsule, and then closed by welding. Furthermore, the inner capsule was conducted via bubble method in water media. After that, the inner capsule was inserted into the outer capsule for irradiation process in a reactor RSG-GA Siwabessy (BATAN, Serpong) at CIP (Center Irradiation Position) with neutron flux of 1 x $10^{14}$ $n.cm^{-2}.s^{-1}$ for ± 94.5 hours. Irradiation was completed with Sn Safety Analysis Reports (SARs) document.

Irradiated material target was removed from the aluminum container, then put into a 250-ml beaker and dissolved in 100 ml of HCl 6 N. Dissolving process has been done on a heater with temperature of 80 °C, and was stirred gently at the same time with a magnetic stirrer to completely dissolved, then added 2 drops of concentrated $H_2O_2$. Thereafter, the solution was evaporated slowly until 50 mL left. $Sn^{2+}$ qualitative test was performed by pipeting 20 μL of the solution diluted to 1 mL, and were added by 1 drop of ammonium molybdate. The change of blue color indicates cation $Sn^{2+}$ (Allan et al., 2010). In this process Sn was expected to be in the form of $Sn^{4+}$ cations. The solution was transferred into a glass vial and sealed using a cover vial glass (rubber cap). The whole work was conducted in hot cells. Activity measurement was done using spectrometry- γ MCA-HPGe at energy 225 keV ($^{113}$Sn) and 393 keV ($^{113m}$In).

### Determination of optimum condition for impregnation process of irradiated Sn metal into silica gel.

#### *Solvent concentration variation*

About ± 250 mg silica gel was weighted and placed into 5 vial bottle. Each vial is filled each vial with 1 mL solution of radioactive Sn-metal (item 2.1) and set to be variation of solvent concentration of HCl (0.05; 0.1; 0.2;

0.5; 1 N) by adding distilled water. The solution was homogenized by stirring for ± 25 minutes. Then, silica gel was separated from the solution using a centrifuge. Next, pipette as ± 20µL filtrate and measured the radioactivity using Multi-Channel Analyzer instrument. Finally, do the calculation of the distribution coefficient. The distribution coefficient of three key radioisotopes i.e. $^{113}$Sn, $^{113m}$In, and $^{117}$mSn was calculated as the ratio of the radioactivity concentration of each radioisotope on solid matrix compared to the radioactivity concentration of those radioisotopes on the solution.

***Stirring time variation***

Weigh ± 250 mg silica gel into 6 vial bottle, fill each vial with 1 mL solution of active Sn- metal with concentration of HCL 0.1 N (item 2.2.1). The solution was homogenized by stirring for 5, 10, 15, 20, 25, and 30 minutes. Then, silica gel was separated from the solution using a centrifuge. Next, pipette as ± 20µL filtrate and measured the radioactivity using Multi-Channel Analyzer instrument. Finally, do the calculation of the distribution coefficient.

***Weight variation of silica gel***

Weigh silica gel into 6 vial bottle with different mass (100;200;250;300;400;500 mg), then fill each vial with 1 ml solution of active Sn- metal in HCl 0,1 N (item 2.2.1). The solution was homogenized by stirring for 25 minutes (item 2.2.2). Then, silica gel was separated from the solution using a centrifuge. Next, pipette as ± 20µL filtrate and measured the radioactivity using Multi-Channel Analyzer instrument. Finally, do the calculation of the distribution coefficient.

**Separation process of $^{113m}$In by using silica gel chromatography column**

Weigh ± 250 mg of silica gel into vial bottle then fill each vial with 1 mL solution of active Sn- metal in HCl 0,1 N (item 2.2.1). The solution was homogenized by stirring for 25 minutes (item 2.2.2). The solution was added to a glass column chromatography (ø 0.5 cm x 7.5 cm). Silica gel remains in the column, while the parent solution is removed and collected. $^{113m}$In elution using 0.05 M HCl solution, the filtrate is collected 10 times of 1 mL per fraction/ vial. Then, $^{113m}$In radioactivity was measured by the Multi Channel Analyzer (HPGe-MCA).

**Radionuclide and radiochemical purity test of $^{113m}$In.**

Radionuclide purity was determined from gamma ray spectrum of $^{113m}$In as the HPGe-MCA analysis result. 5 µL dosage of radionuclide $^{113m}InCl_3$ was dropped on filter paper, dried, put in a plastic wrapper, and counted for 60 minutes. γ-ray spectrum characteristic of $^{113m}InCl_3$ at 391 keV energy peaks. The radionuclide purity (KRN) of $^{113m}$In was calculated using the equation below:

$$KRN = \frac{Activity\ 113mIn\ radioisotope}{Total\ Activity} x100\%$$

While the radiochemical purity was determined by paper chromatography using a mixture of 90% ethanol and 5 M HCl 10% (v / v) or acetone 85% as mobile phases and Whatman No.I chromatography paper as a stationary phase was observed at retention factor as 0.7 of In (III). $^{113m}InCl_3$ solution was dropped at a distance of 2 cm from the bottom of Whatman 1, then the paper was put to the chamber that had been saturated by the eluent. Elution was carried out until the migration distance of the mobile phase reaches 14 cm. The paper was dried, cut every 1 cm and counted using γ-counter single channel analyzer (SCA).

## RESULT AND DISCUSSION

**Irradiation result of $^{112}$Sn target**

10 µL solution of the target (section 2.1) dropped on the vial and radioactivity concentration was measured using a spectrometry-γ MCA-HPGe. Gamma spectrum was analyzed, measurements were performed 3 times of repetition. Identification $^{113}$Sn and $^{113m}$In radioisotopes were determined using BATAN BANDUNG NAA software (Lestiani, Muhayatun, & Adventini, 2009) and resulted in a graph between counts and the energy as shown in **Figure 1**. The peak at 255 keV was $^{113}$Sn counts and 391 keV for $^{113m}$In radioisotope. **Figure 1** showed the presence of other radionuclides than $^{113}$Sn and $^{113m}$In which formed, were $^{117m}$Sn and $^{125}$Sb radionuclides which derived from the core reaction of their isotopes by neutron at irradiation process. $^{117m}$Sn came from the decay of irradiated result $^{116}$Sn (natural isotopic abundance of 14 %), while $^{125}$Sb resulted from the decay of irradiated $^{124}$Sn. $^{116}$Sn and $^{124}$Sb isotopes were

contained in metal Sn target (Cradarelli, 2008; Duyeh, Nana, & Titin, 2013).

Radioactivity $^{113}$Sn as a form $[^{113}SnCl_6]^{2-}$ obtained by measurement using the spectrometry-γ MCA-HPGe was 0.0467 mCi/mL, while theoretically at EOI (End Of Irradiation) should be 0.0504 mCi/mL, means obtained yield was 92.65%. Decay reactions that occur between parent radioisotopes ($^{113}$Sn) and daughter radioisotope ($^{113m}$In) were secular equilibrium reaction where in $^{113}$Sn has a half life 115 days, relatively longer than $^{113m}$In as 1.6 hours, allowed for separation by column chromatography using silica gel matrix (Saha, 2010).

## Distribution Coefficient (KD) Determination

### *Solvent concentration variation*

Distribution coefficient values resulted from the experiment (show as **Figure 2**) indicated that the radionuclide $^{113}$Sn and $^{113m}$In can be separated using silica gel in a suitable solvent. The higher value of KD indicated the nuclide had stronger interaction and well absorbed into silica gel matrix. The optimum concentration of HCl to impregnate $^{113}$Sn as parent nuclide of $^{113m}$In was achieved in 0.1N where KD of $^{113}$Sn was 156.27 ml/g. Although this process could not separate $^{113}$Sn from other Sn radionuclide $^{117m}$Sn which had KD of 110.44 mL/g, impregnation process using this concentration can separated $^{113m}$In from its parent nuclide. On the optimum point (using HCl 0.1 N), $^{113m}$In had low KD. The KD of $^{113m}$In was around 20 mL/g which mean some $^{113m}$In was trapped in silica gel matrix. Higher KD value of $^{113}$Sn and lower KD value of other nuclide indicated the better impregnation process. Beside solvent type and concentration, KD value is influenced by contact time and the amount of solid matrix. **Figure 2** showed the optimum distribution coefficients was achieved in 0.1N HCl with KD of $^{113}$Sn was 156.27 mL/g and KD of $^{113m}$In was 16.11 mL/g, therefore KD of $^{117m}$Sn was 110.44 mL/g.

### *Stirring time variation*

The results of the analysis of the distribution coefficient determination based on agitation time as shown in **Figure 3**. **Figure 3** visualized the optimum distribution coefficient was obtained at stirring process or contact time for 25 minutes with the values of KD of $^{113m}$In, $^{113}$Sn and $^{117m}$Sn were 8.86 mL/g, 206.95 mL/g and 159.98 mL/g respectively. The effect of agitation time on KD was not clear enough. This might be caused from inconsistency of stirring speed. Generally, KD of $^{113}$Sn is higher than $^{113m}$In almost in all variation of time. However, the KD value resulted from 25 minutes of agitation showed the highest KD of $^{113}$Sn with the low KD of $^{113m}$In. This time parameter was used in preparation of the column chromatography.

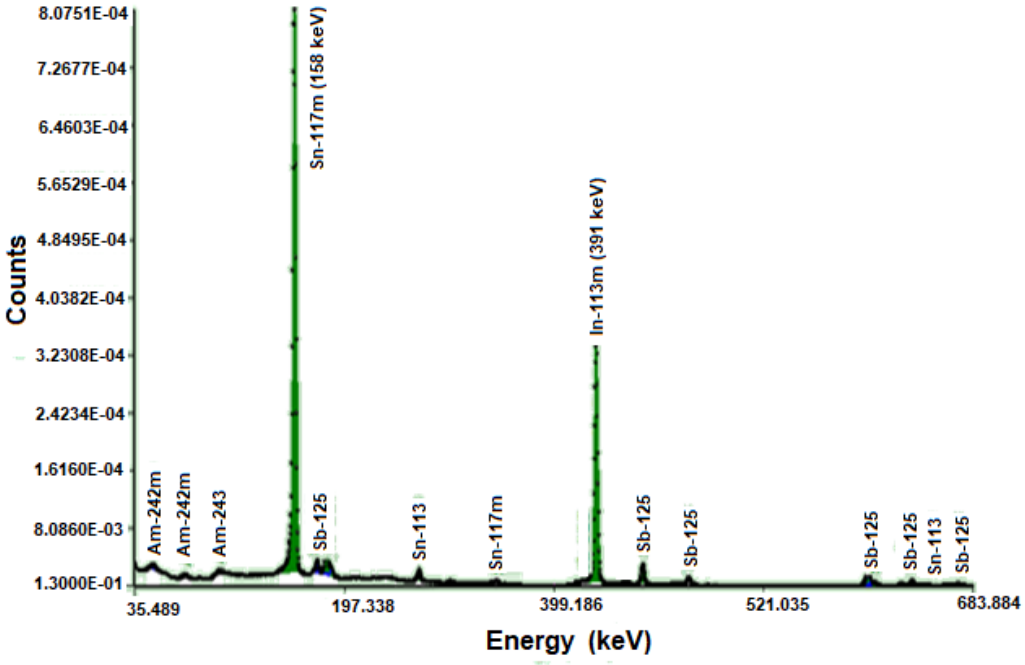



**Figure 1.** Gamma spectrum of $^{113}$Sn – $^{113m}$In after irradiation.

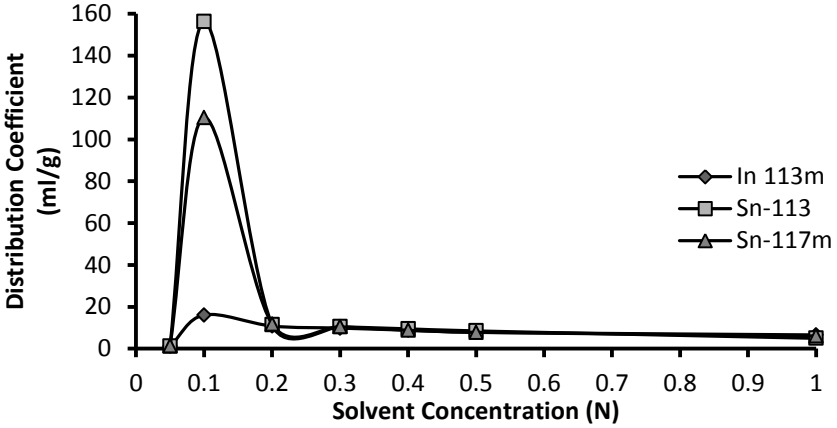



**Figure 2**. Coefficients distribution at solvent concentration variation

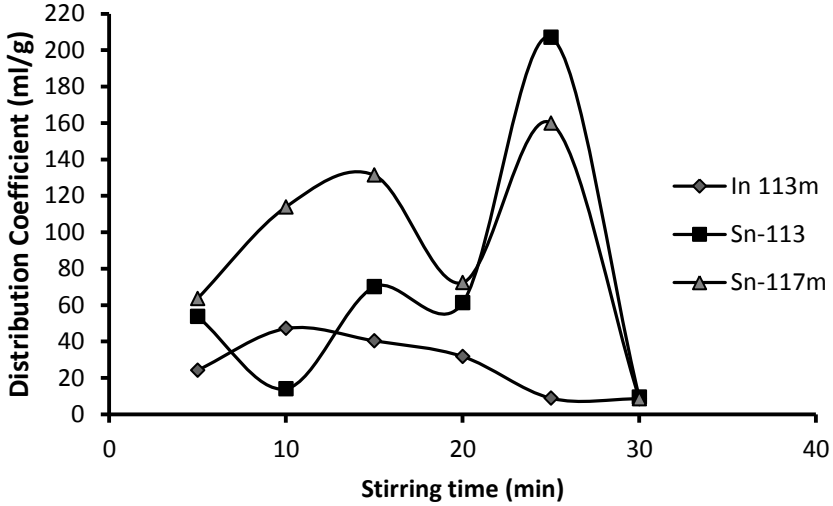



**Figure 3**. Coefficients distribution at contact time variation

***Weight of silica gel variation***

The analysis result of the distribution coefficient based on the weight variation of silica gel as shown in **Figure 4**. **Figure 4** visualized the optimum distribution coefficient was obtained at 250 mg silica with the values of KD of $^{113}$Sn was 240.12 mL/g, KD of $^{113m}$In was 8.79 mL/g, and KD of $^{117m}$Sn was 179.30 mL/g. Similar to time parameter determination result, the effect of the amount of solid matrix on KD of $^{113}$Sn did not show the good pattern. Since the using of 250 mg of silica gel showed the optimum KD, this amount of solid matrix with agitation process within 25 minutes in 0.1 N of HCl were used in preparation stationary phase for separation process of $^{113m}$In from $^{113}$Sn via column chromatography technique.

**$^{113m}$In separation process using silica gel column chromatography**

$^{113m}$In and $^{113}$Sn separation process was performed by the silica gel column chromatography system. The column was rinsed by 0.05 N HCl and the bottom of the column was sealed with a rubber stopper and aluminum cap (**Figure 5a**). To the 250 mg of silica gel was added 1 mL of $^{113}$Sn in 0.1 N HCl. $^{113}$Sn solutions containing silica gel were homogenized by stirring for 25 minutes while heated at a temperature of 60 °C. Furthermore, the solution was added to a glass column (ø 0.5 cm x 7.5 cm) that the bottom of the column already contained activated carbon (**Figure 5a**), the aim of activated carbon addition was to absorb radionuclides $^{113}$Sn and $^{117m}$Sn. Then the column was eluted by 0.05 N HCl so we get the final results of the elution profile of $^{113m}InCl_3$ solution (**Figure 5b**). **Figure 5b** showed there was no $^{113}$Sn carried by HCl 0.05 N in all fraction. $^{113m}InCl_3$ radioactivity measurements using spectrometer-γ MCA-HPGe was obtained of 0.0388 μCi/mL ($^{117m}$Sn: 0.0034 μCi/mL), with the percent yield (separation) 81.83%. The most of $^{113m}$In was obtain in the first four fraction (4 mL elution) without $^{113}$Sn being carried. This results show

that the silica gel-based column chromatography combined with carbon active are potential to be used for separation of $^{113m}$In from irradiated natural tin.

### Radionuclide and radiochemical purity test of $^{113m}InCl_3$

***Radionuclide purity (KRN)***

$^{113m}InCl_3$ radionuclide purity was tested using gamma ray (γ) spectroscopy, Multi-Channel Analyzer (MCA) equipment wiyh a gamma ray (γ) spectrum pattern as shown in **Figure 6**. **Figure 6** indicates the characteristics of the γ-ray spectrum of $^{113m}InCl_3$ at 391 keV peak, which means that this production process can generate both $^{113m}$In products as $^{113m}InCl_3$. These results are relevant to the information described by Allan, et al. (2010), that the $^{113m}$In has gamma emission with an energy of 391 keV and a half-life of 1.7 hours. Data collection from gamma ray spectrum analysis $^{113m}InCl_3$ is summarized in **Table 1**. **Table 1** showed $^{113m}InCl_3$ radionuclide purity was 90.22%, but there were other radionuclides, $^{125}$Sb (0.47%) as the result of the core reaction of $^{124}$Sn (n, γ) $^{125}$Sb, and $^{117m}$Sn radionuclides (7.90%) as the core reaction product $^{116}$Sn (n, γ) $^{117}$Sn which has a natural isotopic abundance of 14% (Creech, Moynier, & Badullovich, 2017).

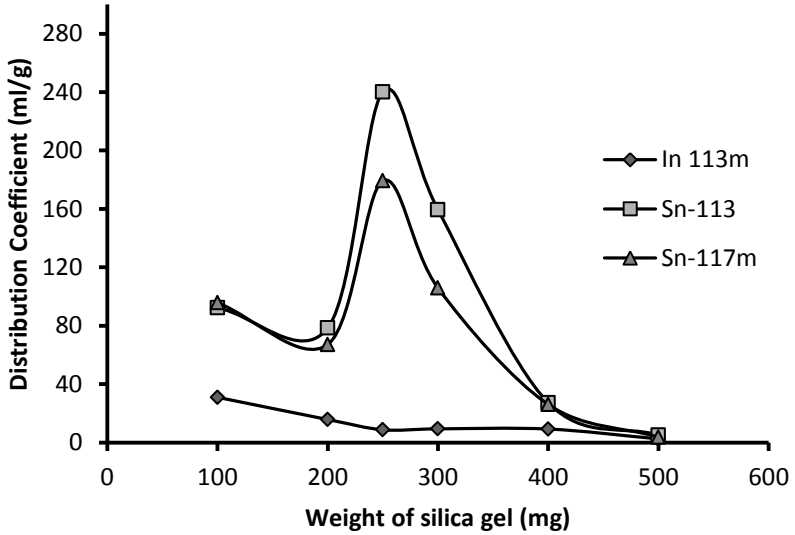
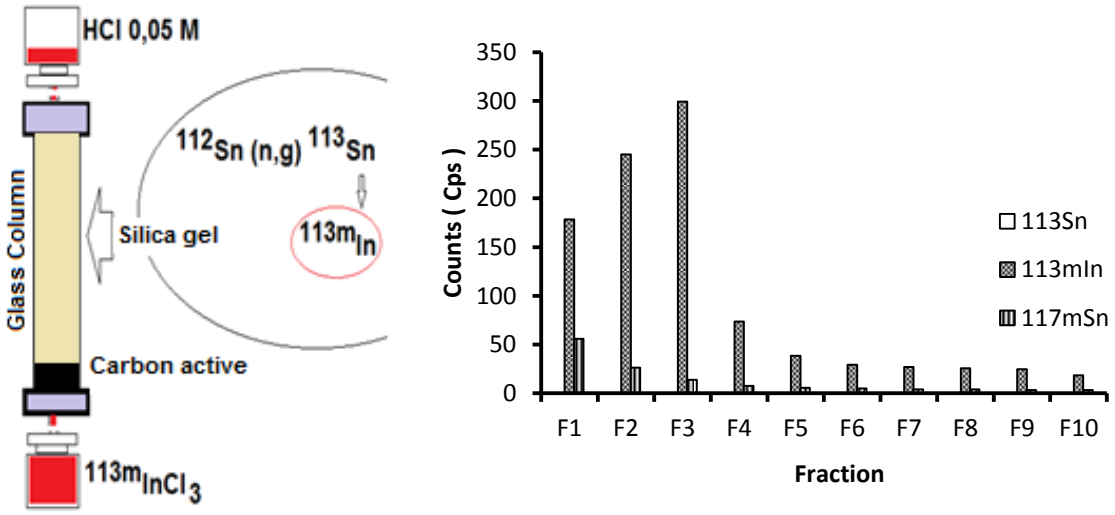



**Figure 4**. Coefficients distribution at weight of silica gel variation




(a) (b)

**Figure 5**. (a) silica gel column chromatography, (b) elution profile of $^{113m}InCl_3$ solution

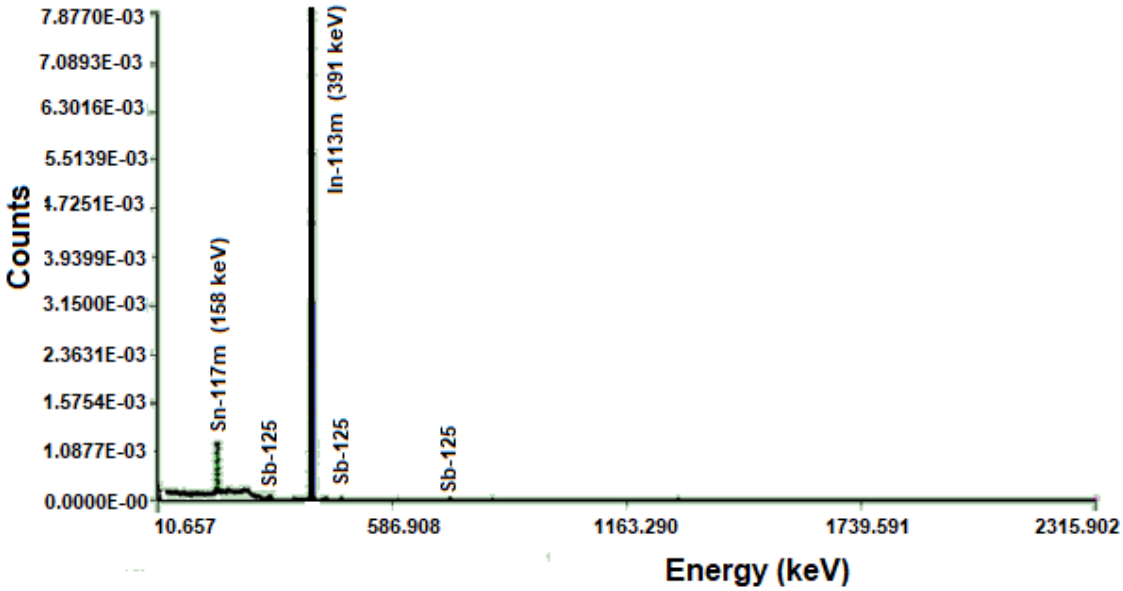



**Figure 6**. Gamma spectrum of 113mInCl3

**Table 1**. Analysis result of $^{113m}$In gamma spectrum

| Energy (keV) | Counts (cps) | Activity (µCi/mL) | Intensity (%) | Radionuclide | KRN (%) |
|---|---|---|---|---|---|
| 158,56 | 129,327 | 0,0034 | 2,11 | $^{117m}$Sn | 7,90 |
| 391,68 | 960,603 | 0,0388 | 64,97 | $^{113m}$In | 90,22 |
| 175,90 | 2,270 | 0,0002 | 6,84 | $^{125}$Sb | 0,47 |
| 427,87 | 2,553 | 0,0002 | 29,60 | $^{125}$Sb | 0,47 |
| 462,83 | 0,786 | 0,0002 | 10,49 | $^{125}$Sb | 0,47 |
| 635,71 | 0,100 | 0,0002 | 11,22 | $^{125}$Sb | 0,47 |
| TOTAL | | 0,043 | | | |

***Radiochemical Purity***

$^{113m}InCl_3$ radiochemical purity was determined by paper chromatography method by control the stationary phase compared to variable mobile phases. In the preparation of the solution of $^{113m}InCl_3$, $^{113m}$In was expected to be in the form of a single compound, namely $^{113m}InCl_3$. Paper chromatography systems were conducted in this study as summarized in **Table 2**. **Table 2** showed the chromatographic system that was used is Whatman 1 chromatography paper as the stationary phase and a solution mixture of 90% ethanol: 10% HCl 5 N as the mobile phase with an elution time of 95 minutes. Using of Whatman 31ET chromatograph system as the stationary phase and a solution mixture of 90% ethanol: 10% HCl 5 N as the mobile phase with the elution time of 148 minutes also can be used. Chromatogram sample with this system is shown in **Figure 7**. **Figure 7** showed the paper chromatography system which was used to determine the radiochemical purity $^{113m}$In are using Whatman 31ET paper as the stationary phase and solution mixture of 90% ethanol: 10% HCl 5 N as the mobile phase. In this study resulted in a chromatogram with Rf 0.7 to 0.8 as $^{113m}$In's peak with a radiochemical purity of 91.61 ± 0.29%. These results are relevant with study that used a mixture of 90% ethanol: 10% HCl as eluent with Rf 0.7 at Whatman 1 chromatography paper (Allan et al., 2010).

***Stability determination***

$^{113m}InCl_3$ solution stability in 3 days storage at room temperature showed that $^{113m}InCl_3$ remained clear and unchanged pH. While radiochemical stability was conducted by paper chromatography method by using mixtures of 90 % ethanol: 10 % HCl 5 N eluent. Radiochemical stability is showed in **Figure 8**. **Figure 8** showed the results of stability test, $^{113m}InCl_3$ solution was stable and the purity was maintained at 90.96 ± 1.20% at room temperature. According to Uccelli et al., (2018), radiochemical purity of radiopharmaceutical, including radioisotope solution, could be affected by physico-

chemical factor of the solution and environmental factor. Thus, by storing $InCl_3$ radioisotope solution in room temperature, it was sufficient to maintain radiochemical purity between 80% - 100% (Uccelli et al., 2018).

**Table 2**. Paper chromatography system variation

| No | Paper chromatography system | | $R_f$ $^{113}InCl_3$ | Elution time (minutes) | Remark |
|---|---|---|---|---|---|
| | Stationary phases | Mobile phases | | | |
| 1 | Whatman 1 chromatography paper | Acetone 85% | 0 | 69 | Cannot be used |
| 2 | Whatman 31ET chromatography paper | Acetone 85% | 0 | 34 | Cannot be used |
| 3. | Whatman 1 chromatography paper | 90 % Ethanol : 10 % HCl 5 N | 0,7 -0,8 | 95 | Can be used (optimum) |
| 4. | Whatman 31ET chromatography paper | 90 % Ethanol : 10 % HCl 5 N | 0,7 -0,8 | 148 | Can be used (optimum) |

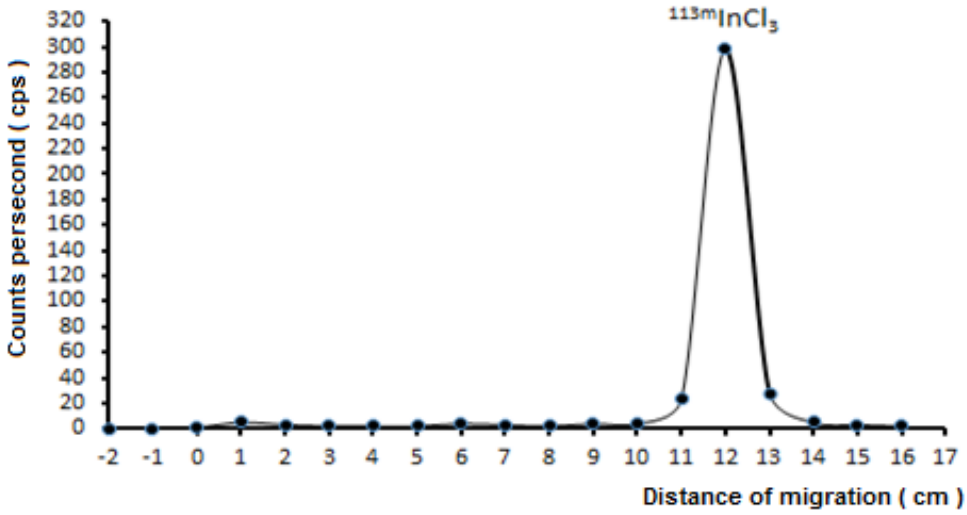



**Figure 7**. $^{113m}InCl_3$ in chromatography system solution 90% ethanol: 10% HCl 5 N as eluent and Whatman 31ET chromatography paper

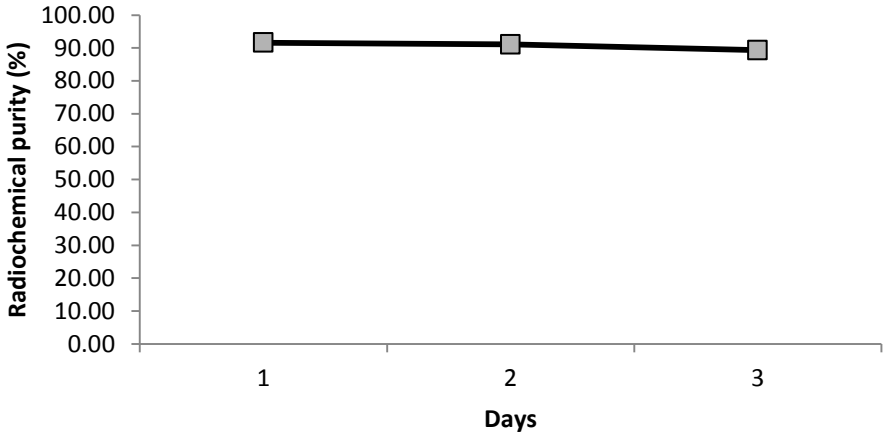



**Figure 8**. Radiochemical stability of $^{113m}InCl_3$ in mixture 90 % etanol : 10 % HCl eluent

## CONCLUSION

$^{113m}InCl_3$ radioisotopes were obtained by irradiation of a metal tin target material (Sn-metal) in the form of nature (Stano-112) by neutron activation at neutron flux of $1 \times 10^{14}$ $n.cm^{-2}.s^{-1}$ for 94.5 hours. Characteristics of $^{113}Sn$ radioisotopes in $^{113}SnCl_3$ chemical form were a clear solution, pH 2, the radioactive concentration of 0.0467 mCi/mL while theoretically was 0.0504 mCi/mL, obtained a yield of 92.65%. The optimum conditions of separation of radioisotopes $^{113}Sn$ / $^{113m}In$ were obtained at a solvent concentration of HCl 0.1 N, stirred for 25 minutes and a silica gel's weight of 250 mg. The obtained $^{113m}InCl_3$ was a clear solution, pH 2, the activity was 0.0388 μCi/mL with a yield of 81.83%, radionuclide purity of 90.22%, and radiochemical purity of 91.61 ± 0.29%.

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