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Preparation of [^{68}Ga]PSMA-11 for PET–CT imaging using a manual synthesis module and organic matrix based ^{68}Ge/^{68}Ga generator

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ABSTRACT

Introduction: [^{68}Ga]PSMA-11 is a relatively recently introduced radiopharmaceutical for PET–CT imaging of prostate cancer patients. The availability of ^{68}Ge/^{68}Ga generator and PSMA-11 ligand from commercial sources is facilitating the production of the radiopharmaceutical in-house. This paper describes our experience on the preparation of ~200 batches of [^{68}Ga]PSMA-11 for conducting PET–CT imaging in patients suspected/suffering from prostate cancer.

Methods: The radiosynthesis of [^{68}Ga]PSMA-11 was done in a hospital based nuclear medicine department using ^{68}Ge/^{68}Ga generator and a manual synthesis module, both supplied by Isotope Technologies Garching (ITG), Germany. The production involved the reaction of 5 μg (5.3 nmol) of PSMA-11 ligand in 1 ml of 0.25 M sodium acetate buffer with 4 ml of $^{68}GaCl_3$ in 0.05 M HCl for 5 min at 105 °C; followed by purification in a C18 cartridge and collection through a 0.22 μm pore size filter.

Results: The radiochemical yields obtained were consistently high, 93.19% ± 3.76%, and there was hardly any batch failure. The radiochemical purity of the product was >99% and the product was stable for over 2 h; however it was used in patients immediately after preparation. About 200 batches of [^{68}Ga]PSMA-11 were prepared during the period and more than 300 patients received the tracer during the 14 months of study. No adverse reaction was observed in any of the patients and the image qualities were consistent with literature reports.

Conclusion: [^{68}Ga]PSMA-11 with high radiochemical and radionuclidic purity is conveniently prepared by using a ^{68}Ge/^{68}Ga generator and manual synthesis module. The radiochemical yields are very high; and activity sufficient for 3–4 patients can be prepared in a single batch; multiple batches can be done on the same day and when needed after a gap of 1.5–2 h.

1. Introduction

Prostate cancer (PCa) is one of the major cancers affecting men and a major cause of morbidity and mortality in elderly men in the entire world [1]. A number of radiopharmaceuticals have been developed for positron emission tomography (PET) imaging of prostate cancer [2]. The introduction and wide practice of PET–CT imaging with [^{68}Ga] PSMA-11 (^{68}Ga labeled Glu-NH-CO-NH-Lys-(Ahx)-HBED-CC) are making a significant impact on the management of prostate cancer patients [3–5]. Gallium-68 ($T_{1/2}$ = 68 min) is a positron emitter produced by the decay of ^{68}Ge ($T_{1/2}$ = 271 days) and has ideal characteristics to be used as a radionuclide for PET–CT imaging [6–8]. The short half life of 68 min limits the radiation dose to the patients and at the same time it is sufficient for collecting clinically useful images with many of the targeting vectors. Gallium-68 decays to ^{68}Zn (inactive) 88.88% by positron emission and 11.11% by electron capture. The major advantage of ^{68}Ga is that it is available from a ^{68}Ge/^{68}Ga generator; the long half life ($T_{1/2}$ = 271 days) permits the generator to be used for several months, even up to a year [9]. The short half life of ^{68}Ga ($T_{1/2}$ = 68 min) allows multiple elution of the generator on the same day; thus enabling the preparation of several radiopharmaceuticals for different indications. Gallium-68 is emerging as one of the preferred radionuclides in PET–CT imaging.

The enzyme prostate membrane specific antigen (PSMA) is overexpressed in PCa cells and hence is an ideal target for molecular tracers [10,11]. PSMA being an enzyme, it can be targeted by using an inhibitor and several such inhibitors have been developed in the past; and many of these molecules have been used as pharmacophore for designing radiopharmaceuticals [12,13]. The inhibitors have structure mimicking the substrate molecule, N-acetyl-aspartyl-glutamate (NAAG); yet different that after binding with the enzyme further metabolism is stopped. Several dipeptide molecules, having uriedo (-NH-CO-NH-) structure and having high enzyme inhibition capacity as denoted by low IC_{50} (half maximal inhibitory concentration) have been developed as part of drug research and they are useful targeting vectors [14,15].

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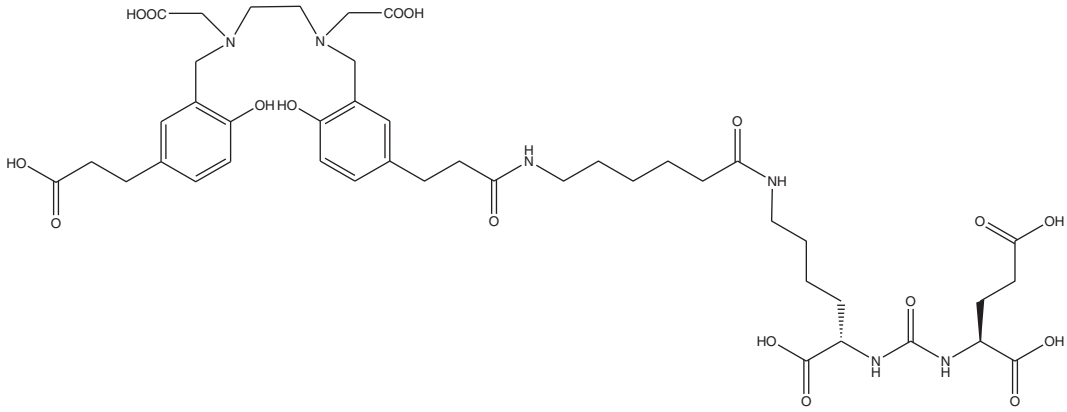

Fig. 1. PSMA-11 ligand; (Glu-^{CO}-Lys(Ahx)-HBED-C$^{C;}$ Glu-NH-CO-NH-Lys(Ahx)-HBED-CC); M. wt. 947 (product no. 9920.0000; ABX Advanced Compounds, Germany).

The ligand PSMA-11(Glu-CO-Lys(Ahx)-HBED-CC or Glu-NH-CO-NH-Lys(Ahx)-HBED-CC; also called DKFZ-PSMA-11) (Fig. 1) has been developed for radiolabeling with $^{68}GaCl_3$ [16]. There are several acronyms used for this product; the authors prefer to use PSMA-11, the acronym used by ABX Advance Chemical Compounds supplying the ligand. The pharmacophore of the ligand, PSMA-11, is Lys-uriedo-Glu, a PSMA inhibitor molecule; conjugated with the chelating agent HBED-CC (*N,N′*-bis[2-hydroxy-5-(carboxyethyl)benzyl]ethylenediamine-*N,N′*-diacetic acid) at the lysine end through a spacer molecule [17]. [^{68}Ga]PSMA-11 is used in several nuclear medicine departments across the world for imaging of not only prostate cancer but also several other indications [18,19].

While the preparation of [^{68}Ga]PSMA-11 is relatively easy, there are issues with radiation protection aspects as well as to ensure pharmaceutical purity to the final product. A 30 mCi open source of ^{68}Ga at one feet distance could give >1.5 mSv/h radiation dose and hence adequate safety measures need to be taken to limit the radiation dose to the radiopharmacist. A purification step at the end of synthesis is essential; in order to remove uncomplexed ^{68}Ga; more importantly ^{68}Ge coming as breakthrough product from the generator. Essential features to ensure good manufacturing practices (GMP) need to be built in the process to ensure the production of sterile and pyrogen free product safe for administration in patients. Most of the commercially available synthesis modules ensure the above features.

This paper summarizes our experience in the production of 200 batches of [^{68}Ga]PSMA-11 using an organic matrix immobilized on silica resin based ^{68}Ge/^{68}Ga generator and a manual synthesis module, both obtained from ITG Germany.

2. Materials

The ligand, PSMA-11 was purchased from ABX advanced biochemicals compounds. Hydrochloric acid (30%, Suprapur), sodium acetate (Suprapur), trisodium citrate dihydrate (Empura), ethanol (Emsure), water (Emsure) and pH paper (MColorpHast, pH 0–6) were from Merck, Germany. C18 light cartridges were purchased from Waters. 0.22 μm pore size syringe filters were of Millipore or Sartoriuos, Germany. All radioactivity measurements were done with Capintec CRC® 25 PET dose calibrator in ^{68}Ga-window mode.

iQS® Ga-68 fluidic labeling module [20] and the ^{68}Ge/^{68}Ga generator were procured from Isotope Technologies Graching (ITG), Germany. During the period of this study of 13 months, we used ^{68}Ge/^{68}Ga generators of 30 mCi (1110 MBq) and 20 mCi (740 MBq) ^{68}Ge activity.

3. Experimental

3.1. Preparation of reagents and pretesting procedures

The stock peptide, 500 μg of PSMA-11 (MW 947 Da) was dissolved in 5 mL of 0.25 M sodium acetate solution, and aliquots of 50 μL (5 μg, 5.28 nmol) or 100 μL (10 μg, 10.56 nmol) were dispensed in 1 mL Eppendorf tubes, closed and frozen at −20 °C. The product was found to be stable for over 6 months by which time the frozen aliquots were exhausted.

0.05 M HCl used for eluting the generator was prepared by adding 264 μL of 30% HCl to 50 mL of Emsure water in a 50 mL Corning tube. 0.25 M sodium acetate solution was prepared by dissolving 1.025 g of sodium acetate in 50 mL of Emsure water in a 50 mL Corning tube. As a practice, 250 μL of 0.25 M sodium acetate buffer is mixed with 1 mL of 0.05 M HCl; and the pH was ensured to be ~4.5 by using a pH paper. This step was done as an inbuilt quality assurance check to ensure the reaction mixture pH is ~4.5.

Ethanol (60%) used for eluting the product from the cartridge was prepared by mixing 20 mL of ethanol with 30 mL of Emsure water in a 50 mL corning tube. 70% ethanol solution is used for conditioning the cartridge; which was prepared by diluting 15 mL of ethanol with 35 mL of Emsure water.

A C18 cartridge is conditioned by passing 5 ml of 70% ethanol followed by passing 10 mL of Emsure water. A large sterile filter was also conditioned by passing 2 mL of 60% ethanol. The integrity of the filter is tested by passing 5 mL of air using a syringe to feel the pressure as well as to remove excess ethanol in it.

3.2. Elution of the generator

^{68}Ge/^{68}Ga generator is eluted with 4 mL of 0.05 M HCl and the activity measured in a Capintec CRC® 25 PET dose calibrator. Germanium-68 breakthrough was checked by measuring the activity of a completely decayed (~2 days) old generator eluted solution in a CRC® 25 PET dose calibrator. The dose measured is compared with the ^{68}Ga activity measured at the time of elution to check the ^{68}Ge breakthrough. By this method, breakthrough $>10^{-4}$ could be measured; as the dose calibrator has a sensitivity >1 μCi. Counting the decayed samples in a calibrated NaI(Tl) solid scintillation counter could have given breakthrough values more accurately; but was not considered essential. For synthesis of the radiopharmaceutical, the generator eluent is directly passed to the reactor through the fluidic system.

3.3. Preparation of [^{68}Ga]PSMA-11

3.3.1. Logics of the fluidic system

The wet chemistry was done in an iQS fluidic labeling module supplied by ITG, Germany; and a standard operating procedure (SOP) for the preparation of the radiopharmaceutical was also provided by the manufacturer. The iQS fluidic labeling module is designed such that features needed for taking care of GMP as well as radiation protection are inbuilt in the system. The essential features of the fluidic system are that it is a completely closed system and all the reagents needed for