RESEARCH ARTICLE OPEN ACCESS

Chemical Kinetic Of The Synthesis Of 2-(1-{6-[(2-2'-[^{18}F]Fluoroethoxyethyl)(Methyl)Amino]-2-Naphthyl}Ethylidene)Malononitrile([^{18}F]FEONM) As A Tau Protein Imaging Agent

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ABSTRACT
[^{18}F]FEONM is a new Alzheimer's disease imaging agent with the same core structure but has higher lipophilicity than that of 2-(1-{6-[(2-[^{18}F]fluoroethyl)(methyl)amino]-2-naphthyl}ethylidene)malononitrile ([^{18}F]FDDNP). In order to optimize the radiochemical yield of [^{18}F]FEONM, we have studied its radiofluorination kinetic at constant temperature and concentration. The results showed that the first order radiofluorination rate constant can be determined at optimized gap area condition. Although the first order reaction rate constant calculated by Gauss apodization function is an error function, it can be calculated with Welch apodization function and expressed as an analytical form. Consequently, the ideal size of a microfluidic plug flow reactor can be determined.

Keywords: [^{18}F]FEONM, radiofluorination kinetic, Welch apodization

Date of Submission: 30-03-2019 Date of acceptance: 13-04-2019

I. INTRODUCTION

Radiofluorination process is one of the major phase transfer reaction unit operation procedure for producing fluorine-18 labeledPET imaging agent. The radiofluorinationreaction takes place when both the phase transfer solvent and the catalyst are present at the same time at medium to high temperature [1,2]. [^{18}F]FEONM (Figure 1A) is a potent brain positron emission tomography (PET) imaging agent. It is designed to increase the lipophilicity of [^{18}F]FDDNP (Figure1B) [3-5] which is the first tau protein PET imaging radiopharmaceuticals for Alzheimer's disease and chronic traumatic encephalopathy. Preparing [^{18}F]FEONM, gas-liquid phase transfer reaction is operated at medium to high temperature in the presence of phase transfer catalyst and aprotic solvent. Several types of polymers and small molecules have been applied. Small molecule phase transfer catalysts are cheaper butless stable than those of polymer catalysts such as macrocyclic and macrobicyclic polydentate ligands [6]. In this study, we have studied and reported herein the chemical kinetic of [^{18}F]FEONMproduction in aprotic solvent (Acetonitrile) and the macrobicyclic polydentate polymer phase transfer catalyst K[^{18}F]/Kryptofix®-2.2.2 reaction system.

II. EXPERIMENT

2.1 Materials and Methods

All chemicals and reagents were obtained from commercial vendors and were used as received without further purification. Oxygen-18 water was purchased from Cambridge Isotope Laboratory. Aqueous [^{18}F]Fluoride was produced in our TR30/15 cyclotron (EBCO) via an ^{18}O(p, n)^{18}F

nuclear reaction. Precursor (TEON) and reference standard (FEON) were purchased from Huayi Isotopes Co.,kryptofix®-222 and potassium carbonate were purchased from Merck. Anion exchange resin was purchased from Bio-Rad (AG®1x8, hydroxide form, 200-400 mesh). Sep-Pak® Plus Alumina N cartridge was purchased from Waters. Membrane filter (MILLEX®) was purchased from Merck Millipore Ltd.Both semi-preparative HPLC column(Fortis® Part No. FPH 100905, 5μm diphenyl, size: 250x10mm) and analytical column (Cogent C18 100A 5μm, e Series, 150mm x 4.6mm, Cat. No. 78018-15P) were purchased from Inpac International Corporation. Waters 300E pump, 2489 UV detector (wavelength set at 254nm),Raytest Gabi gama detector and Bioscan AR-2000 Scanner were used for this study.

2.2 Preparation of [^{18}F]FEONM

The [^{18}F]FEONM (Scheme 1B) was synthesized with a Nuclear Interface synthesizer as described previously [4]. A mixture of the precursor (TEON,Scheme 1A, 7 mg) and K[^{18}F]/$K_{2.2.2}$ in 2-5 ml of acetonitrile was heated at 95℃ for 5 min. After cooling down to room temperature, the reaction mixture was flowing through an alumina cartridge (Sep-Pak® Plus Alum N) followed by purification with a semi-preparative HPLC (Fortis® Part No. FPH 100905, 5μm diphenyl, size: 250x10mm, mobile phase: 95% ethanol, flow rate: 1.6 ml/min). The peak corresponding tofinal product [^{18}F]FEONM was collected and passed through a 0.22μm membrane filter into a sterilized vial. The radiochemical yield of product was 20-30% at the end of synthesis (EOS) in a synthesis and purification time of 40 min from end of bombardment (EOB) (Scheme 1). The chemical and radiochemical purities of [^{18}F]FEONM were verified by both Radio-HPLC (Cogent C18 100A 5μm, e Series, 150mm x 4.6mm; mobile phase: 99.5% acetonitrile; flow rate: 0.3ml/min equipped with Waters 300E pump, Waters 2489 UV detector and Raytest Gabi gama detector) and Radio-TLC (Merck TLC Silica gel 60 F_{254} developed with95% acetonitrile and detected with a Bioscan AR-2000 Scanner) and was greater than 95% as compared to authentic sample.

2.3 Radiofluorination kinetic analysis

Radiofluorination reaction yield of [^{18}F]FEONM was calculated by measuring the activity of K[^{18}F] and radiofluorinated product with radio-TLC. In a previous study, when a mixture of 1 mg of precursor (Scheme 1A)and K[^{18}F]/$K_{2.2.2}$ in 1 ml of acetonitrilewas heated at 105℃ for 5 min, up to eighty percent of activity was identified on the region of interest (ROI) as the radiofluorinated compound(Scheme 1B). Value of gap function factor at this time, FG5, under this radiofluorinationcondition was 0.249 [4]. In this study, a mixture of 7 mg of precursor (Scheme 1A) and K[^{18}F]/$K_{2.2.2}$ in 2 ml of acetonitrilewas heated at 95℃ for various time points (5, 10, 30, 60, 90 and 120 minutes) and the radiofluorination rate was monitored with radio-HPLC.

III. RESULTS

The radiochemical yield of [^{18}F]FEONM at 5, 10, 30, 60, 90, 120 minutes are 84.54%, 87.14%, 92.23%, 92.48%, 93.07%, 93.35% (ROI, region of interest) and 78.16%, 81.08%, 86.21%, 85.12%, 86.38%, 87.88% (non-ROI). Since the value of ROI is increasing with reaction time, the radiochemical yield of [^{18}F]FEONM represents by ROI is closer to the real data of [^{18}F]FEONM activity. Both the decay corrected yield and non-decay corrected yield are reported according to the percentage of region of interest (ROI) or the percentage of total activity by the software WinScan Version 3.13. Radiofluorination kinetic curve of [^{18}F]FEONM at this almost optimized condition is listed as Figure2.

IV. DISCUSSION

Fluoride-18 is an ideal radionuclide to study the fluorination reaction kinetic in the reactors of chemical engineering factory. It provides all the hydrogen fluoride properties to form the strongest covalent bond with different precursors for making fluorinated compounds. In this study, the reaction mixture was spiked to measure the relationship of fluorinating yield to reaction time at several temperature and specific pressure as the reference standard curve for constructing a new kinetic model (Figure 2). This sampling pathway is similar to the well mixed chemical engineering reaction mixture when taking a single sample. From the reaction kinetic curve, we found that the reaction rate is first order in the beginning of the reaction. This is similar to most reactions in a batch reactor of chemical engineering. Here we named the end of the first order reaction time as t_a. After the first order reaction, it follows n order reaction; the highest yield is named at time t_b. This is also called the thermolysis or radiolysis time. The end of the reaction fully thermolysis time beyond time t_b is named t_c.

The first part of the radiofluorination reaction is considered to be a first order reaction, its region is from the beginning of the reaction to time t_a, and the second part and third part of the radiofluorination is n order reaction. Therefore the area can be described as the summation of integration form of Gauss or Welch apodization function. When we describe the n order phase transfer reaction yield curve from t_a to t_c by Gauss

or Welch apodization function, then t_a to t_b is similar to an integrated Gauss or Welch apodizationfunction and t_b to t_c is another Gauss or Welch apodization function without integration. This equation can be expressed as Gap function[4], named FG(1GA); 1: first order, GA: Gauss apodization.FG(1GD); GD: Gauss distribution.

$$FG = 1 - \int_0^t ROI/100t\,dt$$

$$FG(1GD) = 1 - \int_0^{t_a} kxdx - \iint_{t_a-1}^{t_b-1} \frac{1}{\sigma\sqrt{2\pi}} e^{-\frac{1}{2}\left(\frac{x-\mu}{\sigma}\right)^2} dx - \int_{t_b}^{t_c} \frac{1}{\sigma\sqrt{2\pi}} e^{-\frac{1}{2}\left(\frac{x-\mu}{\sigma}\right)^2} dx$$

$$FG(1GA) = 1 - \int_0^{t_a} kxdx - \iint_{t_a-1}^{t_b-1} exp\left(-\frac{x^2}{2\sigma^2}\right) dx - \int_{t_b}^{t_c} exp\left(-\frac{x^2}{2\sigma^2}\right) dx$$

The minimum area between ideal reaction and real reaction yield curve is at the differential form of Gap function when the result is zero. Although the solution of differentiating the integration form of Gauss apodization function is an error function,the rate constant can be determined with an analytical form by applying Welch apodization function and the length of microfluidic plug flow reactor can be expressed[7].

V. CONCLUSION

From the radiofluorination kinetic study by way of the research and development for [^{18}F]FEONM, the length of the microfluidic plug flow reactor can be determined by Gauss or Welch apodization gap function area calculus.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge Huang Ping-Yen and Chang Mao-Hsung for maintaining cyclotron's high vacuum system and constructing hydrogen fluoride-18 target; the PET center of Tri-Service General Hospital and National Taiwan University Hospital for inspiring this study.

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Figure 1: Structures of [^{18}F]FEONM(A) and [^{18}F]FDDNP(B).

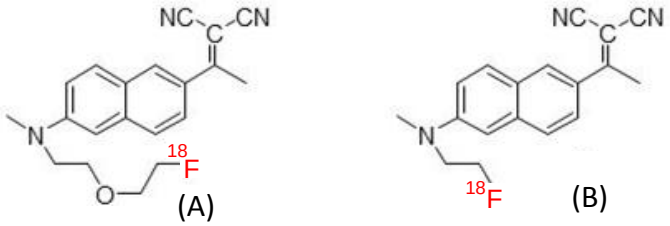
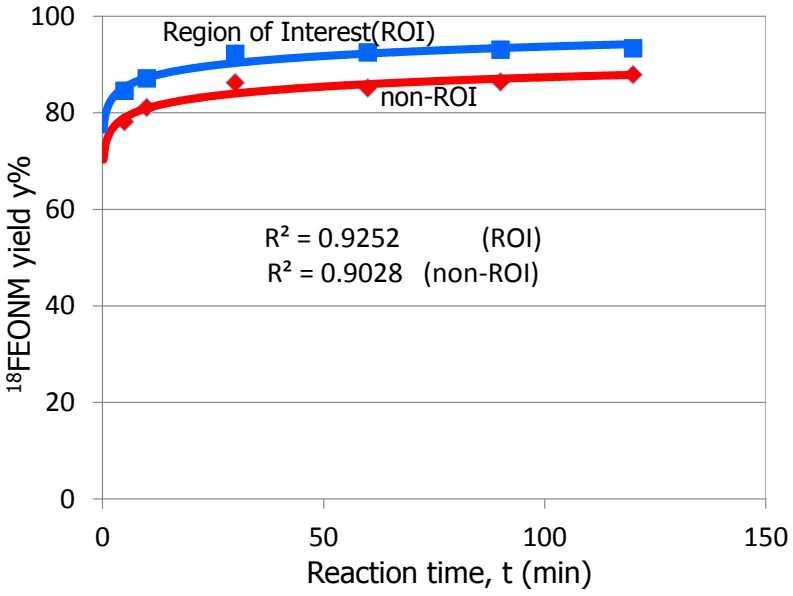
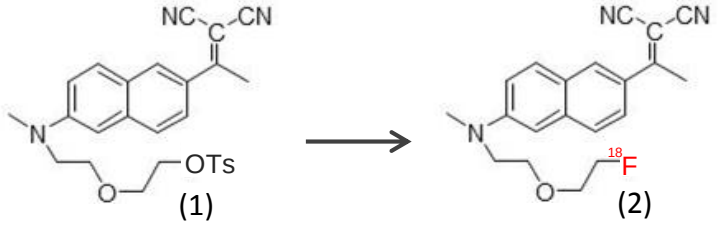


Figure2: Chemical kinetic of the synthesis of [^{18}F]FEONM. Region of Interest (ROI) was determined by radio-TLC activity ratio. R^2 show little different from ROI and non-ROI which represent the variance of activity distribution peaks.



Scheme 1: Synthesis of [^{18}F]FEONM.



Precursor TEON
2-(1-{6-[(2-2′-p-Toluenesulfonic acid-ethoxyethyl)(methyl) amino]-2-naphthyl}ethylidene) malononitrile

[^{18}F]FEONM
2-(1-{6-[(2-2′-[^{18}F]Fluoroethoxyethyl) (methyl)amino]-2-naphthyl}ethylidene) malononitrile

Chen Jenn-Tzong"Chemical Kinetic Of The Synthesis Of 2-(1-{6-[(2-2'-[18F]Fluoroethoxyethyl)(Methyl)Amino]-2-Naphthyl}Ethylidene)Malononitrile([^{18}F]FEONM) As A Tau Protein Imaging Agent" International Journal of Engineering Research and Applications (IJERA), Vol. 09, No.03, 2019, pp. 19-22