Automated synthesis of ^{18}F-labeled ligands for pre- and postsynaptic PET imaging of the dopaminergic system using IBA Synthera modules

V. Kramer[1)], M. Piel[2)], C. Elgueta[1)], S. Höhnemann[2)], A. Amaral[1)], M. Avila[1)], J. Ribbec[1)], E. Perez[3)], R. Pruzzo[4)], P. Chana[5)], C. Juri[6)], F. Rösch[2)] and H. Amaral[1,2)]

[1)]Positronpharma S.A., Santiago, Chile; [2)]Institut für Kernchemie, Johannes Gutenberg-Universität, Mainz, Germany; [3)]Instituto de química organica, Pontifica Universidad Catolica de Chile, Santiago, Chile [4)]Medicina Nuclear, Fundación Arturo Lopez Perez, Santiago, Chile; [5)]CETRAM Universidad de Santiago, Santiago, Chile [6)]Pontifica Universidad Católica de Chile, Santiago

Abstract

- Positron emission tomography for non invasive *in vivo*-diagnosis of DAT- and D_2/D_3-like receptor functions is considered to be a valuable tool for differential diagnosis and early detection of Parkinsons disease. [1]
- The aromatic amino acid decarboxylase (AADC), dopamin transporters (DAT) and vesicular monoamine transporters (VMAT2) are valuable targets for preclinical detection of PD.
- Out of those, DAT seems to be the most sensitive target in early phases [1] (see Figure 1)

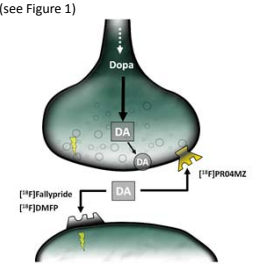
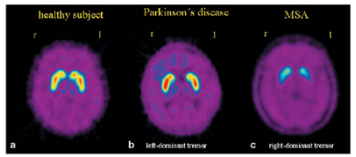


Figure 1: Dopaminergic synapse and radioligands for pre- and postsynaptic imaging.

- Postsynaptic D_2/D_3-receptor ligands like Fallypride or DMFP have been used successfully for receptor quantification, occupancy studies and for differential diagnosis of idiopathic and atypical parkinson syndroms [2,3] (see Figure 2).



- **Figure 2:** Differential diagnosis of parkinsonism with **[^{18}F]DMFP**
- Aim of this study was to provide a fully automated synthesis for routine application of ^{18}F-Fallypride, ^{18}F-DMFP and ^{18}F-PR04.MZ, a new selective and high affine DAT ligand for PET imaging, using the IBA Synthera platform.

Materials & Methods

- ^{18}F-Fluoride was produced by ^{18}O(p,n)^{18}F-reaction (Cyclone 18/9 IBA) and transferred to a spare vial.
- ^{18}F-PR04.MZ, ^{18}F-Fallypride and ^{18}F-DMFP were labeled by direct nucleophilic fluorination of corresponding mesyl and tosyl precursors (see Figure 3)

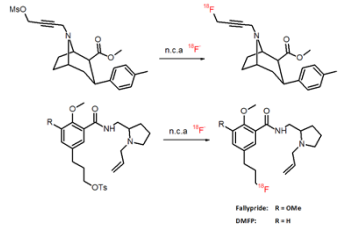


Figure 3: Radiosynthesis of **[^{18}F]PR04.MZ**, **[^{18}F]FP** and **[^{18}F]DMFP** .

- Known labeling conditions from the literature were adapted to the IBA Synthera plattform[2,4]. The different reaction steps were optimized and saved in a script for automatic control.
- Labeling precursors, consumables and IFPs were purchased from ABX and used without any modifications.
- For the labeling step, standard IFPs for FDG synthesis were used.
- Purification was reached by HPLC, solid phase extraction and sterile filtration. For postprocessing standard IFPs for alkylation were used without any modifications. Experimental setup is shown in Figure 4:

Figure 4: Experimental setup for labeling, purification and post-processing.

- Depending on the radioligand, different consumables and reaction conditions were used (see Table 1).

Table 1: Reaction and purification conditions:

	PR04.MZ	Fallypride	DMFP
Vial 1	15 mg K222, 15 µmol K_2CO_3	15 mg K222, 15 µmol K_2CO_3	15 mg K222, 15 µmol K_2CO_3
Vial 2	5 mg Mesyl-PR04MZ	5 mg Tosyl-FP	5 mg Tosyl-DMFP
Reaction-temperature	88 °C	88 °C	88 °C
Reaction-time	10 min.	20 min.	20 min.
Vial 3	Water/MeCN 1:1	Water/MeCN 1:1	10 % H_3PO_4
HPLC-solvent	4 mL/min MeCN/Solvent A 60 : 40	4 mL/min MeCN/Solvent A 30 : 70	4 mL/min MeCN/Solvent A 30 : 70
HPLC-column	Phenomenex Luna C18 10 x 250 mm	Phenomenex Luna C18 10 x 250 mm	Phenomenex Luna C8 10 x 250 mm
Retention time Product	15.5 min	16 min	13.5 min
HPLC-dilution	45 mL water	45 mL water	45 mL 0.15 M Na_2HPO_4-Buffer
Vial 4/5/6	2 mL water / 1 mL ethanol / 9 mL 0.9 % sodium chloride		

Results

- After labeling, crude product solution was successfully transferred to HPLC-loop using a 0.22 µm nylon filter.
- Thereby about 15 % of activity was retained in the filter.
- The product retained on C18 cartrige after HPLC-purification was successfully eluted with 1 mL ethanol. Thereby around 10 % were still retained on cartrige (see table 2).
- [^{18}F]PR04.MZ, [^{18}F]Fallypride and [^{18}F]DMFP were obtained as injectable solution (10 % ethanol in 0.9% sodium chloride.
- Full QC according to guidelines by European Pharmacopoeia was performed and passed and radiochemical purity was in the range of 96-98 % for all ligands.
- From starting activities of 9-35 GBq, [^{18}F]PR04.MZ, [^{18}F]Fallypride and [^{18}F]DMFP were obtained in high RCYs of 41±7, 37±9 and 35±4 %, respectively.
- Total synthesis time for all ligands was in the range of 57-68 minutes and specific activities were in the range of 47-480 GBq/µmol (mean: 110 GBq/µmol).

Table 2: Typical activity distribution for a synthesis of **[^{18}F]DMFP**

	Activity / mCi	Time	RCY / % (d.c.)
Start synthesis	364.0	0	100
QMA	8.23	78	3.7
Nylon filter	32.7	89	15.7
Alumina N	0.6	90	0.3
C18	25.3	79	11.4
Steril filter	6.89	72	3.0
Product	88.7	68	37.4

Summary & Outlook

- A fully automated synthesis of 3 different ligands for pre- and postsynaptic imaging of the dopaminergic system was established for the IBA Synthera platform.
- A new automated method for post-processing was developed using IFPs for alkylation.
- [^{18}F]PR04.MZ, [^{18}F]Fallypride and [^{18}F]DMFP were obtained in high radiochemical yields and purities.
- QC in accordance to guidelines made my the European Pharmacopoeia was passed for all ligands.
- The RCYs obtained on IBA Synthera modules by the described methods are high, but still lower than those published in the literature. Therefore especially loop-loading and elution of the C18 cartrige have to be optimized in the near future.
- Specific activities obtained by this method are sufficient for clinical application and injected mass of tracer would be < 1 µg. Nevertheless there ist still a need for further optimization of the specific activity.

References

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vkramer@positronpharma.cl http://www.positronpharma.com

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