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SYNTHESIS OF 3-[^{18}F]-FLUOROMETHYL-TCP[1], A POTENTIAL TOOL FOR PET STUDY OF THE NMDA RECEPTOR CHANNEL COMPLEX.

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ABSTRACT

In an attempt to visualize the NMDA glutamatergic receptors and after checking the biological activity of the cold 3-fluoromethyl-TCP **3**, 3-[^{18}F]-fluoro-methyl-TCP **4** was synthesized by a nucleophilic substitution of 3-bromomethyl-TCP **5** with [^{18}F$^-$].

KEYWORDS: ^{18}F, PET, NMDA receptor, TCP[1] or 1-[1-(2-thienyl)-cyclohexyl]-piperidine.

INTRODUCTION

The very important role of excitatory amino acids in the pathology of a number of neurodegenerative disorders led us to look for a radioactive ligand capable to visualize the receptors by the positron emission tomography (PET).
TCP, the thienyl analog of phencyclidine (PCP), is now well known to specifically bind to the NMDA gated ionic channel (1,7,9,10). Several labelled compounds ([^{3}H] and [^{18}F] analogs) have been synthesized in order to evaluate the biodistribution of these analogs (2,4,5,8). But, neither *in vivo* results in the baboon brain (with [^{18}F] analog), nor *ex vivo* studies (with [^{3}H]-TCP) have been reported (3). Recently, *ex vivo* studies of mouse brain (6) showed that cerebral uptake was specific and that regional distribution was in agreement with previous studies and encouraging for their use in PET studies.
We report here the synthesis of the cold fluoro compound via the triflate pathway method and the radiosynthesis by a nucleophilic substitution.

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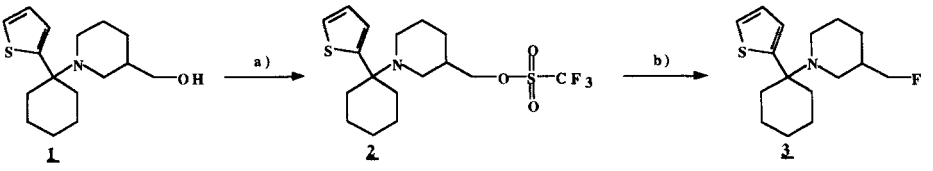
SCHEME 1 : Triflate pathway method



1 2 3

3-HYDROXYMETHYL-TCP 3-(TRIFLUOROMETHANE-SULFONYLOXY)METHYL-TCP 3-FLUOROMETHYL-TCP

a) $(CF_3SO_2)O, CH_2Cl_2, C_5H_5N$ b) KF, K222, CH_3CN

CHEMISTRY

The starting material, 3-hydroxymethyl-1-[1-(2-thienyl)-cyclohexyl]-piperidine **1**, was selected as a good chemical precursor to obtain the 3-fluoromethyl-TCP **3** via the classical triflate pathway (SCHEME 1); the reasons for this choice were:

- elimination product was obtained by the triflate pathway when the hydroxy group was directly bound to the piperidine ring (5).
- 3-substituted analogs have a very good affinity for the receptors.

The synthesis of 3-(trifluoromethanesulfonyloxy)-methyl-1-[1-(2-thienyl)-cyclohexyl]-piperidine **2** was carried out in anhydrous methylene chloride with trifluoromethanesulfonic anhydride in the presence of pyridine with a 47% yield. The cold fluoro-compound **3** was then obtained with a 61 % yield by heating in acetonitrile the triflate **2** in the presence of potassium fluoride and kryptofix. In a second run, compound **3** was obtained in one step with about the same global yield.

TABLE 1 : Parameter studies for radiolabelling of 3-bromoTCP

TCP-CH_2-Br mg	K 222 mg	K_2CO_3 mg	Solvent	Time min	Temperature °C	*Yield %
base 3.68	14.7	3	CH_3CN	10	85	2.6
base 5.92	15.2	3	DMSO	10	120	12.85
base 4.61	15.3	3.2	DMSO	15	125	30.8
HCl 4.8	14.6	2.5	DMSO	10	120	-

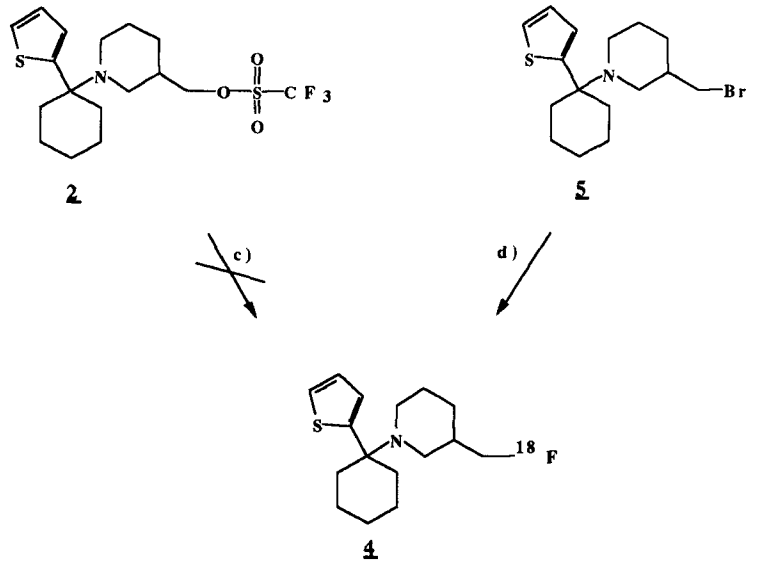
* yield corrected.

Neither 3-[^{18}F]-fluoro-methyl-TCP **4**, nor even a trace of 3-fluoromethyl-TCP **3** were obtained in the radiosynthesis via the triflate pathway method. After unsuccessful attempts to obtain 3-[^{18}F]-fluoro-methyl-TCP **4**, the radiosynthesis was carried out by the nucleophilic substitution of 3-bromo-methyl-TCP **5** (SCHEME 2) with [$^{18}F^-$] produced by a (p,n)

reaction with 16 Mev protons on 50% [^{18}O] enriched water (1.3 ml). After one hour, irradiation was stopped and the solution was transferred into a tube containing K-222 and K_2CO_3 and then evaporated to dryness under a stream of nitrogen. Then after dissolution of [^{18}F]-KF in an appropriate solvent, the solution was dropwise added into a vial containing the 3-bromomethyl-TCP **5**.

Parameters of the radiosynthesis (solvent, time and temperature) were studied in order to improve the radiochemical yield (TABLE 1). The best conditions found were DMSO as a solvent and 15 minutes heating at 125°C which gave a 30% yield (corrected for decay). A large amount of impurities were formed under these drastic conditions (Figure 1). Two reverse phase HPLC purifications were necessary to obtain about 10 mCi of pure 3-[^{18}F]-fluoro-methyl-TCP **4**, with a specific radioactivity of 728mCi/μmol.

SCHEME 2 : Radiosynthesis of 3-[^{18}F]-fluoro-methyl-TCP 4

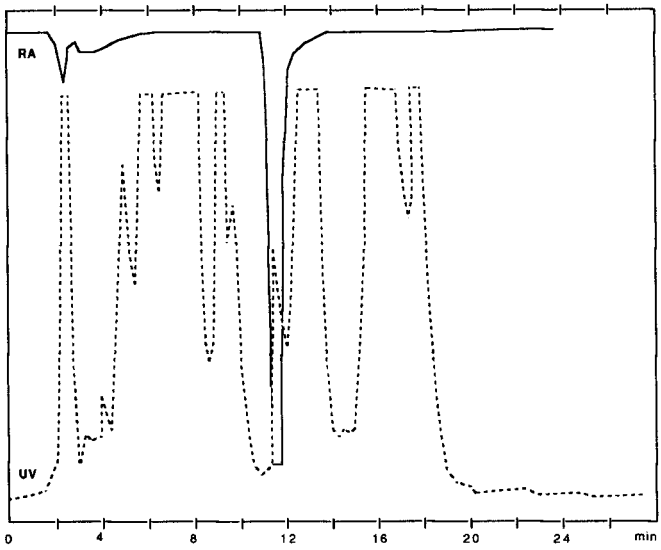


3-[^{18}F]-FLUOROMETHYL-TCP

c) K ^{18}F, K 2 2 2, C H 3 C N d) K ^{18}F, K 2 2 2, D.M.S O

In conclusion, the radiosynthesis appears to be useful for *in vivo* studies. The preliminary PET results on baboon brain with 3-[^{18}F]-fluoro-methyl-TCP **4** were presented as a poster at the IXTH International Symposium on Radiopharmaceutical Chemistry (PARIS, 6-10 april 1992). The abstract of our presentation will be pusblished in the Proceedings of this Meeting.

Figure 1 : Chromatogram of the first HPLC purification of 3-[^{18}F]-fluoro-methyl-TCP 4



EXPERIMENTAL

Compounds 1 and 4 were a gift of Dr KAMENKA J.M.. All commercial chemicals were used without purification and solvents were freshly distilled.
NMR spectra were recorded on a Brucker AM 250 and mass spectra were obtained using a Nermag R10-10 instrument at 70ev. U.V spectra were recorded using a Kontron (Uvikon 810) spectrometer. Mass, N.M.R. and U.V. analysis were in agreement with the assigned structures.
HPLC analysis and purifications were performed using a Waters system(501 pump) on μ Bondapak C 18 column (30cm, 7.8mm O.D, 10μm) using ethanol/water/triethylamine (70/30/0.05 : v/v/v) with U.V detection at 235 nm (Waters 484).

3-(trifluoromethanesulfonyloxy)-methyl-1-[1-(2-thienyl)-cyclohexyl]-piperidine : 2.
To a solution of 0.32 mmol (90mg) of 1 in 25 ml of anhydrous methylene chloride, 65 μl of anhydrous pyridine (63.7mg, 0.85mmol) were added. To this mixture, cooled in an ice-ethanol bath, was then added 2.13eq of trifluoromethanesulfonic anhydride(192mg, 0.68mmol) in 0.7 ml of methylene chloride. After two hours, a precipitate was observed. The crude material was extracted with water(2x100ml), the organic layers were dried over sodium sulfate and concentrated under vacuum to give 62.7mg of a colorless oil with 47.6% yield.
M.S. (DCI/NH_3): (M+1) = 412.

3-fluoromethyl-1-[1-(2-thienyl)-cyclohexyl]-piperidine : 3-fluoro-methyl-TCP : 3.

25mg of potassium fluoride (0.43mmol, 1.3eq) and 257.5mg of Kryptofix (0.68mmol, 2.1eq) were added to a solution of 2 (0.33mmol, 136mg) in acetonitrile and the mixture was refluxed for 20 hours at 80°C under an atmosphere of N_2. After evaporation to dryness, the crude material was dissolved with methylene chloride, extracted and purified by flash chromatography on silicagel. 55.4mg of a pure colorless oil 3 were obtained (61% yield). The hydrochloride salt was prepared with a solution of hydrogen chloride in ether.

HPLC : retention time : 11.5mn (4ml/mn).

M.S. (DCI/NH_3): (M+1) = 282.

1H R.M.N.(CD_2Cl_2) : δ: 0.8-3 (m, 19H), 4.27 (d,2H,J-CH_2F =57Hz), 6.85(s,1H), 7 (t,1H), 7.2 (s, 1H).

U.V.(H_2O): λ max = 229 nm (ε = 5216), λ min = 203 nm (ε = 1436).

3-[^{18}F]-fluoromethyl-1-[1-(2-thienyl)-cyclohexyl]-piperidine : 3-[^{18}F]-fluoro-methyl-TCP : 4.

262mCi of n.c.a K^{18}F / K 222 in 0.6 ml of dry D.M.S.O was added to about 14μmol (5mg) of free base of 5 in a pyrex vial. After fitted with a septum plug, the vial was heated at 120°C for 15min. The mixture was then dropped in 10ml of water For elimination of unreacted [^{18}F]-KF, the crude material was poured onto a C18 Sep-Pack cartridge; the radioactive compound 4 was then eluted on a silica Sep-Pack cartridge with methylene chloride. From this cartridge 3-[^{18}F]-fluoro-methyl-TCP 4 was eluted with methylene chloride containing few% of triethylamine.

After two HPLC purifications 10 mCi of 3-[^{18}F]-fluoro-methyl-TCP 4 were obtained with a specific radioactivity of 760 mCi/μmol.

One hundred and twenty six minutes were necessary to obtain chemically and radiochemically pure 4.

REFERENCES

1. Allaoua H. and Chicheportiche R.- Eur. J. Pharmacol., 163: 327 (1989).

2. De Costa B.R., and Mattson M.V.- J. Label. Comp. Radiopharm. 29: 63 (1991).

3.Ferrarese C., Guidotti A., Costa E., Rice K.C., De Costa B.R, Miletich R.S., and Di Chiro G.- J. Cereb. Blood Flow Metab., 11: S873(1991).

4. Kiesewetter D.O., Rice K.C., Mattson M.V., Finn R.D.- J. Label. Comp. Radiopharm. 27: 277(1989).

5. Maeda M., Tsukiyama S., Fukumara T., Orita K., and Kojima M.- Appl. Radiat. Isot. 42: 563(1991).

6. Maurice T., and Vignon J. - J.Neurosci. Res. 26: 377(1990).

7. Simpson M.D.C., Royston M.C., Deakin J.F.W., Cross A.J., Mann D.M.A., and Slater P. - Brain Res. 426: 76(1988).

8. Thurkauf A., De Costa B.R., Kiesewetter D.O., Jacobson A.E., and Rice K.C. - J. Label. Comp. Radiopharm. 27: 653(1989).

9. Vignon J., Chicheportiche R., Chicheportiche M, Kamenka J.M., Geneste P. and Lazdunski M.- Brain Res., 280, 194(1983).

10. Vignon J., Privat A., Chaudieu I., Thierry A., Kamenka J.M. and Chicheportiche R. Brain Research, 378, 133(1986).