PRECURSOR SYNTHESIS AND RADIOLABELLING OF THE DOPAMINE D₂ RECEPTOR LIGAND [¹¹C]RACLOPRIDE FROM [¹¹C]METHYL TRIFLATE

Oliver LANGER[1,3*], Kjell NÅGREN[2], Frédéric DOLLE[3], Camilla LUNDKVIST[1], Johan SANDELL[1], Carl-Gunnar SWAHN[1], Françoise VAUFREY[3], Christian CROUZEL[3], Bernard MAZIERE[3] and Christer HALLDIN[1]

[1]Karolinska Institutet, Department of Clinical Neuroscience, Psychiatry Section, Karolinska Hospital, S-17176 Stockholm, Sweden.
[2]Turku PET Center, Radiopharmaceutical Chemistry Laboratory, Porthaninkatu 3, FIN-20500 Turku, Finland.
[3]Service Hospitalier Frédéric Joliot, Département de Recherche Médicale, CEA, 4 place du Général Leclerc, F-91406 Orsay, France.

Summary

Desmethyl-raclopride was synthesized *via* a straightforward, three-step synthetic approach and used for the preparation of [¹¹C]raclopride from [¹¹C]methyl triflate. Conditions for the radiolabelling were optimized to obtain a simple and reproducible procedure suitable for automation. [¹¹C]Raclopride was prepared with an average radiochemical yield of 55-65% (decay corrected, based on starting [¹¹C]methyl triflate) in a total synthesis time (including purification and formulation of product) of 35 min. The radiolabelling procedure used significantly less precursor, avoided the use of DMSO, and was shorter compared to the standard radiolabelling procedure with [¹¹C]methyl iodide.

Key words : [¹¹C]methyl triflate, [¹¹C]raclopride, dopamine D₂ receptor antagonist

Introduction

[¹¹C]Methyl triflate ([¹¹C]MT) (1) has been shown to be a highly reactive alternative to [¹¹C]methyl iodide ([¹¹C]MI) (2-3) as a radiolabelled reagent in the synthesis of radioligands for positron emission tomography (PET). Different types of reactions such as ¹¹C-methylations of amine, phenol and thiol functions as well as amide and carboxylic acid functions have been performed (4-7). It has been demonstrated that [¹¹C]MT is compatible with low concentrations of aqueous bases such as sodium hydroxide and tetrabutylammonium hydroxide. It has also been found that reactions are completed after trapping of [¹¹C]MT, as additional reaction time with heating does not increase the radiochemical yield (6-7).

Raclopride (Fig. 1) is a selective dopamine D_2/D_3 receptor antagonist with an affinity of 1-2 nM (8). [^{11}C]Raclopride has during recent years become the "golden standard" for PET examination of striatal dopamine D_2 receptors and is used extensively by PET Centers in Europe, Japan and USA. The main areas of interest are human PET studies of the involvement of striatal dopamine receptors in neuropsychiatric diseases and drug abuse. Additionally, for the development of drugs used in the treatment of neuropsychiatric disorders receptor occupancy studies with [^{11}C]raclopride and PET have been shown to be useful (9).

The standard radiolabelling procedure for [^{11}C]raclopride is [^{11}C]MI alkylation of the precursor-phenolate - generated *in situ* from the corresponding hydrobromide salt of the O-des-methyl-compound NCQ 259 (previously available from ASTRA, Södertälje, Sweden) -, using aqueous NaOH in dimethylsulfoxide (DMSO) and heating at 80°C for 3 min. (10). Radiochemical yields and specific radioactivities are in most cases sufficiently high for PET-studies in human. However, the standard synthetic procedure occasionally fails to give the desired product in normal yields. In addition to that the viscosity of the reaction mixture makes automation difficult. There is thus a need for a simplified and reproducible radiolabelling procedure which is suited for automation of the entire production.

The benzamide analogues FLB 457 and epidepride (Fig. 1), which are structurally closely related to raclopride, have in their ^{11}C-labelled form been used for studies of extrastriatal dopamine D_2 receptors. These radioligands have been prepared in an automatic synthetic procedure from [^{11}C]MT (6,11). As [^{11}C]MT presumably reacts with the bromide ion of the hydrobromide salt of the precursor to afford the unwanted volatile by-product labelled methyl bromide the free base of the precursor has been used (6,11).

In this work we reinvestigate the synthesis of raclopride and its corresponding desmethyl-precursor (12-13) and present a new, simplified radiolabelling procedure of [^{11}C]raclopride with [^{11}C]MT.

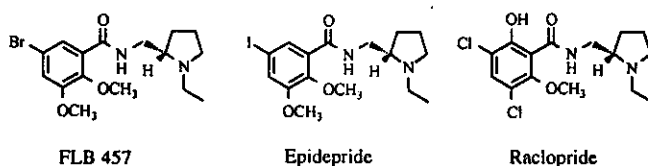
FLB 457   Epidepride   Raclopride

Figure 1 : Structures of the dopamine D_2 receptor antagonists FLB 457, epidepride and raclopride.

Results and Discussion

Chemistry

Raclopride (**1**) was synthesized with slight modifications according to a method reported in the literature from commercially available 3,5-dichloro-2,6-dimethoxybenzoic acid (**2**) and (S)-(-)-2-aminomethyl-1-ethylpyrrolidine (**12**) (Fig. 2). A similar synthetic pathway has been recently employed for the synthesis of the structurally closely related benzamide epidepride (11).
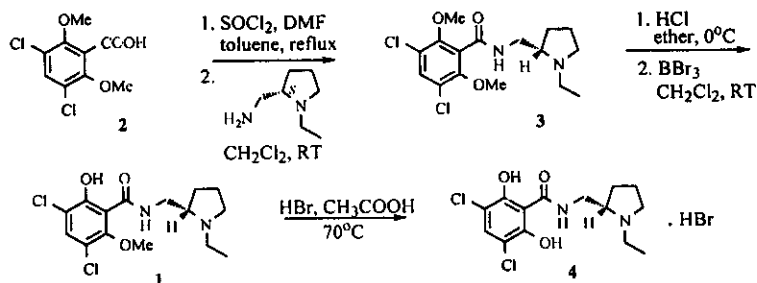
Figure 2 : Synthesis of raclopride (1) and desmethyl-raclopride (4).

The substituted benzoic acid **2** was converted into its acid chloride with $SOCl_2$ in toluene at reflux containing traces of DMF followed by reaction with (S)-(-)-2-aminomethyl-1-ethylpyrrolidine in dichloromethane at room temperature to obtain the benzamide **3** in excellent yield (95%) (Fig. 2). Compound **3** was then converted into its hydrochloride salt (using a solution of HCl in diethylether) and mono-demethylated with boron tribromide in dichloromethane to afford raclopride (**1**) in high yield (91%) (Fig. 2). As confirmed by NMR analysis and mass spectrometry, only one methoxy group had been cleaved while the other one had been left unaffected, even when an excess of boron tribromide had been used. Cleavage of the remaining methoxy group was done as reported in the literature by the use of HBr in acetic acid and gave the carbon-11 radiolabelling precursor **4** (nor-raclopride) in the form of its hydrobromide salt in around 50% yield (13) (Fig. 2).

Compound **4** was meant to be used for radiolabelling with [^{11}C]MT under basic conditions. As [^{11}C]MT presumably reacts with the bromide ion to afford the unwanted volatile by-product methyl bromide, the amine **4** had to be liberated from its hydrobromide before being used for the radiolabelling reaction. This was done either by extracting an aliquot of the hydrobromide from water made alkaline with diluted ammonia (pH 8-9) or by precipitation of the bromide ion with silver acetate. The free amine **4** had the appearance of a fine yellowish powder, that could be stored at -20°C without any degradation.

Radiochemistry

Reaction of desmethyl-compound **4** hydrobromide with [^{11}C]MI in DMSO/aqueous NaOH employing the standard conditions that have so far been used in our laboratory for the routine synthesis of [^{11}C]raclopride ([^{11}C]**1**) (10) regularly yielded [^{11}C]**1** in radiochemical yields of 50-60% (decay corrected and calculated from inital [^{11}C]MI) (Fig. 6A). On the other hand the reaction of **4** hydrobromide with [^{11}C]MT only afforded traces of [^{11}C]raclopride and - as expected - [^{11}C]methyl bromide as the only radiolabelled product (Fig. 6B).

Thus, two different approaches were investigated for removal of the bromide ion from the precursor : (1) treatment of the precursor salt with silver acetate (AgOAc) or silver triflate (AgOTf) for precipitation of the bromide ion or (2) liberation of the free base of the precursor by extraction from diluted aqueous ammonia.

Surprisingly, the radiochemical yield in the silver acetate experiments increased when excess amounts of silver acetate were used to precipitate the bromide ion (Fig. 3). The use of 4.5 eq of AgOAc resulted in an average radiochemical yield of 50-60% which can be compared to the [11C]MI experiments (Fig. 3). A radioactive by-product was observed on HPLC (Fig. 6C), which we identified as [11C]methyl acetate by coelution with authentic unlabelled methyl acetate.
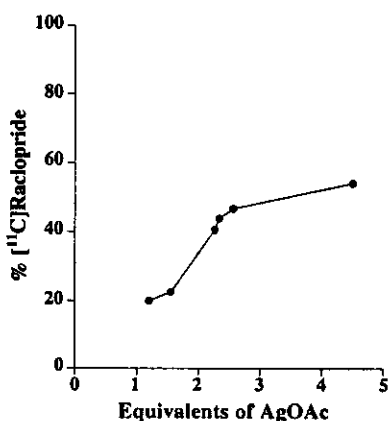

Figure 3 : [11C]Raclopride from [11C]MT : AgOAc precipitation of bromide from precursor salt.

We therefore tried to use AgOTf for the bromide-precipitation as this reagent would afford [11C]MT itself as a possible radiolabelled side-product, thus presumably leading to an increase in the radiochemical yield. As predicted no radiolabelled methyl acetate could be detected (Fig. 6D). However, surprisingly enough, the average radiochemical yield did not significantly improve compared to the use of AgOAc.

We wanted to simplify the above described procedures by using directly the free base of the precursor for the radiolabelling. This was further supported by the fact that the free base was relatively stable under normal storage conditions (-20°C) and did not have to be freshly prepared from the hydrobromide salt for each experiment. The structurally closely related benzamide FLB 457 has previously been radiolabelled with [11C]MT (6). It has been demonstrated that neither the use of excess base nor additional reaction time and heating after trapping of [11C]MT were necessary to increase radiochemical yields (6-7). We used for our initial experiments similar conditions as those reported for FLB 457 : 1 mg (3 µmol) of the desmethyl-precursor 4 (free base) dissolved in acetone (0.2 mL) and aqueous NaOH (1 eq). The trapping of [11C]MT was performed at room temperature (Fig. 4).

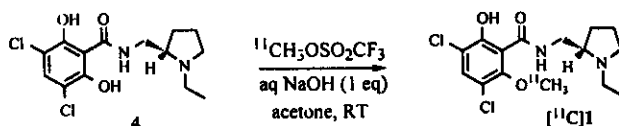Figure 4 : Synthesis of [^{11}C]raclopride from [^{11}C]MT.

Using these conditions [^{11}C]raclopride was obtained in an average radiochemical yield of 55-65% which was comparable to the above described "precipitation method" and the standard conditions with [^{11}C]MI.

In order to be able to economize on the radiolabelling precursor **4** we performed a series of experiments with continuous reduction of the amount of precursor being used. These experiments demonstrated that until down to 0.05-0.10 mg (0.15-0.30 µmol) of **4** (free base) radiochemical yields could be maintained at 55-65% (Fig. 5).
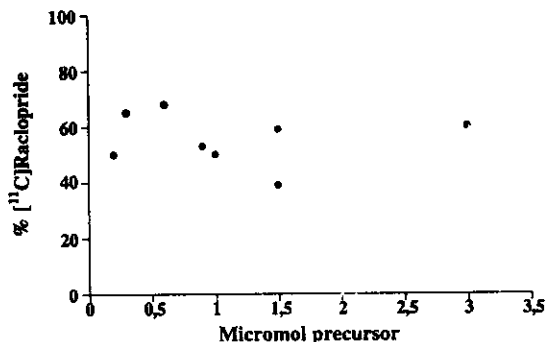

Figure 5 : [^{11}C]Raclopride with reduced precursor amounts.

It is important to avoid contamination of the radiolabelled product with the radiolabelling precursor. By comparison to the conventional [^{11}C]raclopride radiosynthesis with [^{11}C]MI, in our [^{11}C]MT experiments HPLC separations of unreacted desmethyl-precursor **4** and [^{11}C]raclopride ([^{11}C]**1**) have been improved due to a significant reduction of precursor amounts (30-60 times less than in the [^{11}C]MI experiments) (Fig's. 6A & 6E). Fig. 6E depicts a typical semi-preparative HPLC chromatogram for the synthesis of [^{11}C]raclopride using [^{11}C]MT and the precursor **4** in the form of its free base.

For radiolabelling of raclopride with [^{11}C]MI DMSO has been employed as a solvent (10, 13). Purification by semipreparative normal phase HPLC, which offers certain advantages in terms of good separation of the desmethyl-precursor from the product and fast removal of HPLC solvent, is difficult to perform as DMSO interferes with the chromatographic separation. Acetone - on the other hand - can be easily removed and offers the advantage that normal phase HPLC purification should thus become feasible.

Our new method to prepare [¹¹C]raclopride by use of desmethyl-raclopride (free base) and [¹¹C]MT in acetone/aqueous NaOH was entirely automated in a recently developed system containing a GEMS MeI MicroLab (14). The total synthesis time including HPLC purification and formulation of product was about 35 min and the specific radioactivity of [¹¹C]raclopride at the end of synthesis (EOS) was in the range of 1500-2000 Ci/mmol (56-74 GBq/µmol). We have performed this radiosynthesis for several months now in our laboratory on a routine scale and have so far observed good reproducibility of radiochemical yields.
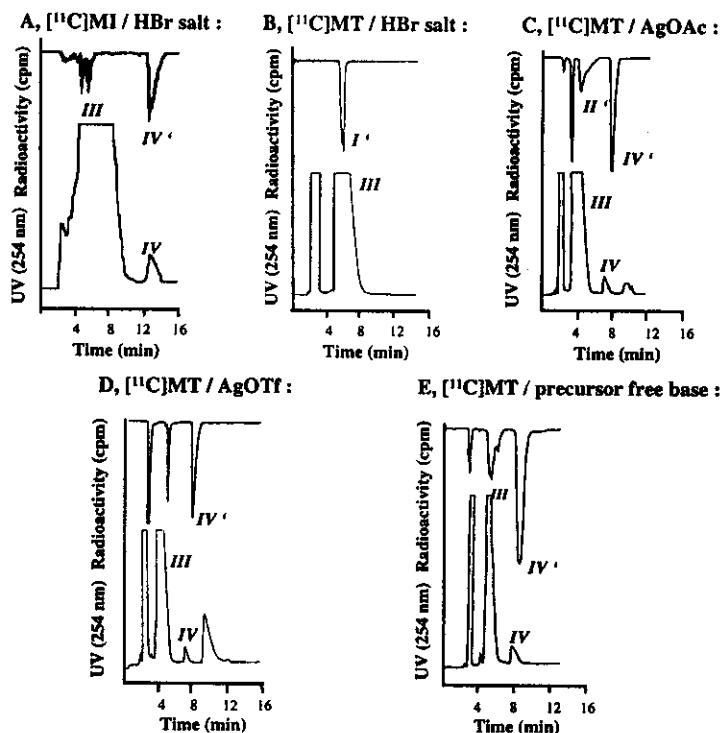

Figure 6 : HPLC chromatograms from the purification of [¹¹C]raclopride. *I ʻ* : [¹¹C]methyl bromide; *II ʻ* : [¹¹C]methyl acetate; *III* : desmethyl-raclopride; *IV* : raclopride; *IV ʻ* : [¹¹C]raclopride.

Experimental

General

Chemicals :   3,5-Dichloro-2,6-dimethoxybenzoic acid was purchased from Maybridge, UK and (S)-(-)-2-aminomethyl-1-ethylpyrrolidine from TCI, Japan. Other chemicals were purchased from Aldrich, Fluka or Sigma France and were used without further purification, unless otherwise stated.

Production of [¹¹C]MT :   The [¹¹C]carbon dioxide was produced at the Accelerator Laboratory of Åbo Akademi with a 103 cm isochronous Efremov cyclotron and at the Karolinska Hospital with a

Scanditronix RNP 16 cyclotron using the ^{14}N(p,α)^{11}C reaction. [^{11}C]MI was prepared from [^{11}C]carbon dioxide either (A) via [^{11}C]CH$_3$OH in a one-pot reaction set up as described elsewhere (15) (Turku) or (B) via [^{11}C]CH$_4$ by catalytic gas-phase iodination in a GEMS MeI MicroLab (14) (Karolinska). [^{11}C]MT was prepared by sweeping the [^{11}C]MI vapour through a glass column, heated at 150-200°C, containing silver triflate impregnated graphitized carbon (1) (Fig. 7).

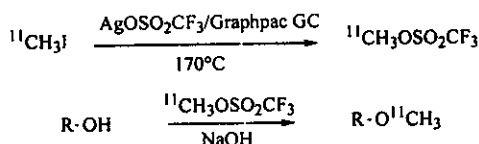$$^{11}CH_3I \xrightarrow[170°C]{AgOSO_2CF_3/Graphpac\ GC} {}^{11}CH_3OSO_2CF_3$$

$$R\text{-}OH \xrightarrow[NaOH]{^{11}CH_3OSO_2CF_3} R\text{-}O^{11}CH_3$$

Figure 7 : Preparation of [^{11}C]methyl triflate from [^{11}C]methyl iodide and use in the O-[^{11}C]methylation of desmethyl-raclopride.

Analysis and purification :   Analytical TLC was run on pre-coated plates of silica gel 60 F_{254} (Merck). The compounds were localized using a UV-lamp at 254 nm and/or by dipping the TLC-plates into an aqueous KMnO$_4$ solution (1%) and heating on a hot plate. Flash chromatography was conducted on silicagel 63-200 μm (Merck) at 0.3 bars. Semipreparative reversed phase HPLC was performed using a Kontron 420 pump, an automatic sample injector (type VICI with a 1 mL loop), a Waters μ-Bondapak column (300 x 7.8 mm, 10 μm), and a Kontron 432 UV-detector (254 nm) in series with a GM-tube for detection of radioactivity. A mixture of 0.01M aqueous H$_3$PO$_4$/CH$_3$CN (70/30) was used as mobile phase with a flow rate of 6 mL/min. The radiochemical yields were calculated by expressing the amount of product collected after semipreparative HPLC (decay-corrected from initial [^{11}C]MT) over total trapped [^{11}C]MT in percent. The radiochemical purity was analyzed by reversed-phase HPLC using a Waters μ-Bondapak-C18 column (300 x 3.9 mm, 10 μm) and the same mobile phase as above with a flow rate of 2 mL/min. NMR spectra were recorded on a Bruker AMX 300 MHz apparatus using the hydrogenated residue of the deuteriated solvents (CD$_2$Cl$_2$, δ = 5.32 ppm ; DMSO-d$_6$, δ = 2.51 ppm) and/or TMS as internal standards for ^{1}H NMR as well as the deuteriated solvents (CD$_2$Cl$_2$, δ = 53.8 ppm ; DMSO-d$_6$, δ = 39.7 ppm) and/or TMS as internal standards for ^{13}C NMR. The chemical shifts (δ) are reported in ppm, downfield from TMS (s, t, bs, bt for singlet, triplet, broad singlet and broad triplet respectively). The mass spectra (MS), DCI/NH$_4^+$, were measured on a Nermag R10-10 apparatus.

Chemistry

Precursor synthesis

(S)-(-)-3,5-Dichloro-N-((1-ethyl-2-pyrrolidinyl)methyl)-2,6-dimethoxybenzamide (3)

To a solution of 3,5-dichloro-2,6-dimethoxybenzoic acid (2) (3.0 g, 11.9 mmol) in toluene (20 mL) SOCl$_2$ (2.6 mL, 36 mmol) and three drops of freshly distilled DMF were added. The reaction solution was stirred under nitrogen at 60°C for two hours. Concentration to dryness under repeated

addition of CH_2Cl_2 yielded the acid chloride as an orange solid that was not purified any further. The residue was dissolved in CH_2Cl_2 (10 mL) and (S)-(-)-2-aminomethyl-1-ethylpyrrolidine (1.5 g, 11.9 mmol, 1 eq) dissolved in CH_2Cl_2 (5 mL) was added dropwise. The reaction mixture was stirred overnight at ambient temperature. The solvent was removed, the residue redissolved in CH_2Cl_2 and washed with aqueous NaOH (1M). The aqueous phase was re-extracted with CH_2Cl_2 and the combined organic phases were dried (Na_2SO_4) and concentrated. The residue was purified by flash chromatography on silica gel (CH_2Cl_2/CH_3OH 84/16) to afford the title compound **3** in the form of white crystals (4.1 g, 95% yield).

Rf (CH_2Cl_2/CH_3OH 7/3) : 0.55-0.60. ^{1}H NMR (CD_2Cl_2, 298.0 K) : δ : 1.10 (t, J = 6.0 Hz) ; 1.60-3.80 (11H) ; 3.90 (s, 6H) ; 7.15 (bs, $w_{1/2}$ = 22 Hz) ; 7.48 (s, 1H). ^{1}H NMR (DMSO-d$_6$, 298.0 K) : δ : 1.10 (t, J = 6.0 Hz) ; 1.50-3.70 (11H) ; 3.80 (s, 6H) ; 7.72 (s, 1H) ; 8.49 (bt, $w_{1/2}$ = 14 Hz). ^{13}C NMR (CD_2Cl_2, 298.0 K) : δ : 13.9 (CH_3) ; 23.0 (CH_2) ; 28.6 (CH_2) ; 41.7 (CH_2) ; 48.5 (CH_2) ; 53.8 (CH_2) ; 62.5 (CH_3) ; 63.0 (CH) ; 123.8 (C) ; 130.3 (C) ; 131.1 (CH) ; 152.6 (C) ; 163.8 (C). ^{13}C NMR (DMSO-d$_6$, 298.0 K) : δ : 13.4 (CH_3) ; 22.1 (CH_2) ; 28.3 (CH_2) ; 42.1 (CH_2) ; 48.0 (CH_2) ; 53.0 (CH_2) ; 61.8 (CH_2) ; 63.0 (CH) ; 122.6 (C) ; 130.1 (CH) ; 130.2 (C) ; 151.7 (C) ; 162.7 (C). MS (DCl/NH$_4$$^+$) : $C_{16}H_{22}Cl_2N_2O_3$: 365, 363, 361 (M + H$^+$).

3, Hydrochloride salt

Compound **3** (3.5 g) was dissolved in CH_2Cl_2 and 3N HCl in Et_2O (15 mL) was added. Concentration to dryness yielded the hydrochloride of **3** as a fluffy white solid (3.8 g, 81% yield).

^{1}H NMR (CD_2Cl_2, 298.0 K) : δ : 1.40 (t, J = 6.0 Hz) ; 1.90-4.20 (11H) ; 3.86 (s, 6H) ; 7.45 (s, 1H) ; 8.66 (bs, $w_{1/2}$ = 19 Hz) ; 11.44 (bs, $w_{1/2}$ = 28 Hz). ^{1}H NMR (DMSO-d$_6$, 298.0 K) : δ : 1.30 (t, J = 6.0 Hz) ; 1.70-4.00 (11H) ; 3.80 (s, 6H) ; 7.78 (s, 1H) ; 9.06 (bt, $w_{1/2}$ = 18 Hz) ; 10.92 (bs, $w_{1/2}$ = 27 Hz). ^{13}C NMR (CD_2Cl_2, 298.0 K) : δ : 10.7 (CH_3) ; 23.5 (CH_2) ; 27.8 (CH_2) ; 39.8 (CH_2) ; 50.9 (CH_2) ; 54.0 (CH_2) ; 62.7 (CH_3) ; 67.8 (CH) ; 123.9 (C) ; 129.8 (C) ; 131.7 (CH) ; 152.5 (C) ; 164.6 (C). ^{13}C NMR (DMSO-d$_6$, 298.0 K) : δ : 10.3 (CH_3) ; 21.6 (CH_2) ; 27.5 (CH_2) ; 39.8 (CH_2) ; 48.5 (CH_2) ; 52.6 (CH_2) ; 62.0 (CH_2) ; 65.5 (CH) ; 122.7 (C) ; 129.5 (C) ; 130.7 (CH) ; 151.6 (C) ; 163.2 (C).

(S)-(-)-3,5-Dichloro-N-((1-ethyl-2-pyrrolidinyl)methyl)-2-hydroxy-6-methoxybenzamide (raclopride, 1)

To a solution of (S)-(-)-3,5-dichloro-N-((1-ethyl-2-pyrrolidinyl)methyl)-2,6-dimethoxy-benzamide (**3**) hydrochloride (3.7 g, 9.3 mmol) in CH_2Cl_2 (20 mL), cooled to -20°C, BBr_3 in CH_2Cl_2 (1.0M, 14 mL, 14 mmol) was added and the solution was stirred for two hours at room temperature. The reaction mixture was concentrated to dryness and the residue was treated with 2M aqueous NaOH. The solution was stirred at room temperature for 30 min and subsequently acidified with concentrated aqueous HCl. Repeated extraction with EtOAc, drying of the combined organic phases with Na_2SO_4 and concentration to dryness yielded the crude product in the form of yellow crystals. The crude product was redissolved in a small quantity of water, made alkaline with concentrated aqueous NH_4OH and repeatedly extracted with EtOAc. The combined organic phases were dried

(Na$_2$SO$_4$) and the residue obtained after concentration was purified by silica gel flash chromatography (CH$_2$Cl$_2$/CH$_3$OH 95/5) to afford the pure title compound **1** in the form of a yellowish oil (2.9 g, 91% yield).

Rf (CH$_2$Cl$_2$/CH$_3$OH 7/3) : 0.50-0.55. ^{1}H NMR (CD$_2$Cl$_2$, 298.0 K) : δ : 1.30 (t, J = 6.0 Hz) ; 1.60-3.90 (11H) ; 4.00 (s, 3H) ; 7.49 (s, 1H) ; 9.24 (bs, w$_{1/2}$ = 19 Hz) ; 12.92 (bs, w$_{1/2}$ = 39 Hz). ^{13}C NMR (CD$_2$Cl$_2$, 298.0 K) : δ : 12.1 (CH$_3$) ; 23.3 (CH$_2$) ; 28.9 (CH$_2$) ; 41.1 (CH$_2$) ; 50.1 (CH$_2$) ; 53.9 (CH$_2$) ; 62.4 (CH$_3$) ; 64.8 (CH) ; 109.9 (C) ; 117.1 (C) ; 118.9 (C) ; 134.2 (CH) ; 154.5 (C) ; 158.5 (C) ; 169.5 (C). MS (DCI/NH$_4^+$) : C$_{15}$H$_{20}$Cl$_2$N$_2$O$_3$: 351, 349, 347 (M + H$^+$).

Note : ^{1}H NMR as well as ^{13}C NMR were fully superimposable with those obtained from authentic raclopride furnished as a gift by ASTRA (Södertälje, Sweden).

(S)-(-)-3,5-Dichloro-N-((1-ethyl-2-pyrrolidinyl)methyl)-2,6-dihydroxybenzamide (4)

To a solution of (S)-(-)-3,5-dichloro-N-((1-ethyl-2-pyrrolidinyl)methyl)-2-hydroxy-6-methoxybenzamide (raclopride, **1**) (1.1 g, 3.11 mmol) in 100% CH$_3$COOH (3 mL) HBr in CH$_3$COOH (5.0M, 2 mL, 10 mmol) was added and the reaction solution was stirred under nitrogen at 60°C for two hours. The solution was concentrated to dryness under repeated addition of toluene to obtain the crude product in the form of brownish crystals. Recrystallization from EtOH/heptane afforded the pure hydrobromide of **4** as white crystals (700 mg, 54% yield).

^{1}H NMR (DMSO-d$_6$, 298.0 K) : δ : 1.30 (t, J = 6.0 Hz) ; 1.70-4.10 (11H) ; 7.67 (s, 1H) ; 9.43 (bs, w$_{1/2}$ = 27 Hz) ; 9.74 (bs, w$_{1/2}$ = 44 Hz). ^{13}C NMR (DMSO-d$_6$, 298.0 K) : δ : 10.4 (CH$_3$) ; 21.7 (CH$_2$) ; 27.6 (CH$_2$) ; 39.2 (CH$_2$) ; 49.1 (CH$_2$) ; 53.2 (CH$_2$) ; 65.6 (CH) ; 106.8 (C) ; 111.7 (C) ; 132.5 (CH) ; 154.0 (C) ; 169.3 (C).

Note : ^{1}H NMR as well as ^{13}C NMR were fully superimposable with those obtained from authentic nor-raclopride furnished as a gift by ASTRA (Södertälje, Sweden).

4, Free amine

An aliquot of the hydrobromide salt of **4** (100 mg) was dissolved in 15 mL H$_2$O. The solution was made alkaline to pH 8-9 with aqueous NH$_4$OH (2M) and repeatedly extracted with EtOAc. Drying (MgSO$_4$) of the combined organic phases and concentration to dryness yielded 70 mg (87%) of the amine **4** in the form of a fine, yellowish powder that could be stored at −20°C without any degradation. Alternatively, to aliquots of the hydrobromide of **4** (1 mg, 2.4 μmol) dissolved in acetone (0.1 mL) AgOAc (10.8 μmol, 1.8 mg, 4.5 eq) suspended in acetone (0.2 mL) was added. The mixture was put into the freezer (-20°C) for two hours and then filtered directly into a reaction vessel (1.0 mL mini-vial, Alltech) and used immediately afterwards for [^{11}C]MT radiolabelling.

Rf (CH$_2$Cl$_2$/CH$_3$OH 7/3) : 0.45-0.50. ^{1}H NMR (DMSO-d$_6$, 298.0 K) : δ : 1.22 (t, J = 9.0 Hz) ; 1.50-4.00 (11H) ; 7.08 (s, 1H) ; 13.31 (bs, w$_{1/2}$ = 20 Hz). ^{13}C NMR (DMSO-d$_6$, 380.0 K) : δ : 11.2 (CH$_3$) ; 22.0 (CH$_2$) ; 28.0 (CH$_2$) ; 39.2 (CH$_2$) ; 48.7 (CH$_2$) ; 53.3 (CH$_2$) ; 65.6 (CH) ; 103.8 (C) ; 106.0 (C) ; 130.4 (CH) ; 161.8 (C) ; 173.5 (C). MS (DCI/NH$_4^+$) : C$_{14}$H$_{18}$Cl$_2$N$_2$O$_3$: 337, 335, 333 (M + H$^+$).

Radiochemistry

[¹¹C]Raclopride ([¹¹C]1)

a) Using the precursor in the form of the free amine:

The [¹¹C]MI vapour was swept through a glass column (diameter 3 mm, length 4 cm, heated at 150-200°C) containing graphitized carbon impregnated with silver triflate. The [¹¹C]MT was trapped at room temperature into a reaction vessel (1.0 mL mini-vial, Alltech) containing 0.05-0.10 mg (0.15-0.30 μmol) of the desmethyl-precursor **4** (free amine) dissolved in acetone (200 μL) and aqueous NaOH (0.1M, 1.50-3.00 μL, 0.15-0.30 μmol, 1.0 eq). The amount of trapped radioactivity was measured in a well counter. Mobile phase (0.6 mL) was added before injection onto the semipreparative HPLC column. [¹¹C]Raclopride was collected at 8-10 min. The fraction containing the pure product was concentrated and redissolved in sterile phosphate buffered saline (pH 7.4) and sucked into an empty vial. The radioactivity in this vial was measured and the yield calculated (decay corrected) on the amount initially trapped in the reaction solution.

b) Using silver acetate to precipitate the bromide ion:

[¹¹C]MT, prepared as described before, was trapped at room temperature into a reaction vessel (1.0 mL mini-vial, Alltech) containing the filtrate obtained after precipitation of the bromide ion with AgOAc (compare before) and aqueous NaOH (1.0M, 8 μL, 8 μmol, 3.3 eq). The vessel was sealed and heated at 60°C for one minute and the radioactivity in the vial measured in a well counter after cooling. Semipreparative reversed phase HPLC purification and calculation of radiochemical yields were performed as described before.

Conclusion

We prepared desmethyl-raclopride in a straightforward synthetic approach. [¹¹C]Raclopride was synthesized in 55-65% radiochemical yield from [¹¹C]MT by use of desmethyl-raclopride free base (0.05-0.10 mg) dissolved in acetone and aqueous NaOH (1 eq). Compared to the standard radiosynthesis of [¹¹C]raclopride with [¹¹C]MI, this procedure meant an improvement in terms of better HPLC separations due to the use of less precursor and no DMSO, as well as in terms of good reproducibility of radiochemical yields.

Acknowledgements

References

1. Jewett D. M. - Appl. Radiat. Isot. 43: 1383 (1992)
2. Långström B. and Lundqvist H. - Int. J. Appl. Radiat. Isot. 27: 357 (1976)
3. Marazano C., Maziere M., Berger G. and Comar D. - Int. J. Appl. Radiat. Isot. 28: 49 (1977)
4. Någren K., Müller L., Halldin C., Swahn C.-G. and Lehikonen P. - Nucl. Med. Biol. 22: 235 (1995)
5. Någren K., Halldin C., Müller L., Swahn C.-G. and Lehikonen P. - Nucl. Med. Biol. 22: 965 (1995)
6. Lundkvist C., Sandell J., Någren K., Pike V.W. and Halldin C. - J. Label. Compds. Radiopharm. 41: 545 (1998)
7. Någren K. and Halldin C. - J. Label. Compds. Radiopharm. 41: 831 (1998)
8. Köhler C., Hall H., Ögren S-O. and Gawell L. - Biochemical Pharmacology 34: 2251 (1985)
9. Farde L. - TINS 19: 211 (1996)
10. Halldin C., Farde L., Högberg T., Hall H., Ström P., Ohlberger A. and Solin O. - Nucl. Med. Biol. 18: 871 (1995)
11. Langer O., Halldin C., Dollé F., Swahn C.-G., Olsson H., Karlsson P., Hall H., Sandell J., Lundkvist C., Vaufrey F., Loc'h C., Crouzel C., Mazière B. and Farde L. - Nucl. Med. Biol. (in press, 1999)
12. de Paulis T., Kumar Y., Johansson L., Rämsby S., Hall H., Sällemark M., Ängeby-Möller K. and Ögren S-O. - J. Med. Chem. 29: 61 (1986)
13. Ehrin E., Gawell L., Högberg T., de Paulis T. and Ström P. - J. Label. Compds. Radiopharm. 24: 931 (1987)
14. Larsen P., Sandell J., Printz G., Söderholm P. and Halldin C. - J. Label. Compds. Radiopharm. 40: 320 (1997)
15. Halldin C., Farde L., Högberg T., Hall H. and Sedvall G. - Appl. Radiat. Isot. 41: 669 (1990)