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Short Communication

Synthesis of a carbon-14 analogue of N-(aryl-methyl)-3-phenyl-acryl amidine-[carboxy-^{14}C] and its derivatives as NR2B-selective NMDA receptor antagonist

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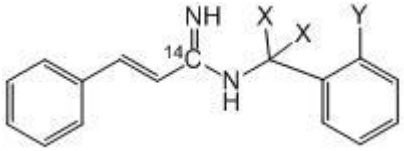
Abstract

Four amidine NR2B-selective NMDA receptor antagonists, N-(2-methoxy benzyl) -3-phenyl-acrylamidine, N-[diduterio(2-methoxyphenyl) methyl]-3-phenyl-acrylamidine, N-benzyl-3-phenyl-acryl amidine and N-[diduterio(phenyl)methyl]-3-phenyl-acrylamidine, all four labeled with carbon-14 in the 1-position, have been synthesized as part of 5-step sequence from $Ba^{14}CO_3$.

Keywords: NMDA-receptor; Carbon-14; Labeling; Amidines.

1. Introduction

The N-methyl-D-aspartate (NMDA) receptor is highly expressed in the central nervous system (CNS) and is comprised of a minimum of two different subunits, NR1 and NR2. The NR1 subunit has at least eight isoforms (NR1a-h) and the NR2 subunit has four distinct subtypes (NR2A-D) [1-5]. It has been suggested that NR2B selective compounds may have a reduced side-effect profile when compared to NMDA receptor antagonists, and have been shown to be efficacious in preclinical pain models [6, 7].



	X	Y
1a	H	CH_3O
b	H	H
c	D	CH_3O
d	D	H

Fig. 1. Chemical structure the synthesis of four N-(aryl-methyl)-3-phenyl-acrylamidine -[1-^{14}C].

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Among the known NR2B-selective compounds are ifenprodil, CP-101,606, Ro-25-6981 and a novel series of amidines [8-9]. Based on the amidine class of compounds, recently, a novel series of amidines has been described and shown to exhibit high NR2B-subtype selective NMDA antagonistic activity [10]. Within the subclass of benzamidines, N-(2-methoxybenzyl)-3-phenyl-acrylamidine,has already been labelled with carbon-11 and is currently under investigation [11]. For studies of pharmacokinetics and metabolism of this compound, versions with a metabolically suitable carbon-14 label was required [12-13]. This paper reports the synthesis of four N-(aryl-methyl)-3-phenyl-acrylamidine -[1-^{14}C] **1a-d** (Fig. 1).

2. Experimental

IR spectra were recorded on a Bruker FT-IR, Vector 22 instrument and the ^{1}H- NMR spectra were recorded on a Varian Unity plus 400 spectrometer (400 MHz). Radioactivity was determined using a Beckman LS6500 liquid scintillation spectrometer. Mass spectra were obtained on a Finnigan TSQ-70 instrument.

2.1. Potassium [^{14}C] –cyanide 3

A mixture of barium [^{14}C] carbonate **2** (46MBq) and ammonium chloride, under nitrogen was added potassium, which was placed in a furnace and heated for 60min. at 100 °C. The contents were cooled and treated with water to destroy the excess potassium. The mixtuer was transferred into a distilling flask and acidified with sulfuric acid, then the product was distilled into a potassium hydroxide solution. The solution was concentrated under reduced pressure and gave (37 MBq) of the title compound **3**.

2.2. 2-Cyanoacetic acid-[cyano-^{14}C] 5

In a 25 ml round-bottomed flask, 500 mg (5.3 mmol) of 2-chloroacetic acid **4** was dissolved in 1 ml of water. The solution was warmed to 50 °C and neutralized with anhydrous sodium carbonate of which about 290 mg (2.7 mmol) was required. Meanwhile (390 mg, 37MBq, 6mmol) of potassium [^{14} C] –cyanide **3** was dissolved in 1ml of water warmed to 55 °C. The potassium [^{14} C] –cyanide **3** solution was then added to the sodium 2-chloroacetic acid solution, which had been cooled to room temperature, with rapid mixing of two solutions and cooling under water bath. The temperature rapidly rises; when it reached 95 °C the solution should be cooled by adding 0.2 mL of cold water and this repeated, if necessary, until the temperature no longer rises. The solution was heated to the boiling point and boilded for 5 min, and finally cooled to room temperature. The solution was filtered and the 2-cyanoacetic acid-[cyano-^{14}C] **5** was set free by adding with thorough stirring 0.6 (5.8 mmol) of hydrochloric acid. The solution was evaporated on a water bath at 60-70 °C under a pressure of 20-30 mm. and the evaporation continued until no more distillate comes over. To the residue was added 1ml of 95 % alcohol. The solution was filtered from the sodium chloride, and the residue was washed with another 1ml portion of alcohol. On evaporating the alcoholic solution under reduced pressure from a water bath held at 50-60 °C to give the title compound **5** (408 mg, 29.6 MBq, 80 %). IR(KBr): 3522, 22929, 2266, 1734,11395, 1191, 934 Cm^{-1}

2.3. Cinnamonitrile-[3-^{14}C] 7

A mixture of (550 mg, 5.18 mmol) of freshly distilled benzaldehyde **6**, (400 mg, 29 MBq, 4.7 mmol) of 2-cyanoacetic acid-[cyano-^{14}C] **5**, 14 mg of ammonium acetate, 5 mL of toluene, 2.5 mL of pyridine was placed in a 25 mL round-bottomed flask equipped with a Stark and Dean water trap and reflux condenser. The mixture was boiled under reflux for 2 days. The theoretical quantity of water was collected in trap within 1 hour. Upon completion of the reflux period, the

solvent was removed under reduced pressure by heating on water bath. The crude product was purified by silica gel chromatography using ethyl acetate: hexane (1: 9) as eluant to give the title compound **7** (461mg, 22MBq, 76%). ^{1}H-NMR (CCl_4, TMS) : δ 7.44(d, 1H, J=17Hz), δ 7.31 (pseudo-s , 5H aromatic protons), δ 5.71 (d, 1H, J=17Hz) (Signals for E-Isomer), & δ 7.31 (pseudo-s , 5H aromatic protons), δ 6.98(d, 1H, J=12Hz), δ 5.31 (d, 1H, J=17Hz) (Signals for Z-Isomer), (E/Z ratio:2.68).

2.4. (E)-3-phenyl-acrylimidic acid ethyl ester, hydrochloride [1-^{14}C] 8

A solution of cinnamonitrile-[3-^{14}C] **7** (461 mg, 22 MBq) in anhydrous EtOH (6.5 mL) under argon was cooled to 0 °C. Gaseous hydrogen chloride was bubbled through the solution for 3 h and the solution was warmed to room temperature and stirred for an additional 15 h. The reaction mixture was evaporated to dryness, taken up in Et_2O, and filtered to give the corresponding imidate salt **8** (439mg, 12.78MBq) as a white solid. ^{1}H-NMR (CD_2Cl_2, TMS) : δ 12.35(bs, 1H), δ 11.53 (bs , 1H), δ 7.95 (d, 1H, J=16Hz) , δ 7.61-7.68(m, 2H), δ 7.41-7.49(m, 3H), δ 7.14 (d, 1H, J=16Hz) , δ 4.37 (q, 2H, J=6.5Hz), δ 1.56 (t,3H, J=6.5Hz) , MS (70eV): m/z = 178$[M+H]^+$.

2.5. (E)-N-(2-methoxybenzyl)-3-phenyl-acrylamidine- [1-^{14}C] 1a

To a solution of (E)-3-phenyl-acrylimidic acid ethyl ester, hydrochloride [3-^{14}C] **8** (439mg, 12.78MBq) in DMF (4.5 mL) was added triethylamine (300 µL,). The suspension was filtered off and rinsed twice with DMF (1.3 mL). To the combined filtrates containing the free base was added triethylamine (300 µL) and 2-methoxybenzyl amine **9a** (228 µL). The reaction mixture was stirred for 3h at room temperature under argon and concentrated under reduced pressure. The residue was purified by column chromatography (eluent: CH_2Cl_2/MeOH: 9/1) to yield the title compound **1a** as a white solid(227mg, 5.24MBq, 41%).^{1}H-NMR (MeOH-d_4): δ 7.62 (d, 1H, J=16Hz) , δ 7.60(m, 2H), δ 7.41-7.22(m, 5H), δ 6.88-7.02 (m, 2H) , δ 6.63 (d, 1H, J=16Hz), δ 4.42 (s,2H), δ 3.75(s, 3H). MS (70eV): m/z = 269$[M+H]^+$.

2.6. Benzamidine, N-(benzyl)cinnamimidamide-[1-^{14}C] 1b

1b was prepared according to the above described procedure for **1a** by using benzyl amine **9b** instead of 2-methoxybenzyl amine **9a** (yield :46%). ^{1}H-NMR (MeOH-d_4): δ 7.62 (d, 1H, J=16Hz) , δ 7.60(m, 2H), δ 7.23-7.42(m, 8H), δ 6.63 (d, 1H, J=16Hz), δ 4.42 (s,2H), MS (70eV): m/z = 239$[M+H]^+$.

2.7. (E)-N-[diduterio(2-methoxyphenyl)methyl]-3-phenyl-acrylamidine- [1-^{14}C] 1c

1c was prepared according to the above described procedure for **1a** by using (2-methoxyphenyl)methanamine-[methan-d_2] **9c** instead of 2-methoxybenzyl amine **9a** (yield :41%). ^{1}H-NMR (MeOH-d_4): δ 7.62 (d, 1H, J=16Hz) , δ 7.60(m, 2H), δ 7.41-7.22(m, 5H), δ 6.88-7.02 (m, 2H) , δ 6.63 (d, 1H, J=16Hz), δ 3.75(s, 3H). MS (70eV): m/z = 271$[M+H]^+$.

2.8. (E)-N-[diduterio(phenyl)methyl]-3-phenyl-acrylamidine- [1-^{14}C] 1d

1d was prepared according to the above described procedure for **1a** by using phenylmethanamine-[methan-d_2] **9d** instead of 2-methoxybenzyl amine **9a** (yield :46%). ^{1}H-NMR (MeOH-d_4): δ 7.62 (d, 1H, J=16Hz) , δ 7.60(m, 2H), δ 7.23-7.42(m, 8H), δ 6.63 (d, 1H, J=16Hz). MS (70eV): m/z = 241$[M+H]^+$.

2.9. Phenylmethanamine-[methan-d_2] 9d

A solution of (2 mmol) of LAD in 3 mL of ether was placed in a 20 mL, three-necked flask equipped with a reflux condenser, dropping funnel and mechanical stirrer. Through the dropping funnel, a solution of (206 mg, 2 mmole) of benzonitrile in 2 mL of ether was introduced at a rate such as to produce gentle reflux. Shortly after completing the addition, sufficient water was added dropwise, and with cooling of the flask in ice-water, to decompose the excess hydride. There was then added 5 mL of a 20 % solution of sodium potasssium tatrate. The clear mixture was trasferred to a separatory funnel and after separating the ether layer, the aqueous layer was extracted with two 5 mL portions of ether. From the combined ether extracts there was obtained, after drying over calcium sulfate, a 71.5 % of the title compound **9d** (156 mg).

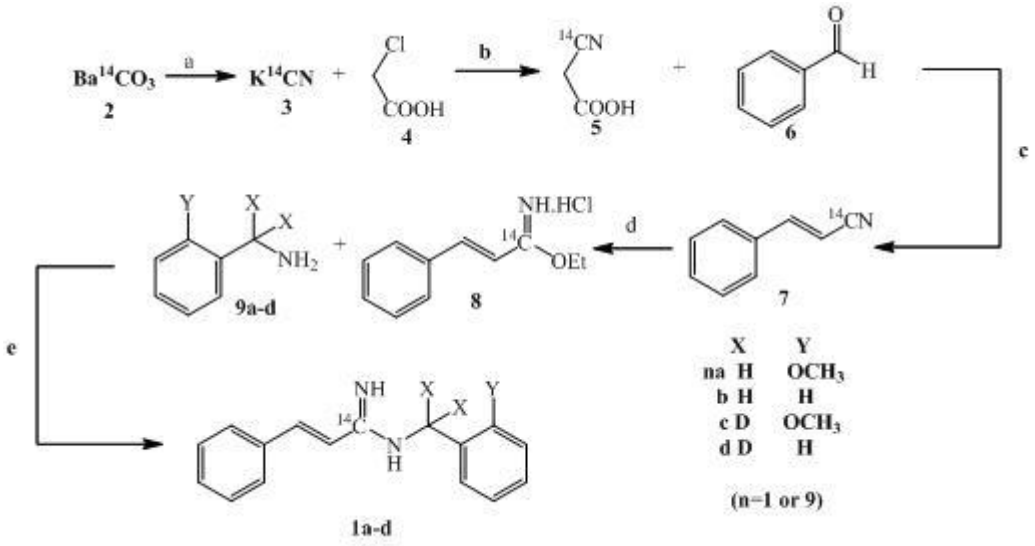
2.10. (2-methoxyphenyl)methanamine-[methan-d_2] 9c

1b was prepared according to the above described procedure by reaction 2-methoxy-benzonitrile (266 mg, 2 mmol) with (2 mmol) of LAD in dry ether to give the title compound (222 mg, 1.6 mmol, 80 %).

3. Results and discussion

Our approach to the synthesis of N-(aryl-methyl)-3-phenyl-acryl amidine-[1-^{14}C] **1a-d** is shown in scheme 1. After conversion of barium [^{14}C] carbonate **2** to potassium [^{14}C] cyanide **3** in the presence of ammonium chloride and potassium, under furnace condition [14], 2-cyanoacetic acid-[cyano-^{14}C] **5** was derived from the addition of potassium [^{14}C] cyanide **3** to 2-chloroacetic acid **4** with good yield [15-16]. In the next step, the latter product was coupled with benzaldehyde **6** by using ammonium acetate in the mixture of toluene and pyridine, and cinnamonitrile-[cyano-^{14}C] **7** was achieved in good yield [17-19].

Then cinnamonitrile-[cyano-^{14}C] **7** could then be converted to 3-phenyl-acrylimidic ethyl ester-[carboxy-^{14}C], hydrochloride **8** via the Pinner synthesis by treatment with gaseous hydrogen chloride in anhydrous ethanol [20-21]. In the final step, coupling of the imidate salt 3-phenyl-acrylimidic ethyl ester-[carboxy-^{14}C], hydrochloride **8** with arylmethan-amine **9a-d** in a mixture of DMF and triethylamine at room temperature gave N-(aryl-methyl)-3-phenyl-acrylamidine-[1-^{14}C], **1a-d**[22-24].



Scheme 1 a) NH_4Cl, K b) Na_2CO_3, water c) NH_4OAc, Toluene, Pyridine d) EtOH (dry), HCl (g) 0 ^oC-RT, e) NEt_3, DMF, RT.

4. Conclusion

In this paper, a convenient synthetic pathway for labeling of a series of N-(aryl-methyl)-3-phenyl-acrylamidine with carbon-14 in the 1-position, has been presented as part of q5-step sequence from $Ba^{14}CO_3$.

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References

[1] H. Mori, M. Mishina, Neuropharm 34 (1995) 1219-1237.
[2] R. Betarbet, R. Porter, J.T. Greenamyre, J. Neurochem, 74 (2000) 1166-1174.
[3] A. Buller, H. Larson, B. Schneider, J. Beaton, R. Morrisett, D. Monaghan, J. Neurosci. 14 (1994) 5471-5484.
[4] R. Dingledine, K. Borges, D. Bowie, S. Traynelis, Pharmacol. Rev. 51 (1999) 7-61.
[5] H. Monyer, N. Burnashev, D.J. Laurie, B. Sakmann, P.H. Seeburg, Neuron 12 (1994) 529-540.
[6] B.A. Chizh, P.M. Headley, T.M. Tzchentke, Trends Pharmacol Sci. 22 (2001) 636-642.
[7] S. Boyce, A. Wyatt, J.K. Webb, R.O. Donnell, G. Mason, M. Rigby, D. Sirinathsinghji, R.G. Hill, N.M. Rupniak, Neuropharm 38 (1999) 611-623.
[8] J. Johnson, P. Ascher, Nature 325 (1987) 529-531.
[9] T. Haradahira, J. Maeda, T. Okauchi, M.R. Zhang, J. Hojo, T. Kida, T. Arai, F. Yamamato, S. Sasaki, M. Maeda, K. Suzuki, T. Suhara, Nucl. Med. Biol. 29 (2002) 517-522.
[10] C.F. Clairborne, J.A. McCauley, B.E. Libby, N.R. Curtis, H.J. Diggle, J.J. Kulagowski, S.R. Michelson, K.D. Anderson, D.A. Claremon, R.M. Freidinger, R.A. Bednar, S.D. Mosser, S.L. Gaul, T.M. Connolly, C.L. Condra, B. Bednar, G.L. Stump, J.J. Lynch, A. Macaulay, K.A. Wafford, K.S. Koblan, N.J. Liverton, Bioorg. Med. Chem. Lett. 13 (2003) 697-704.
[11] C. Thominiaux, B. Bruin, Y. Bramoulle, F. Hinnen, S. Demphel, H. Valette, M. Bottlaender, L. Besret, M. Kassiou, F. Dolle, Appl. Radiat. Isot. 64 (2006) 343-351.
[12] N. Saemian, G. Shirvani, H. Matloubi, J. Labelled Cpd. Radiopharm. 49 (2006) 71-76.
[13] D. Seidel, P. Brehmer, Y. Schoof, U. Weinber, U. Niewohner, M. Nowakowski, J. Labelled Cpd. Radiopharm. 46 (2003) 1019-1025.
[14] N. Saemian, G. Shirvani, S.S. Oliyaee, M. Javaheri, J. Iran. Chem. Res. 2 (2009) 63-67.
[15] N. Saemian, G. Shirvani, M. Javaheri, J. Radioanal. Nuc. Chem. 281 (2009) 421-423.
[16] N. Saemian, G. Shirvani, H. Matloubi, J. Radioanal. Nuc. Chem. 268 (2006) 545-548.
[17] J.H. Ha, J.H. Ahn, Bull. Korean Chem. Soc. 27 (2006) 121-127.
[18] J. Sandel, O. Langer, P. Larsen, F. Dolle, F. Vaufrey, S. Demphel, C. Crouzel, C. Hanlldin, J. Labelled Cpd. Radiopharm. 43 (2000) 331-338.
[19] T.G. Hamill, J.A. McCauley, H.D. Burns, J. Labelled Cpd. Radiopharm. 48 (2005) 1-7.
[20] G. Shirvani, N. Saemian, H. Matloubi, J. Labelled Cpd. Radiopharm. 50 (2007) 1234-1235.
[21] N. Saemian, G. Shirvani, H. Matloubi, J. Radioanal. Nuc. Chem. 274 (2007) 631-633.
[22] D. Haddenham, L. Pasumansky, J. Desoto, S. Eagon, B. Singaram, J. Org. Chem. 74 (2009) 1964-1970.
[23] N.R. Curtis, H.J. Diggle, J.J. Kulagowski, C. Londen, S. Grimwood, P.H. Hutson, F. Murray, P. Richards, A. Macaulay, K.A. Wafford, Bioorg. Med. Chem. Lett.13 (2003) 693-698.
[24] N. Saemian, G. Shirvani, H. Matloubi, J. Radioanal. Nuc. Chem. 274 (2007) 643-646.