Journal of Labelled Compounds and Radiopharmaceuticals—Vol. XXXIV, No. 3

SYNTHESIS OF SODIUM 6-ISOPROPYL-3-[4-(P-CHLOROBENZENESULFONYLAMINO)-BUTYL]-[2-^{14}C]AZULENE-1-SULFONATE

Takashi Yanagisawa[a], Masayuki Yokota[a], Tsuyoshi Tomiyama[a] and Shiro Ikegami[b]
[a]Research Laboratories, Kotobuki Pharmaceutical Co., Ltd.
6351 Sakaki-Machi, Nagano 389-06, Japan
[b]Faculty of Pharmaceutical Sciences, Teikyo University
1099-1 Suarashi Sagamiko-Machi, Tsukuigun, Kanagawa 199-01, Japan

SUMMARY

Sodium 6-isopropyl-3-[4-(p-chlorobenzenesulfonylamino)-butyl]azulene-1-sulfonate (KT2-962) 1, which has been found to be a potent and selective thromboxaneA_2 receptor antagonist, was synthesized in ^{14}C-labelled form by using potassium [^{14}C]-cyanide. ^{14}C-labelled 1 with a specific activity of 2.36 GBq/mmol was prepared with no-carrier-added and in nine steps in 64% overall radiochemical yield.

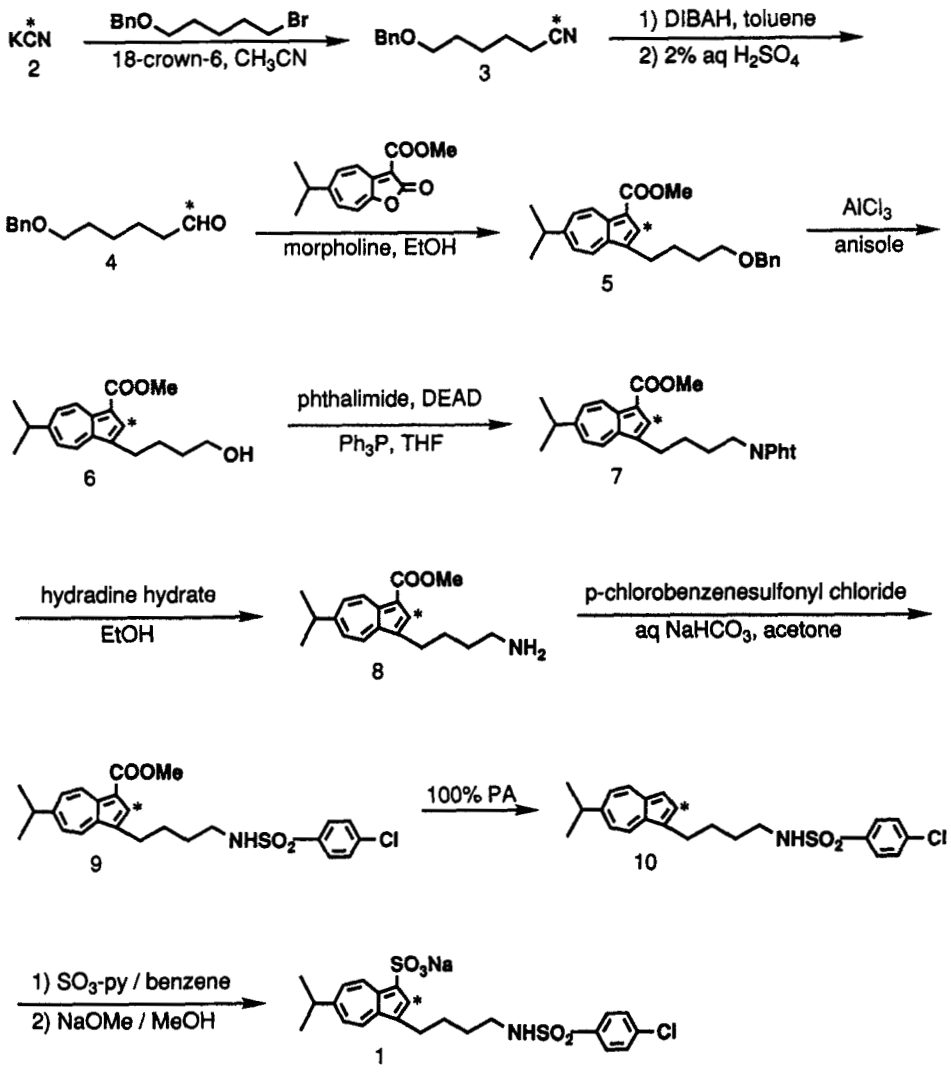
Key Words: thromboxaneA_2, receptor antagonist, sodium 6-isopropyl-3-[4-(p-chlorobenzene-sulfonylamino)-butyl]-[2-^{14}C]azulene-1-sulfonate.

INTRODUCTION

ThromboxaneA_2 (TXA_2), an unstable metabolite of arachidonic acid (AA), is an extremely potent vasoconstricting and platelet-aggregating agent.[1,2] An excess of TXA_2 has been considered to be one of the factors associated with angina pectoris and other cardio- and cerebrovascular diseases. A TXA_2 receptor antagonist is considered to be useful for these diseases. Recently KT2-962 (sodium 6-isopropyl-3-[4-(p-chlorobenzenesulfonylamino)-butyl]azulene-1-sulfonate) 1 has been reported to possess excellent TXA_2 receptor antagonistic activity without any agonistic activity[3,4]. In the course of our metabolism and disposition studies of 1, it was necessary to prepare a ^{14}C-labelled compound. In this paper, we describe the synthetic method for the preparation of 1 (KT2-962) labelled with carbon-14 at the 2-position of the azulene ring.

CCC 0362–4803/94/030205–08

Received 29 June, 1993
Revised 25 October, 1993



Scheme 1

RESULTS AND DISCUSSION

Our synthetic strategy for the azulene ring was based on the reaction of 2H-cyclohepta[b]furan-2-one with enamine generated in situ from aldehyde 4.[5,6] Additionally, our laboratory group has reported the synthesis of sodium 3-ethyl-7-isopropyl-[2-^{14}C]azulene-1-sulfonate.[7] By using this method, we investigated the synthetic route of ^{14}C-KT2-962. Therefore we established the

synthetic route of ^{14}C-KT2-962 1 by using ^{14}C-labelled 4 obtained from potassium [^{14}C]-cyanide as shown in scheme 1. Reaction of potassium [^{14}C]-cyanide 2 with 5-benzyloxy-1-pentylbromide in the presence of 18-crown-6[8] afforded 3 in 98% yield. Reduction of 3 with diisobutylalumium hydride (DIBHA) followed by hydrolysis with 2% aqueous H_2SO_4 afforded the [^{14}C]-hexanal 4 in quantitative yield. The condensation of 4 with methyl 6-isopropyl-2-oxo-2H-cyclohepta[b]furan-3-carboxylate[9] in the presence of morpholine afforded the [2-^{14}C]azulene 5 in 98.6% yield. Deprotection of 5 using aluminum chloride-anisole[10] afforded 6 in 90% yield. Phthalimide 7 was obtained by the Mitsunobu reaction[11] of 6 with phthalimide, diethylazodicarboxylate (DEAD) and triphenylphosphine (Ph_3P) in tetrahydrofuran (THF) in 88% yield. Treatment of 7 with hydrazine hydrate afforded primary amine 8 in quantitative yield. Coupling of the primary amine 8 with p-chlorobenzenesulfonyl chloride in aqueous acetone afforded the [^{14}C]-sulfonamide 9 in 96.2% yield. Decarboxylation of 9 with 100% phosphoric acid afforded 10 in 98.6% yield. Sulfonation with pyridine-sulfur trioxide complex, followed by treatment with sodium methoxide afforded the sodium sulfonate 1 in 85% yield. We could obtain 1 (^{14}C-KT2-962, 1.90 GBq) with no-carrier-added and in 64% overall radio chemical yield from potassium [^{14}C]-cyanide.

EXPERIMENTAL

Potassium [^{14}C]-cyanide (2.96 GBq, 2.13 GBq/mmol) was purchased from Amersham International plc. The reactions in the labelling synthesis were monitored by thin layer chromatography (TLC). Analytical TLC was carried out on a Merck silica gel 60 F_{254} plate (0.25 mm). Fuji Division BM-820MH silica gel was used for column chromatography. Melting points were determined on a micro melting point apparatus. ^{1}H-NMR spectra were measured at 90MHz on a Hitachi R-90H Fourier Transform NMR spectrometer. Chemical shifts are quoted in δ (ppm) downfield from tetramethylsilane (TMS) as an internal standard, and coupling constants (J) are given in Hz. The following abbreviations are used: s=singlet, d=doublet, t=triplet, m=multiplet and brs=broad singlet. Infrared spectra were recorded on a Hitachi 270-

30. These spectral data were obtained in trial experiments using unlabelled material. Radioactivity was determined with a Beckman LS-900 liquid scintillation counter using 2,5-diphenyloxazole in toluene or a mixture of toluene and Triton X-100 as a liquid scintillator. The Radio high performance liquid chromatography (RHPLC) was performed on a 655A-II liquid chromatograph (Hitachi Co., Ltd.,Japan) was equipped with a 655A UV detector (Hitachi Co.) and a RS-8000 Radioanalyzer (Toso). A stainless steel column packed with octadesyl silane (TSK-gel, 80Tm, id, 4.6 x 150 mm) was used for analysis of 1. Operating conditions: mobile phase 20 mM phosphate Buffer (pH 6.0)/CH_3CN=7:3 and 5:5 (v/v); flow rate 1.0 ml/min.; UV 299 nm; retention time for 1 30 min .

5-Benzyloxy-1-[1-^{14}C]hexanonitrile. 3

A suspension of potassium [^{14}C]-cyanide (90.4 mg, 1.38 mmol, 2.98 GBq) and 18-crown-6 (729.5 mg, 2.76 mmol) in CH_3CN (20 ml) was stirred at room temperature for 20 min. To the mixture, 5-benzyloxy-1-pentylbromide (712.1 mg, 2.76 mmol) was added. After stirring at this temperature for 8 hr, the reaction mixture was poured into ice-satd.$NaHCO_3$, then extracted with ethyl acetate. The organic layer was washed with satd. $NaHCO_3$ and satd.brine, dried and evaporated to give a slightly yellow oil. This crude product was purified by column chromatography on silica with ethyl acetate/n-hexane (1:5) as the eluent, to give 275 mg (97.4 %) of 3 (2.88 GBq). IR (neat) cm^{-1}: 2240 (CN). ^{1}H-NMR ($CDCL_3$) δ = 1.40-1.85 (6H, m,-CH_2-), 2.32 (2H, t, -CH_2-), 3.45 (2H, t, -CH_2-), 4.49 (2H, s, -$\underline{CH_2}$-C_6H_5), 7.30 (5H, s, -CH_2-$C_6\underline{H_5}$).

6-Benzyloxy-1-[1-^{14}C]hexanal. 4

A solution of 1.0 M DIBAH in toluene (2.8 ml, 2.8 mmol) was added dropwise to a solution of nitrile 3 (2.88 GBq, 275 mg, 1.35 mmol) in toluene (10 ml) at -78°C under argon atmosphere. After stirring at this temperature for 1.5 hr, 2% sulfuric acid (25 ml) was added at 0°C and the stirring was continued for another 30 min. The mixture was extracted with ethyl acetate. The extract was dried over anhydrous Na_2SO_4, and evaporated to give 4 (2.87 GBq, 278 mg) as a colorless oil. IR (neat) cm^{-1} 1725: (CHO). ^{1}H-NMR ($CDCL_3$) δ = 1.20-1.80 (6H, m, -CH_2-),

2.40 (2H, t, -CH_2-), 3.43 (2H, t, -CH_2-), 4.48 (2H, s, -$C\underline{H}_2$-C_6H_5), 7.30 (5H, s, -CH_2-$C_6\underline{H}_5$), 9.72 (1H, s, -CHO).

Methyl 3-(4-benzyloxybutyl)-6-isopropyl-[2-^{14}C]azulene-1-carboxylate. 5

A mixture of the hexanal 4 (2.87 GBq, 278 mg, 1.35 mmol), morpholine (235 mg, 2.70 mmol) and methyl 6-isopropyl-2-oxo-2H-cyclohepta[b]furan-3-carboxylate (665 mg, 2.70 mmol) in EtOH (15 ml) was stirred under reflux for 8 hr. The mixture was concentrated under reduced pressure, extracted with ethyl acetate, and washed with 2% sulfuric acid, satd. brine. The extract was dried over anhydrous Na_2SO_4, and evaporated to give a violet oil. This crude product was purified by column chromatography on silica with ethyl acetate/n-hexane (1:4) as the eluent, to give 520 mg (98.8%) of 5 (2.84 GBq) as a violet oil. ^{1}H-NMR ($CDCL_3$) δ = 1.35 (6H, d, i-proCH_3), 1.50-1.90 (4H, m, -CH_2-), 2.80-3.20 (3H, t+m, -CH_2-, i-proCH), 3.45 (2H, t, -CH_2-), 3.92 (3H, s, -$COOCH_3$), 4.48 (2H, s, -$C\underline{H}_2$-C_6H_5), 7.30 (5H, s, -CH_2-$C_6\underline{H}_5$), 7.20-7.45 (2H, m, $C_{5,7}$-H), 8.11 (1H, s, C_2-H), 8.26 (1H, d, C_4-H), 9.45 (1H, d, C_8-H).

Methyl 3-hydroxybutyl-6-isopropyl-[2-^{14}C]azulene-1-carboxylate. 6

A solution of 5 (2.84 GBq, 520 mg, 1.33 mmol) in anisole (10 ml) was added to a suspension of $AlCl_3$ (532 mg, 3.99 mmol) in anisole 5 ml at 0°C and the mixture was stirred at room temperature for 30 min. The reaction mixture was poured into ice water (50 ml), and the mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried and evaporated in vacuo. The residue was purified by column chromatography on silica with ethyl acetate/n-hexane (1:4) as the eluent, to give 376.7 mg (94.2%) of 6 (2.67 GBq) as violet oil. ^{1}H-NMR ($CDCL_3$) δ =1.36 (6H, d, i-proCH_3), 1.30-1.80 (4H, m, -CH_2-), 2.80-3.20 (3H, t+m, -CH_2-, i-proCH), 3.65 (2H, t, -CH_2-), 3.92 (3H, s, -$COOCH_3$), 7.20-7.50 (2H, m, $C_{5,7}$-H), 8.11 (1H, s, C_2-H), 8.30 (1H, d, C_4-H),9.45 (1H,d,C_8-H)

Methyl 3-(4-phthalimidebutyl)-6-isopropyl-[2-^{14}C]azulene-1-carboxylate. 7

A mixture of 6 (2.67 GBq, 376.7 mg, 1.3 mmol), phthalimide (386.0 mg, 2.6 mmol),

triphenylphosphine (681.5 mg, 2.6 mmol) and diethylazodicarboxylate (452.8 mg, 2.6 mmol) in dry THF 15 ml was stirred at 0°C for 20 min and room temperature for 10 hr. After the solvent was removed the residue was extracted with ethyl acetate. The ethyl acetate layer was washed with brine, dried and evaporated. The residue was purified by column chromatography on silica with ethyl acetate/n-hexane (1:5) as the eluent, to give 475 mg (88%) of 7 (2.35 GBq) as violet oil. ^{1}H-NMR ($CDCL_3$) δ =1.36 (6H, d, i-proCH_3), 2.50-2.90 (4H, m, -CH_2-), 2.90-3.30 (3H, t+m, -CH_2-, i-proCH), 3.71 (2H, t, -CH_2-), 3.92 (3H, s, -$COOCH_3$), 7.20-7.50 (2H, m, $C_{5,6}$-H), 7.60-7.79 (4H, m , $NC_2O_2C_6H_4$), 8.10 (1H, s, C_2-H), 8.32 (1H, d, C_4-H), 9.47 (1H, d, C_8-H).

Methyl 3-(4-aminobutyl)-6-isopropyl-[2-^{14}C]azulene -1-carboxylate. 8

A mixture of 7 (2.35 GBq, 473 mg, 1.10 mmol), 80% hydrazine hydrate (276 mg, 4.40 mmol) in EtOH 15 ml was stirred under reflux for 2 hr. After cooling, the mixture was diluted with n-hexane (15 ml) and filtered off the insoluble material. The filtrate was evaporated in vacuo to give 8 (2.34 GBq, 329.4 mg). ^{1}H-NMR($CDCL_3$) δ = 1.36 (6H, d, i-proCH_3), 1.50-1.80 (4H, m, -CH_2-), 2.30 (2H, brs, -NH_2), 2.60-2.85 (2H, m, -CH_2-), 2.90-3.20 (3H, t+m, -CH_2-, i-proCH), 3.92 (3H, s, -$COOCH_3$), 7.20-7.50 (2H, m, $C_{5,7}$-H), 8.11 (1H, s, C_2-H), 8.30 (1H, d, C_4-H), 9.45 (1H, d, C_8-H).

Methyl 3-{4-(p-chlorobenzenesulfonyaminobutyl)}-6-isopropyl-[2-^{14}C]azulene-1-carboxylate. 9

10% Aqueous sodium hydroxy carbonate (1.5 ml) and p-chlorobenzenesulfonylchloride (348.3 mg, 1.65 mmol) in acetone (1 ml) were added to a solution of 8 (2.34 GBq, 329.4 mg) in acetone (8 ml) and the mixture was stirred at 0°C for 20 min and room temperature for 2 hr. The mixture was concentrated under reduced pressure, extracted with ethyl acetate, and washed with satd. brine, dried and evaporated. The residue was purified by column chromatography on silica with ethyl acetate/n-hexane (1:4) as the eluent, to give 500.7 mg (96.2%) of 9 (2.25 GBq), as a violet oil. ^{1}H-NMR ($CDCL_3$) δ =1.36 (6H, d, i-proCH_3), 1.30-1.45 (2H, m, -CH_2-), 1.45-1.90 (2H, m, -CH_2-), 2.70-3.40 (5H, t+m, -CH_2-, i-proCH), 3.91 (3H, s, -$COOCH_3$), 5.10-

5.30 (1H, m, -N<u>H</u>SO_2-), 7.20-7.50 (2H, m, $C_{5,7}$-H), 7.39 (2H, d,-C_6<u>H_4</u>Cl), 7.70 (2H, d-C_6<u>H_4</u>Cl), 8.03 (1H, s, C_2-H), 8.22 (1H, d, C_4-H), 9.45 (1H, d, C_8-H).

1-{4-P-Chlorobenzenesulfonlaminobutyl)}-6-isopropyl-[2-^{14}C]azulene. <u>10</u>

A mixture of <u>9</u> (2.25 GBq, 500.7 mg, 1.06 mmol) and 100% phosphoric acid (15 mg), was heated at 100°C for 30 min, cooled and poured into water (100 ml), then extracted with ethyl acetate. The extract was dried over anhydrous Na_2SO_4, and evaporated to give a blue violet oil. This crude product was purified by column chromatography on silica with ethyl acetate/<u>n</u>-hexane (1:5) as the eluent, to give 434.3 mg (98.6%) of <u>10</u> (2.22 GBq), as a blue violet oil.

^{1}H-NMR ($CDCL_3$) δ = 1.35 (6H, d, i-proCH_3), 1.50-1.90 (4H, m, -CH_2-), 2.80-3.20 (5H, m, -CH_2-, i-proCH), 4.18-4.40 (1H, m, -N<u>H</u>SO_2), 6.90-7.10 (2H, m, $C_{5,7}$-H), 7.20 (1H, d, C_3-H), 7.40 (2H, d, -C_6<u>H_4</u>Cl), 7.55 (1H, d, C_2-H), 7.70 (2H, d, -C_6<u>H_4</u>Cl), 8.10 (1H, d, C_4-H), 8.20 (1H, d, C_8-H).

Sodium 6-isopropyl-3-[4-(p-chlorobenzenesulfonylamino)-butyl]-[2-^{14}C]azulene-1-sulfonate. <u>1</u>

A mixture of <u>10</u> (2.22 GBq, 434.3 mg, 1.04 mmol) and pyridine-sulfur trioxide (498.6 mg, 3.13 mmol) in benzene (15 ml) was stilled under reflux for 2 hr. The mixture was concentrated under reduced pressure and the resulting solid dissolved in MeOH (15 ml) and 28% NaOMe (1.20 g, 6.24 mmol) was added at 0°C. After stirring at room temperature for 8 hr, the solvent was concentrated under reduced pressure and water (50 ml) was added to the mixture, then extracted with ethyl acetate. The extract was dried over anhydrous Na_2SO_4, and evaporated to give a violet solid. This crude product was purified by column chromatography on silica with chloroform/methyl alcohol (4:1) as the eluent, further passage through a RP-18 Sep PaK with methyl alcohol as the eluent, to give 459 mg (85%) of <u>1</u> (1.90 GBq). The purity was 99% on RHPLC. mp=207°C; IR (KBr) cm^{-1} 1038 (S=O); ^{1}H-NMR (CD_3OD) δ = 1.30 (6H, d, i-proCH_3), 1.20-1.80 (4H, m, -CH_2-), 2.80-3.10 (5H, t+m, -CH_2-, i-proCH), 7.20 (2H, d, $C_{5,7}$-H), 7.32 (2H, d, -C_6<u>H_4</u>Cl), 7.73 (2H, d, -C_6<u>H_4</u>Cl), 7.90 (1H, s, C_2-H), 8.30 (1H, d, C_4-H), 8.90 (1H, d, C_8-H).

REFERENCES

1. Hamberg M., Svensson J. and Samuelsson B. - Proc. Natl. Acad. Sci. 72: 2994 (1975)
2. Bhagwat S. S., Hammam P. R., Still W. C., Bunting S. and Fitzpatrick F. A. - Nature. 315: 511 (1985)
3. Tomiyama T., Wakabayashi S., Kosakai K. and Yokota M. - J. Med. Chem. 30: 2793 (1990).
4. Tomiyama T., Yokota M., Wakabayashi S., Kosakai K. and Yanagisawa T. - J. Med. Chem 36: 791 (1993)
5. Yang P. W., Yasunami M and Takase, K. - Tetrahedron Lett. 4275 (1971)
6. Yasunami M., Chen A., Yang P.W. and Takase K. - Chem. Lett. 579 (1980)
7. Suzaka H., Tomiyama T. and Ikegami S. - J. Labelled comp. and Radiopharm. 8: 901 (1990)
8. Cook F. L., Bowers C. W. and Liotta C. L. - J. Org. Chem. 39: 3416 (1976)
9. Sato T. - Bull. Chem. Soc. Res. Inst. Non-aqueous solution Tohoku Univ. 8: 47 (1959)
10. Tsuji T., Kataoka T., Yoshioka M., Sendo Y., Nishitani Y., Hirai S., Maeda T. and Nagata W. - Tetrahedron Lett 30: 2793 (1979)
11. Mitsunobu O. - Synthesis 1: (1981)