Synthesis of Carbon-14 Labeled LTD_4 Antagonist MK-571

Dennis C. Dean*, David G. Melillo and Robert L. Ellsworth

Merck Sharp and Dohme Research Laboratories
P.O. Box 2000, Rahway, NJ 07865

Summary

The synthesis of (E)-5-(3-(2-(7-chloroquinolin-2-yl)ethenyl)-phenyl)-[5-^{14}C]-4,6-dithianonane dicarboxylic acid N,N-dimethylamide ([^{14}C]MK-571), a high-affinity LTD_4 antagonist, from sodium [^{14}C]cyanide via a five step sequence is described. Condensation of 3-[^{14}C]cyanobenzaldehyde with 7-chloroquinaldine followed by nitrile reduction provided the [^{14}C]aldehyde **2**. The pivotal formation of the penultimate unsymmetrical dithioacetal intermediate was accomplished in a selective manner from aldehyde **2** by way of an O-trimethylsilyl hemiacetal intermediate. Subsequent ester hydrolysis afforded [^{14}C]MK-571 in 14% overall radiochemical yield.

Key Words: [^{14}C]Leukotriene receptor antagonist, synthesis, sodium[^{14}C] cyanide, unsymmetrical dithioacetal, O-trimethylsilyl hemithioacetal.

Introduction

The critical involvement of the cysteinyl leukotrienes (LTC_4, LTD_4, and LTE_4) in the etiology of human bronchial asthma has been well documented.[1,2] In conjunction with a program directed toward the discovery of new leukotriene receptor antagonists,[3] the compound (E)-5-(3-(2-(7-chloroquinolin-2-yl)ethenyl)-phenyl)-4,6-dithianonane dicarboxylic acid N,N-dimethylamide (**1**, MK-571) was found to be a potent and specific LTD_4 antagonist.[4] In support of the developmental process of this promising antiasthmatic agent, carbon-14 labeled material was required for metabolism and distribution studies in animals and man.[5]

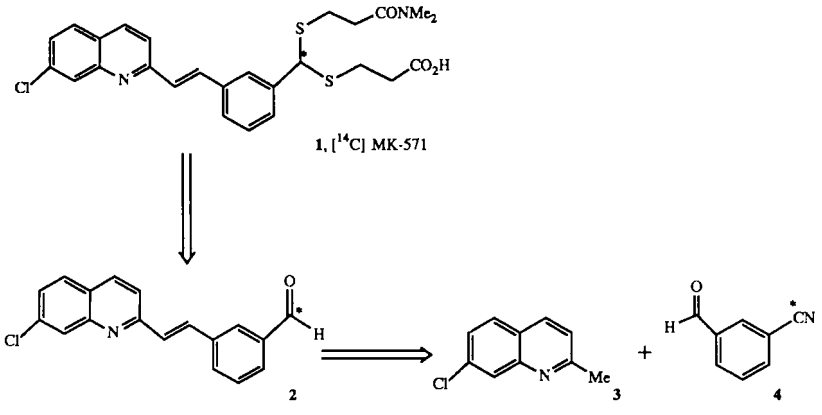
Consideration of a basic retrosynthetic analysis (Scheme 1) suggested preparation of an appropriate labeled aldehyde precursor through the condensation of 7-chloroquinaldine with a 3-[^{14}C]cyanobenzaldehyde followed by subsequent reduction of the nitrile functionality. We chose not to use 1,3-benzenedicarboxaldehyde at this stage in order to localize the ^{14}C-label to the dithioacetal carbon and avoid formation of bis-addition products. Selection of the dithioacetal carbon for label incorporation was also deemed attractive as a position with low potential for degradative metabolism. The efficacy of the synthesis by the course depicted in Scheme 1 hinged on the ability to construct the unsymmetrical dithioacetal moiety from aldehyde in a selective fashion. Fortunately, a recent study by McNamara and coworkers[6,7] formulated a new method to address this problem through initial conversion of **2** to an O-trimethylsilyl hemithioacetal. A second thiol group could then be introduced selectively, taking advantage of

0362–4803/91/020217–06$05.00

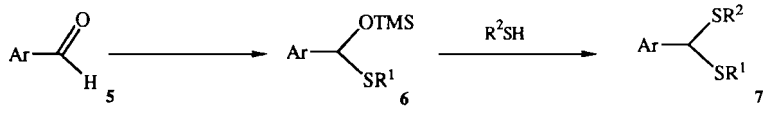
Received August 24, 1990

the greater reactivity of oxygen toward Lewis acid-promoted cleavage relative to sulfur. Herein, we describe the successful implementation of this approach to the synthesis of [^{14}C]MK-571.

Scheme 1



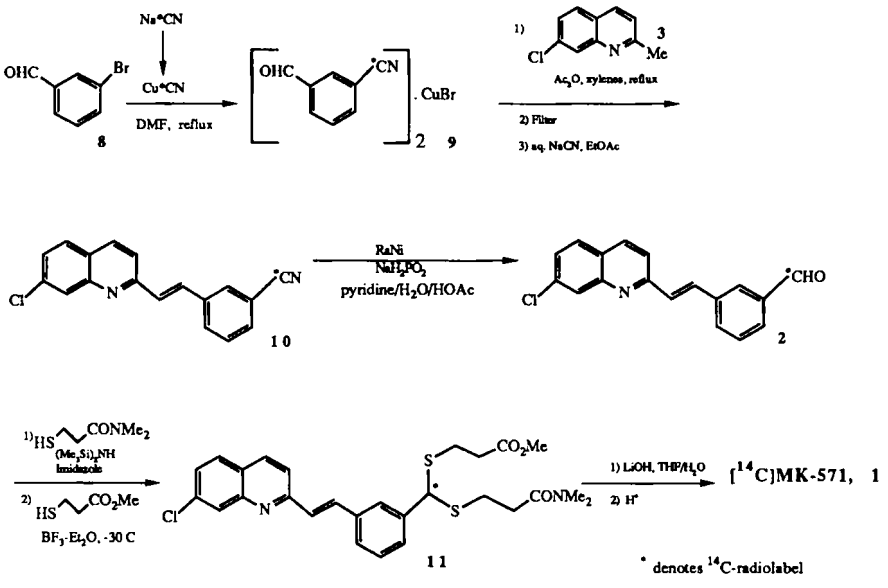
Scheme 2



Results

3-[^{14}C]Cyanobenaldehyde (**4**, Scheme 3) was prepared by reaction of 3-bromobenzaldehyde (**8**) with cuprous [^{14}C]cyanide (formed from sodium [^{14}C]cyanide and cuprous chloride) in refluxing dimethylformamide.[8] Dissociation of the resulting copper complexs of **4** at this stage produced extensive polymerization. Therefore, the crude reaction mixture was diluted with xylene and heated with 7-chloroquinaldine (**3**) and acetic anhydride at 140 °C to afford nitrile **10** as a copper complex. Treatment of this material with 10% aqueous sodium cyanide solution led to smooth complex dissociation and isolation of **10** in 70% overall yield from sodium [^{14}C]cyanide. At this point reduction of the nitrile functionality to the corresponding aldehyde was required. Standard low-temperature DIBAL reduction procedures[9] proved unacceptable due to facile hydride addition to the electrophilic double bond of the unsaturated quinoline. Consequently, a non-nucleophilic reduction method using Raney nickel in the presence of sodium hypophosphite as a mild source of hydrogen[10] was employed to produce the aldehyde **2** in ~40% yield.

Scheme 3



Implementation of the aforementioned protocol for construction of the unsymmetrical dithioacetal was initiated via conversion of aldehyde **2** to the corresponding O-trimethylsilyl hemithioacetal by treatment with 1.1 equivalents of N,N-dimethyl-3-mercaptopropionamide and 1 molar equivalent of 1,1,1,3,3,3-hexamethyldisilazane in the presence of catalytic imidazole using a nitrogen sweep to remove ammonia. The crude product was then reacted with a slight excess of methyl 3-mercaptopropionate in the presence of 3-equivalents of boron trifluoride etherate to afford the desired dithioacetal **11** in 65% overall yield from **2.** The selection of boron trifluoride etherate as Lewis acid was critical, as protic acid or other Lewis acids (such as zinc halides) led to substantial amounts of adducts resulting from conjugate addition of thiol to the unsaturated quinoline.[6] Saponification of **11** with aqueous lithium hydroxide followed by acidic workup provided [^{14}C]MK-571 in 80% yield and 97% radiochemical purity (with a specific activity of 12.3 mCi/mmoL) after final purification by recrystallization from ethanol. The overall radiochemical yield was 14%. This material was found to be particularly sensitive to prolonged exposure to light or heat as a solid or in solution (~10% decrease in purity after 24h at room temperature). Therefore, rapid purification and storage of the labeled drug at -55 ^{o}C was essential.

Experimental

Radioactivity determinations were carried out with a Packard Tri-Carb Model 3320 liquid scintillation counter using 0.42% Omnifluor™ in toluene:ethanol (7:3) as scintillation medium. Analytical TLC was performed using silica gel 60 F-254 (E. Merck) with radioactivity measurements made with a Berthold Model LB2760 scanner. The HPLC system used for analysis consisted of a 4.6 mm x 25 cm Dupont Zorbax RX C-8 column, a Spectra-Physics SP8810 LC pump and

controller, and a Nicolet LC-9563 UV detector. HPLC radioactivity measurements were performed using a Berthold LB-506-C-1 radioactivity monitor and software run on an IBM PS/2 computer. Preparative HPLC was accomplished using a Whatman M20 (22.1 mm x 50 cm) Partisil column.

The identities of each of the labeled intermediates, as well as of the final product (**1**), were established by co-elution via HPLC or TLC of the radio-labeled substance with authentic unlabeled compounds obtained from either Aldrich Chemical Co., Inc. (Milwaukee, Wis.) or from Merck & Co., Inc. (Rahway, N. J.).

(E)-2-(2-(3-[^{14}C]cyanophenyl)ethenyl)-7-Chloroquinoline (10)

A suspension of cuprous [^{14}C]cyanide (1.46 g, 16.4 mmol, 279 mCi, SA=17.01 mCi/mmoL), prepared from sodium [^{14}C]cyanide (SA=54 mCi/mmoL) and cuprous chloride, and freshly distilled 3-bromobenzaldehyde (3.04 g, 16.4 mmol) in N,N-dimethylformamide (7 mL) was stirred at reflux for 5h. The mixture was cooled to room temperature and o-chloroquinaldine (3.2 g, 18.0 mmol), acetic acid (10.8 g, 0.106 mol), and xylene (55 mL) were added. The thick green suspension was heated at reflux for 8h, cooled to room temperature and diluted with hexanes (50 mL). The suspension was filtered and the solid washed with ethyl acetate / hexanes (1:1, 200 mL). The filter cake was dissolved in 10% aqueous sodium cyanide solution (100 mL) and ethyl acetate (100 mL), the two layers separated and the aqueous layer extracted with ethyl acetate (2 X 75 mL). The combined ethyl acetate solutions were washed with 10% aqueous sodium cyanide solution (100 mL), dried over sodium sulfate, and the solvent removed *in vacuo* to provide 4.5 g of a white solid (201 mCi, 71% based on radioactivity) which contained approximately 1g of unreacted 7-chloroquinaldine. Radiochemical analysis by HPLC (Zorbax C-8, methanol : water : trifluoroacetic acid; 30/70/0.1) indicated this material to be 95% **10**. The crude isolate was not further purified and used directly in the subsequent reaction.

(E)-2-(2-(3-[^{14}C]formylphenyl)ethenyl)-7-Chloroquinoline (2)

A solution containing 10 (4.5 g, 201 mCi, 11.2 mmol based on radioactivity) and sodium hypophosphite hydrate (12g, 0.136 mol) in pyridine (80 mL), acetic acid (40 mL), and water (12 mL) was treated with a slurry of water-wet Raney nickel (6.0 g) in pyridine (20 mL). The mixture was heated at 50 ^{o}C for 2h, cooled to room temperature and diluted with ethyl acetate (100 mL). The solution was washed with saturated sodium bicarbonate solution (2 X 50 mL) followed by water (2 X 80 mL) and dried over sodium sulfate. The solvent was removed *in vacuo* and the resulting residue heated at 100 ^{o}C at 0.5 torr to provide a white sublimate (350 mg) which was determined to be 7-chloroquinaldine. The remaining residue was purified by silica gel chromatography using a 25% hexane / chloroform mixture as eluant followed by cyrstallization from ethyl acetate / hexane (2:1) to afford 2 (1.45 g, 78 mCi, 39% radiochemical yield) as a pale yellow solid. Radiochemical analysis by HPLC (RX-C8 column, acetonitrile : water : tetrahydrofuran : trifluoroacetic acid ; 50 : 48 : 2 : 0.1, 1 mL/min) indicated this material to be >98% pure.

Methyl (E)-5-(3-(2-(7-chloroquinolin-2-yl)ethenyl)-phenyl)-[5-^{14}C]-4,6-dithlanonane dicarboxylate N,N-dimethylamide (11)

To a suspension of aldehyde 2 (846 mg, 2.8 mmol, 31 mCi) in anhydrous dichloromethane (10 mL) was added 1,1,1,3,3,3-hexamethyldisilazane (967 mg, 6 mmol), N,N-dimethyl-3-

mercaptopropionamide (465 mg, 3.5 mmol), and imidazole (40 mg, 0.59 mmol) at room temperature under a slow stream of nitrogen (in order to remove ammonia formed in reaction). The reaction was stirred under a gentle stream of nitrogen at room temperature for 18h, filtered and concentrated *in vacuo* to give the crude O-silylated hemithioacetal which had limited stability and was immediately dissolved in nitromethane (20 mL). This solution was cooled to -28 °C and treated with methyl 3-mercaptopropionate (416 mg, 3.5 mmol) followed by dropwise addition of boron trifluoride etherate (1.43 g, 10 .1 mmol) over a 15 min period. The reaction mixture was stirred at -28 °C for 2.5h at which point it was quenched by addition of 10% sodium carbonate solution (35 mL). Additional dichloromethane (25 mL) was added and the layers separated. The organic layer was washed with brine (25 mL), dried over sodium carbonate, and concentrated *in vacuo* at <25 °C. The resulting residue was purified by preparative HPLC reverse phase chromatography (Zorbax M-20 ODS column) using a ternary mixture of 65:20:15 (v/v/v) 0.005 M aqueous dipotassium phosphate solution / acetonitrile / tetrahydrofuran (15 mL/min). Appropriate fractions were combined and concentrated *in vacuo* at <25 °C to remove organic solvent and the aqueous solution extracted with ethyl acetate (3 X 25 mL). The combined ethyl acetate solutions were dried over sodium sulfate and concentrated *in vacuo* at room temperature to yield 11 (980 mg, 20 mCi, 64% based on radioactivity). This material was further purified by recyrstallization from ethanol. Radiochemical analysis by HPLC (RX-C8 column, acetonitrile : 0.005M dipotassium phosphate solution, pH 8 : tetrahydrofuran ; 2:2:1, 1 mL/min) indicated this material to be >98% pure.

(E)-5-(3-(2-(7-chloroquinolin-2-yl)ethenyl)-phenyl)-[5-^{14}C]-4,6-dithianonane dicarboxylic acid N,N-dimethylamide ([^{14}C]MK-571)

To a solution of the ester-amide 11 (874 mg, 1.65 mmol, 20.0 mCi) in tetrahydrofuran (10 mL) at 0 °C was added a 1.0N solution of lithium hydroxide (50 mL, 1.73 mmol) dropwise over a 20 min period. The reaction mixture was stirred at 0 °C for 5.5h, diluted with ice water (14 mL) and the tetrahydrofuran was removed *in vacuo* at <25 °C. The resulting aqueous concentrate (pH=9.5) was extracted with ethyl acetate (3 X 40 mL); an additional portion of ethyl acetate was added (30 mL), and the pH was adjusted to 4.0 with 2N HCl solution. The ethyl acetate layer was removed and the aqueous solution extracted with additional ethyl acetate (2 X 20 mL). The combined pH 4 ethyl acetate extracts were concentrated *in vacuo* at 35 °C and the resulting oily residue dissolved in isopropyl alcohol (25 mL) and distilled *in vacuo* (100 torr) at 50 °C. This process was repeated two times to remove any water present. The solid residue was suspended in absolute ethanol (22 mL) and heated to reflux to effect dissolution. The solution was cooled to 35 °C, 10 mg of non-labeled MK-571 was added as crystal seed, and the crystallization mixture aged at room temperature for 18h in the dark with stirring. The suspension was filtered and the resulting material subjected to recyrstallization as described above to yield [^{14}C]MK-571 (690 mg, 16.1 mCi, SA=12.1 mCi/mmoL, 80% based on radioactivity) as a pale yellow crystalline solid. Radiochemical analysis by HPLC (RX-C8 column, acetonitrile: tetrahydrofuran: water: trifluoroacetic acid; 25:25:50:0.1, 1 mL/min) indicated a radiochemical purity of 97.3%. This material had limited stability with respect to light and temperature, and was therefore stored protected from light at -55 °C.

Acknowledgements

We thank Dr. Allen Jones for performing radioanalytical determinations on the final product and Mrs. Stephen O'Connor and Gregory Gatto for assisting in the preparation of key intermediates. Helpful discussions with Drs. J. M. McNamara, M. Bhupathy, and P.J. Reider and Mr. J. S. Amato of Process Research are also gratefully acknowledged

References and Notes

1. Samuelsson, B.; Dahlen, S.; Lindgren, J.A.; Rouzer, C.A.; Serhan, C.N. - *Science* 237: 1171 (1987)
2. Maron, Z.; Shelhamer, J.H.; Bach, M.K.; Morton, D.R.; Kaliner, M. - *Am. Rev. Respir. Dis.* 126: 449 (1982)
3. Young, R.N.; Guindon, Y.; Jones, T.R.; Ford-Hutchinson, A.W.; Belanger, P.; Champion, E.; Charette, L.; DeHaven, R.N.; Denis, D.; Fortin, R.; Frenette, R.; Gauthier, J.Y.; Gillard, J.W.; Kakushima, M.; Letts, L.G.; Masson, P.; Maycock, A.; McFarlane, C.; Piechuta, H.; Pong, S.S.; Rosethal, A.; Williams, H.; Zamboni, R.; Yoakim, C.; Rokach, J. - *Adv. Prostaglandin Thromboxane Leukotriene Res.* 16: 37 (1986)
4. Jones, T.R.; Zamboni, R.; Belley, M.; Champion, E.; Charette, L.; Ford- Hutchinson, A.W.; Frenette, R.; Gauthier, J.Y.; Leger, S.; Masson, P.; McFarlane, C.S.; Piechuta, H.; Rokach, J.; Williams, H.; Young, R.N.; DeHaven, R.N.; Pong, S.S. - *Can. J. Physiol. Pharmacol.* 67 : 17 (1989)
5. For a prior synthesis of [^{35}S]-labelled MK-571, see: Williams, H.W.R.; Young, R.N.; Zamboni, R.; Shaw, D.R.D. - *J. Labelled Compounds & Radiopharm.* 28: 297 (1989)
6. McNamara, J.M.; Leazer, J.L.; Bhupathy, M.; Amato, J.S.; Reamer, R.A.; Reider, P.J.; Grabowski, E.J.J. - *J. Org. Chem.* 54 : 3718 (1989)
7. For alternative methods of unsymmetrical dithioacetal synthesis, see: Gauthier, J.Y.; Henien, T.; Lo, L.; Therien, M.; Young, R.N. - *Tetrahedron Lett.* 29: 6729 (1988) (and references 5-10 therein).
8. Friedman, L. ; Shechter, H. - *J. Org. Chem.* 26: 2522 (1960)
9. Winterfeldt, E. - *Synthesis* : 617 (1975)
10. Backeberg, O.G.; Staskun, B. - *J. Chem. Soc* : 3961 (1962)