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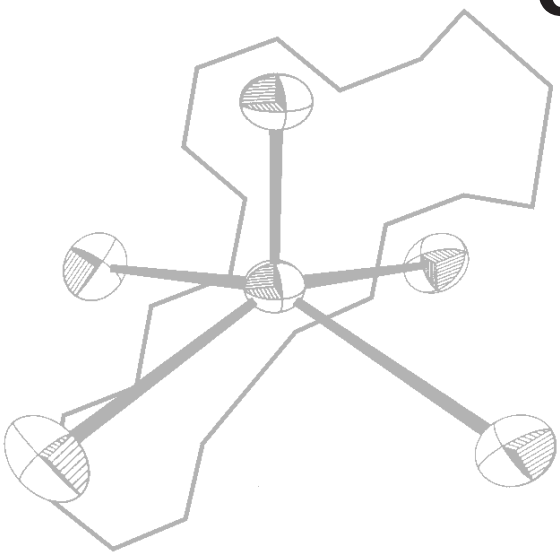

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Improved Epoxidation Methodology for Synthesis of the Highly Selective β_2-Adrenoceptor Antagonist ICI 118551 [*erythro* (±)-3-Isopropylamino-1-(7-methylindan-4-yloxy)butan-2-ol]

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In this communication we describe a much improved methodology for the synthesis of the selective β_2-adrenoceptor antagonist ICI 118551 (1), a procedure which overcomes capricious fractional crystallization and epoxidation methodologies described for its original preparation. Our approach involving a bromohydrin precursor to the key epoxide intermediate (7) yielded an 85 : 15 mixture of the *threo*/*erythro* isomers of (7) which could be conveniently separated by flash chromatography on amine-pretreated silica. This new approach proved much more successful than attempts to separate the precursor alkene isomers (6) by fractional crystallization as described in the original patent literature. The product (1) obtained by using our methodology was found to have identical pharmacological properties to authentic ICI 118551 when tested both *in vitro* and *in vivo*.

Introduction

Over the past decade, it has been recognized that long-acting β_2-adrenoceptor (β_2-AR) agonists such as clenbuterol and cimaterol cause carcass repartitioning effects in animals and in humans, resulting from concomitant increases in protein accretion and lipolysis.[1] Our group has undertaken a number of studies aimed at determining the mechanisms of β_2-agonist action in muscle,[2–6] and to ascertain the functional role of β_3-adrenoceptors.[7–9] In a recent study we needed to effect selective β_2-AR blockade in cattle,[10] which required prohibitively expensive quantities of ICI 118551 [*erythro* (±)-3-isopropylamino-1-(7-methylindan-4-yloxy)butan-2-ol] (1). Compound (1) is the most selective β_2-antagonist yet identified, and has also attracted attention for its inverse-agonist properties.[11]

Synthetic methodology for preparation of the target compound (1) is reported in the patent literature,[12] and an interesting anecdotal account of the problems encountered in the industrial-scale preparation of the drug has been provided by Hutton.[13] The synthesis (Fig. 1) commences with 6-methylcoumarin (2), which is first catalytically reduced to the chromen-2-one (3), and this intermediate then subjected to Lewis acid catalysed rearrangement to produce the indanone derivative (4). The indanone carbonyl group of (4) is then removed reductively, and the resultant indanol (5) reacted with crotyl chloride to afford the key alkenyloxy intermediate (6).

Results and Discussion

Synthesis

In our hands, the four-step conversion[12,13] of (2) into an 85 : 15 *trans/cis* mixture of (6) was achieved in 67% overall yield. While Hutton had reported that the isomers of (6) could be separated by low-temperature fractional crystallization,[13] we found this approach unsatisfactory. Instead we chose to epoxidize the isomeric mixture and then to attempt separation of the corresponding *threo* and *erythro* epoxides.

Epoxidation of (6) using the peroxyimidate methodology described by Hutton[13] proved capricious. The patent report describes the *in-situ* preparation of the peroxyimidate derived from acetonitrile and 50% H_2O_2 in the presence of potassium carbonate, although no yield was reported for the 5-day reaction with (6).[12] For the industrial-scale synthesis of (1), the

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corresponding benzonitrile peroxyimidate was found to react more rapidly with (6); however, the large amount of benzamide side product caused problems in the clean-up of the final product.[13] In our hands, neither *m*-chloroperbenzoic acid nor the peroxyimidate approaches gave satisfactory results (typical yield 30%).

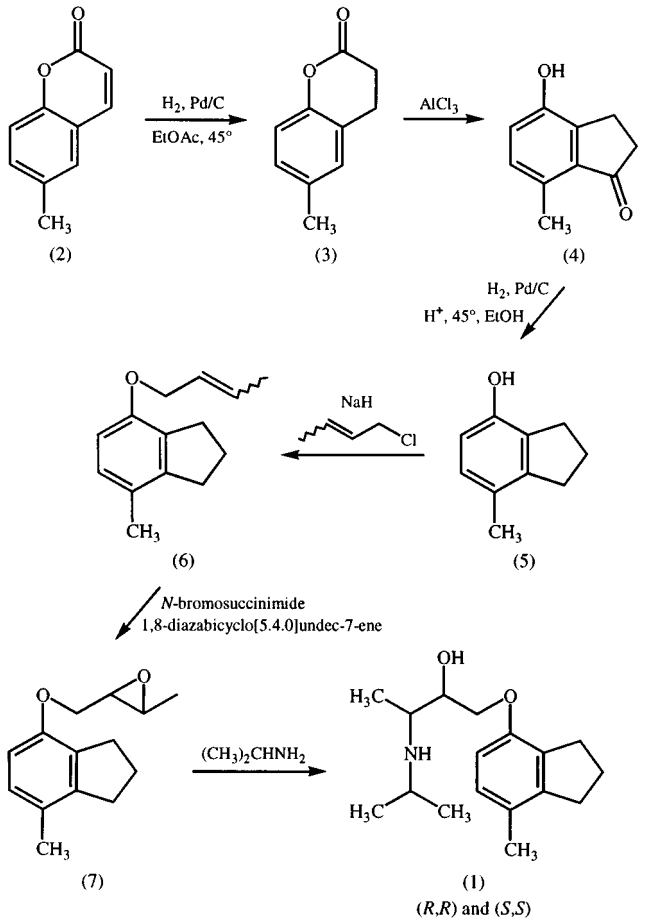

Fig. 1. Synthetic route to ICI 118551.

After some experimentation, we found that the required *threo* epoxide (7) could be obtained routinely in 75% yield from isomeric (6) after a two-step process and subsequent chromatographic purification. Hence the 85 : 15 *trans*/*cis* mixture of (6) was reacted with *N*-bromosuccinimide in a 1,2-dimethoxyethane/water mixture to afford the bromohydrin, which was then treated with 1,8-diazabicyclo[5.4.0]undec-7-ene in 1,2-dimethoxyethane solvent. The resultant 85 : 15 mixture of *threo* and *erythro* epoxides (7) was then subjected to flash chromatography on silica which had been pretreated with 1% triethylamine in 1 : 20 ethyl acetate–hexane. Base pre-treatment of the silica in this manner prevented decomposition of the epoxide during the chromatographic separation of the isomers. The *threo* epoxide (7) was then reacted with isopropylamine in methanol[12,13] to yield the free base (1). The product obtained was found to be spectroscopically identical to an authentic sample of ICI 118551 (1).

Biological Results

The affinity of (1) for β_2-adrenoceptors was determined by using *in-vitro* organ bath techniques. A bovine tracheal strip maintained in oxygenated Tyrode's solution was first contracted with carbachol, and then relaxation of the tissue was achieved through increasing concentrations of isoprenaline. Pre-incubation of the tissue with either 0·1 or 1·0 μM CGP 20712A did not affect the isoprenaline concentration–response curve significantly, indicating few functional β_1-adrenoceptors in the preparation. In contrast, a reference sample of ICI 118551 (ICI Pharmaceuticals, Macclesfield, U.K.) produced concentration-dependent rightward shifts of the isoprenaline response curves, indicating that the drug was antagonizing the β_2-AR mediated relaxation induced by isoprenaline. Schild plot analysis produced a pA_2 value of 7·76±0·12 (mean±s.e.m.) for ICI 118551. Likewise, compound (1) synthesized by the methodology described herein produced rightward shifts of similar magnitudes, giving a pA_2 value of 7·65±0·10, which was not significantly different to that found for the reference ICI 118551 sample.

Conclusions

Our synthetic approach to the key *threo* epoxide intermediate (7), via the bromohydrin, overcomes the low yield/purity problems associated with the peroxyimidate epoxidation methodology described for this conversion in the patent literature.[12] Moreover, we have developed a convenient chromatographic method for separating the required *threo* isomer of (7) from the minor *erythro* isomer. This strategy provides an alternative to separation of the precursor alkene isomers (6) by low-temperature fractional crystallization.[12] The procedures described in this communication have been used to prepare several 20-g batches of ICI 118551 (1) in high purity.

Experimental

General

^{1}H and ^{13}C n.m.r. spectra of $CDCl_3$ solutions of test compounds were obtained by using a Bruker AMX-300 spectrometer. Chemical shifts are quoted as δ values relative to tetramethylsilane. Infrared spectra were recorded with a Perkin–Elmer 1600 Fourier-transform i.r. spectrophotometer. U.v. spectra were obtained with a Varian DMS-100 spectrophotometer and routine mass spectra were obtained by using a Shimadzu QP2000 gas chromatography–mass spectrometer incorporating a direct insertion probe. High-resolution mass spectrometry data were kindly provided by Dr John MacLeod of the Research School of Chemistry, Australian National University, Canberra, Australia. Analytical thin-layer chromatography employed Merck Kieselgel G 60F 254 plates. Pressure-assisted flash chromatography was conducted with Merck silica gel 60, 230–400 mesh. Elemental analyses were performed either by the Canadian Microanalytical Service, Delta, BC, Canada, or by the Microanalytical service at the Australian National University, Canberra, Australia.

Preparation of 2-Methyl-3-(7-methylindan-4-yloxymethyl)oxiran (7) by the N*-Bromosuccinimide/1,8-Diazabicyclo[5.4.0]undec-7-ene Methodology*

Isomeric (6)[12,13] (3·0 g, 14·9 mmol, obtained from 6-methylcoumarin in 67% overall yield) was dissolved in 1,2-dimethoxyethane (69 ml) and water (23 ml); *N*-bromosuccinimide (2·78 g, 15·6 mmol) was added, and the reaction mixture was stirred in the dark for 24 h. Ether (150 ml) was then added and, after partitioning, the ether layer was washed with water (40 ml), dried and filtered. Concentration of the filtrate under vacuum afforded a crude residue which was then taken up in dry 1,2-dimethoxyethane (50 ml) and 1,8-diazabicyclo[5.4.0]undec-7-ene (2·28 g, 15·3 mmol) was added. The reaction mixture was stirred under a nitrogen atmosphere for 6 h. The reaction mixture was then poured into ether (150 ml), washed with water, dried and filtered. Concentration of the filtrate under vacuum and flash chromatography of the residue on silica pretreated with 1% triethylamine in ethyl acetate–hexane (1 : 20), afforded the crystalline epoxide as a colourless solid (2·34 g, 73%), m.p. 61·0–61·5°. ^{1}H n.m.r. δ 1·39, d, J 2·91 Hz, 3H; 2·10, quintet, J 7·47 Hz, 2H; 2·22, s, 3H; 2·85, t, J 7·47 Hz, 2H; 2·93, t, J 7·47 Hz, 2H; 3·06, m, 2H; 4·01, dd, J 5·10, 11·1 Hz, 1H; 4·17, dd, J 3·38, 11·1 Hz, 1H; 6·60, d, J 8·13 Hz, 1H; 6·92, d, J 8·13 Hz, 1H. ^{13}C n.m.r. δ 17·29, 18·34, 24·47, 29·73, 31·90, 52·50, 57·33, 68·39, 109·56, 126·47, 127·77, 131·80, 144·95, 153·14. I.r. (Nujol) 1607, 1495, 1308, 1264, 1079, 868, 801 cm^{-1}. Mass spectrum m/z 218 (17%), 148 (60), 133 (58), 91 (30), 57 (55), 43 (100) (Found: C, 76·9; H, 8·3. Calc. for $C_{14}H_{18}O_2$: C, 77·0; H, 8·3%).

Synthesis of ICI 118551

This oxiran (7) was converted into the title compound (1) by reaction with isopropylamine in methanol.[12] The product (1) was found to be spectroscopically identical to authentic ICI 118551 obtained from ICI Pharmaceuticals, Macclesfield, U.K.

Biological Testing

Bovine trachea were obtained within 30 min of death from a slaughterhouse and transported to the laboratory in cold aerated Tyrode's solution (in mM: NaCl, 136·9; KCl, 5·4; $MgCl_2.H_2O$, 1·05; $NaH_2PO_4.2H_2O$, 0·42; $NaHCO_3$, 22·6; $CaCl_2.2H_2O$, 1·8; glucose, 5·5; ascorbic acid, 0·28). The smooth muscle layer of the trachea was dissected free from the cartilage and epithelial layer, and cut into strips approximately 2 mm wide and 6 mm long. A small stainless steel hook was placed in one end to connect the tissue to the tissue holder, and a silk thread connected the tissue at the other end to a force transducer (Grass FT03, Quincy, MA, U.S.A.). Force of contraction was recorded with a MacLab system (AD Instruments, Cannon Hill, Australia) by a Macintosh LC 475 computer. The preparations were suspended under optimum preload in 25-ml water-jacketed organ baths (35±0·5°) in Tyrode's solution aerated with carbogen (95% O_2, 5% CO_2), and allowed to equilibrate for 60 min with regular replacement of the Tyrode's solution. Indomethacin (3 μM) and corticosterone (100 μM) were added to block the uptake of isoprenaline. Next, the preparations were contracted with 1 μM carbachol until a stable sub-maximal contraction was obtained (2 h). The β_1- or β_2-AR selective antagonists CGP 20712A and ICI 118551 were added and allowed to equilibrate for 30 min before initiation of relaxation. Isoprenaline was added cumulatively at 8-min intervals when the effect of the previous addition (8 min) had reached equilibrium. This induced a concentration-dependent relaxation from which pA_2 values were determined by Schild plot analysis.

CGP 20712A (Ciba–Geigy, Basel, Switzerland) and ICI 118551 (Imperial Chemical Industries, Cheshire, England) were dissolved in dimethyl sulfoxide. Isoprenaline was obtained from Sigma Chemical Company (Sigma, St Louis, MO).

Acknowledgments

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