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Research Article

Improved synthesis of $^{13}C,^{2}H_3$- and $^{2}H_3$-salmeterol by Cs_2CO_3-mediated monoalkylation of a primary amine

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Summary

An abbreviated synthesis of isotopically labelled salmeterol has been achieved. The key improvement utilizes a highly selective Cs_2CO_3-mediated one-pot alkylation of benzylamine by 6-bromo-1-(4′-phenylbutoxy)hexane to prepare the limiting reagent, 6-*N*-benzylamino-1-(4′-phenylbutoxy)hexane without overalkylation. The method was applied to synthesis of the title compounds in >97 at% isotopic purity. Copyright © 2002 John Wiley & Sons, Ltd.

Key Words: phenethylamine; bronchodilator; deuterium; carbon-13; mass spectrometry

Introduction

Synthesis of pharmacologically active *N*-alkylated secondary amines by direct alkylation is often impractical owing to problems of over-alkylation and separation of intractable mixtures of secondary, tertiary and quaternary amines. For example, one preparation of the

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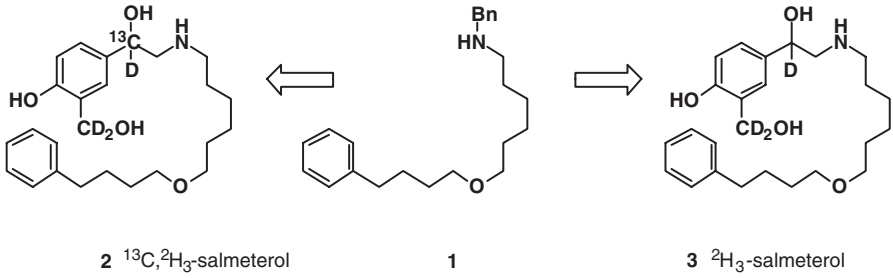


2 $^{13}C,^{2}H_3$-salmeterol 1 3 $^{2}H_3$-salmeterol

Figure 1. Deuterium-labelled salmeterol

bronchodilatory β-adrenergic agonist, salmeterol[1] (Figure 1), uses a work around for the synthesis of the key *N*-benzyl secondary amine **1.** This is achieved by conversion of the primary alkylating agent to the corresponding azide by S_N2 displacement, followed by reduction then conversion to an *N*-benzyl amine by reductive amination with benzaldehyde in the presence of $NaBH_4$ or $NaCNBH_3$.

An alternative method for selective *N*-alkylation relies upon alkylation of an imine, prepared by condensation of the primary amine with an aldehyde, followed by hydrolysis of the resultant *N*,*N*-dialkyliminium salt to the desired secondary amine.[2] Both methods, and other alternatives (for example, alkylation of N-alkylarenesulfonamides followed by hydrolysis),[3] add 1–4 steps to the procedure to achieve the desired outcome-controlled formation of a secondary *N*-benzylamine. The key intermediate *N*-benzylamine **1** (Figure 1),[4] employed in the synthesis of salmeterol, is prepared from commercially available 4-phenylbutanol and 1,6-dibromohexane from operations requiring five steps.[5] We needed a rapid preparation of isotopically $^{13}C,^{2}H$-labelled salmeterol (^{13}C-$^{2}H_3$-**2** and $^{2}H_3$-**3**) for use as internal standards in analytical LC–MS of equine plasma and urine samples. To this end, we sought a one-step method for the preparation of **1** by selective monoalkylation of benzylamine for subsequent conversion to **2** and **3** (Figure 1).

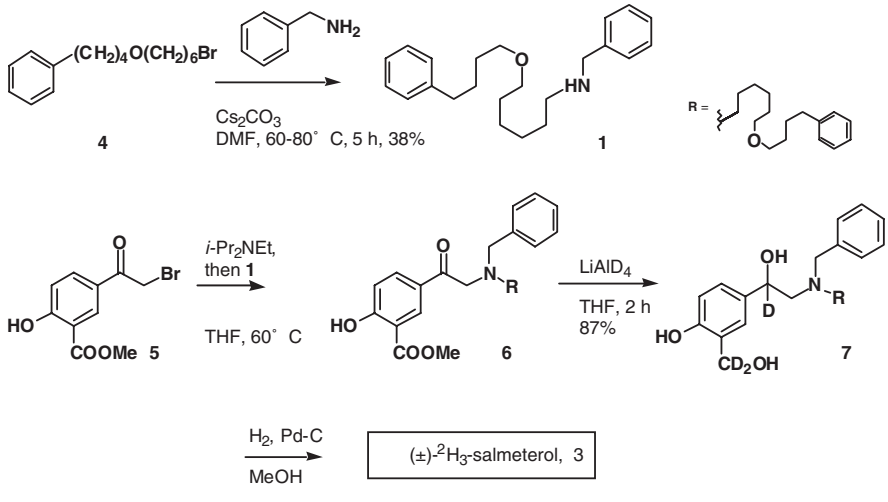
Reports describing chemoselective monoalkylation of primary amines in the presence of the bases Cs_2CO_3 and CsOH and DMF have appeared recently.[6] The improved selectivity for monoalkylation appears to involve initial tight coordination of Cs^+ to the primary amine. Salvatore *et al.* have explored the scope of the reaction and propose a mechanism in which the Cs-coordinated primary amine is deprotonated in the basic medium to give a reactive Cs-amide base.[7] The latter is rapidly alkylated, but further alkylation is prevented by the

steric hindrance to the bulky Cs-coordinated secondary amine product. We found that in the presence of Cs_2CO_3 direct alkylation of the primary bromide **4** gave **1** in one step. This method will find application in other syntheses of secondary and tertiary amines that are prone to the complications of multiple alkylation and mixtures of inseparable products.

Results and discussion

6-Bromo-1-(4′-phenylbutoxy)hexane (**4**, Scheme 1) was prepared by phase-transfer catalyzed alkylation of 4-phenylbutan-1-ol with 1,6-dibromohexane (2 eq, 10 M NaOH, aq, CH_2Cl_2, 5 mol% each of n-Bu_4NHSO_4 and $BnEt_3NCl$, rt, 8 days, 51%). Purification of crude mixture was best achieved by short-passage chromatography over TLC-grade silica gel. Unreacted starting materials and *bis*-ether byproduct were eluted with *n*-hexane followed by 5% MTBE/*n*-hexane which provided pure **4**. Controlled alkylation of **4** with benzylamine (1.1 eq) was smoothly carried out using Cs_2CO_3–DMF (60–80°C) to give **1** (38%, pure, unoptimized) uncontaminated by overalkylation products. The rest of the material was comprised of starting materials, which could be recovered by chromatography.

The unreported deuterium labelled analog **3** was prepared as follows (Scheme 1). Methyl α-bromo 4-acetylsalicylate (**5**), prepared by



Scheme 1. Synthesis of deuterium-labelled salmeterols 2 and 3

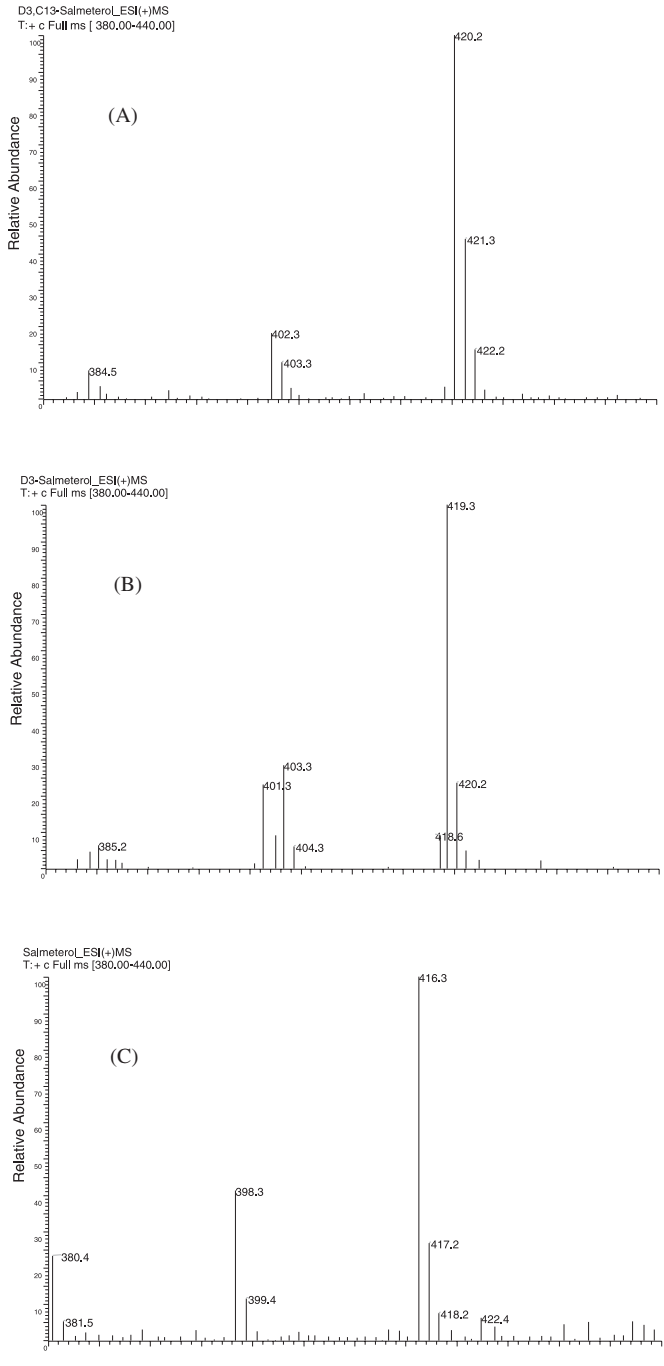


Figure 2. Full scan ESI mass spectra recorded in positive-ion mode after direct sample infusion (see Experimental). Positive-ion mode. (A) $^{13}C,^{2}H_3$-salmeterol (2); (B) $^{2}H_3$-salmeterol (3); and (C) unlabelled salmeterol.

bromination of methyl 4-acetylsalicylate (Br_2, $CHCl_3$), was alkylated with **1** (*i*-Pr_2EtN, THF, 60°C, quantitative) to give **6** which was immediately reduced ($LiAlD_4$) to the corresponding $^2H_3$-alcohol **7**. Hydrogenolysis of **7** (10% Pd–C, H_2) provided pure ($\pm$)-$^2H_3$-salmeterol (**3**) as a clear oil after preparative TLC. The isotopic composition of **3**, as measured by ESI MS, was 98% d_3, 1.5% d_2 and negligible amounts of lower isotopomers.

Synthesis of the known compound **2** followed the method of Goodwin *et al.*[8] Methyl 4-(1-^{13}C-α-bromoacetyl)salicylate, prepared by Friedel–Crafts acylation ($AlCl_3$) of methyl salicylate with 1-^{13}C acetyl chloride (Aldrich, >99 at%) followed by α-bromination,[8] was alkylated with **1**, followed by reduction with $LiAlD_4$ and debenzylation (Pd–C, H_2) to give **2** (>98% d_3, ESIMS), identical in every respect to the reported material.[8] Examination of the ESI mass spectra of **2, 3** and unlabelled salmeterol (**1**) (Figure 2) confirmed that the compositions for each compound were >97% isotopic purity.

Conclusion

In summary, samples of deuterated **2** and the new salmeterol derivative **3** were prepared from methyl 4-acetylsalicylate in good yield by employing a chemoselective Cs^+-mediated monoalkylation of benzylamine. The preparation of the limiting reagent **1** from 4-phenylbutanol was shortened to two steps (*cf* published five-step procedure)[5]. This useful modification can be adapted to recently disclosed syntheses of enantio-enriched salmeterol,[9,5] and preparation of *N*-alkylated phenethylamines substituted with radioisotope labels in the *N*-alkyl side chain.[10] In the latter case, the ability to prepare labelled material with minimum manipulation is most desirable.

Experimental

CH_2Cl_2 and DMF were distilled from CaH_2 and stored under N_2. THF was distilled under N_2 from sodium-benzophenone ketyl. Cs_2CO_3 was dried overnight under high vacuum at 56°C. Compounds were purified by preparative chromatography using flash chromatography[11] or using vacuum-assisted chromatography on TLC-grade silica. Preparative TLC was carried out using $20 \times 20 \times 0.2\,cm^3$ plates (E.M. Merck grade

7749). ESI MS spectra were recorded on a ThermoFinnigan LCQ ion trap. LC–MS analysis was conducted by direct infusion using a mixed solvent (0.1% HCO_2H, 20% CH_3CN, 80% H_2O) with a flow rate of 0.5 ml/min using full scan mode (MS range 150–2000 amu). ^1H NMR spectra were recorded in Varian Inova 400 or Mercury 300 NMR spectrometers in $CDCl_3$ and referenced to residual solvent signals, $\delta_H = 7.26$ ppm and $\delta_C = 77.00$ ppm.

6-bromo-1-(4′-phenylbutoxy)hexane (***4***)

A solution of 4-phenylbutanol (1.50 g, 10 mmol), 1,6-dibromohexane (4.88 g, 20.0 mmol) and tetra-*n*-butylammonium hydrogen sulphate (169 mg, 0.50 mmol) in CH_2Cl_2 (10 ml) was treated with 10 M NaOH aq (1.0 ml) and stirred vigorously for 9 h. The mixture was treated with benzyltriethylammonium chloride (5 mol%) and stirred at room temperature for a total of 8 days, then diluted with CH_2Cl_2. After washing with H_2O, the organic phase was dried (Na_2SO_4) and concentrated to give a crude product that was separated by silica chromatography (TLC grade). Excess dibromide and *bis*-ether byproduct were removed by elution with *n*-hexane and pure **4** (51%) was recovered after elution with 5% MTBE/*n*-hexane. The product gave ^1H NMR and MS data identical with reported values.[4]

Monoalkylation of 6-bromo-1-(4′-phenylbutoxy)hexane with benzylamine in the presence of Cs_2CO_3: 6-N-benzylamino-1-(4′-phenylbutoxy)hexane (***1***)

Anhydrous Cs_2CO_3 (2.27 g, 7.0 mmol) was added to a stirred solution of **4** (1.82 g, 5.8 mmol) and benzylamine (0.69 g, 6.4 mmol) in dry DMF (29 ml) and the mixture was stirred at 60–80°C for 5 h. The mixture was poured into ice–$NaHCO_3$ aq (1 M) and extracted with CH_2Cl_2 (3×100 ml). The combined organic phases were washed with H_2O, dried (Na_2SO_4) and concentrated. The residue (2.00 g) was dried under high vacuum to remove DMF and applied to a short, wide column of TLC-grade silica and eluted with 2–5% MeOH (NH_3-saturated)/CH_2Cl_2 to obtain product **1** as a clear oil (0.75 g, 38%). This oil displayed a single ninhydrin-positive spot by TLC and a ^1H NMR spectrum consistent with the reported values.[4]

(±)-^{13}C,$^{2}H_3$-salmeterol (**2**)

This compound was prepared from **1** and the ^{13}C-labelled analog of **5** as described by Goodwin *et al.*[8] ESI MS *m/z* $[M+H]^+$ 420 (98% d_3), 419 (1.5% d_2), 418 (0.5% *d*). Calcd. for $C_{24}^{13}CH_{35}^{2}H_3NO_4$, 420.

(±)-$^{2}H_3$-salmeterol (**3**)

A solution of compound **6**[8] (90 mg, 0.17 mmol) in THF (4 ml) was cooled in an ice-bath and treated with $LiAlD_4$ (98 at % ^{2}H, 109 mg) in portions for over 30 min then allowed to warm to room temperature over 19 h. The mixture was cooled again and cautiously treated with a few drops of water followed by 1.2 M HCl (10 ml). The mixture was treated with the theoretical amount of solid $NaHCO_3$ required to neutralize the acid, adjusted to pH 7, then diluted with H_2O (50 ml) and extracted with CH_2Cl_2 (3 × 50 ml). The combined extracts were washed with water, dried (Na_2SO_4) and concentrated to give product **7** as a pale oil (87%). An analytical sample was prepared by preparative TLC (0.01:1:1 NH_4OH aq/*n*-hexane/EtOAc). ^{1}H NMR ($CDCl_3$) 7.4–7.1 (m, 10 H, ArH), 6.98 (dd, 1 H, J = 8.6, 2 Hz), 6.91 (d, 1 H, J = 2 Hz), 6.78 (d, 1 H, J = 8.6 Hz), 4.00 (bd, 1 H, $CH(OH)CH_2N$), 3.80 (m, 1 H, $CH(OH)CH_2N$), 3.39 (t, 2 H, OCH_2), 3.35 (t, 2 H, OCH_2), 2.7 (m, NCH_2), 2.61 (t, 2 H, J = 7.2 Hz, $ArCH_2$), 1.7–1.2 (m, 12 H, 6 × CH_2). ESI MS *m/z* $[M+H]^+$ 509 (98% d_3), 508 (2% d_2), 507 (< 0.1% d_1). Calcd. for $C_{32}H_{41}$ $^2H_3NO_4$ 509.

A solution of **7** (28 mg) in ethanol (20 ml) was hydrogenated in the presence of 10% Pd–C (20 mg, 1 atm H_2) for 4 h. The catalyst was removed by filtration through a Celite pad. Removal of the solvent gave a residue which was purified by preparative TLC (developed with 60:8:1 CH_2Cl_2/MeOH/NH_4OH aq) to provide (±)-$^2H_3$-salmeterol, **3** (17 mg, 77%). ESI MS *m/z* $[M+H]^+$ 419 (97.9% d_3), 418 (1.6% d_2), 417 (0.5% *d*). Calcd. for $C_{25}H_{35}$ $^2H_3NO_4$, 419.

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References

1. (a) Finch H, Naylor A, Skidmore IF, Lunts LHC. *US Patent* 1991; **4**: 992,474; (b) Skidmore IF, Lunts LHC, Finch H, Naylor A. *Ger Offen* 1984; **3**: 414, 752.
2. Wawzonek S, McKillip W, Petersen CJ., Baumgarten H (ed). *Organic Syntheses: Collective* vol 5. Wiley, New York, 1973.
3. (a) Hinsberg O. *Ann* 1891; **265**: 178; (b) Lecher H, Graf F. *Ann* 1925; **445**: 68.
4. Nangia A, Praveen P, Dubey, AK. *Ind J Chem* 1995; **34B**: 629.
5. Hett R, Stare R, Helquist P. *Tetrahedron Lett* 1994: **35**: 9375.
6. (a) Salvatore RN, Nagle AS, Schmidt SE, Jung KW. *Org Lett* 1999: **1**: 1893; (b) Salvatore RN, Nagle AS, Schmidt SE, *et al*. *Tetrahedron Lett* 2000: **41**: 9705.
7. Salvatore RN, Nagle AS, Jung KW. *J Org Chem* 2002: **67**: 674.
8. Goodwin TE, Zhou X, Haile PA, *et al*. *J Label Compd Radiopharm* 2000: **43**: 65.
9. Procopiou PA, Morton GE, Todd M, Webb G. *Tetrahedron: Asymmetry* 2001: **12**: 2005.
10. Bowers GD, Bayliss MK, Donnelly MC, Fellows I, Ismail IM, Mookerjee CR. *J Pharm Biomed Anal* 1998: **18**: 461.
11. Still WC, Kahu M, Mitra A. *J Org Chem* 1978: **43**: 2923.

