Stereoselective synthesis of (2*S*,3*R*)- and (2*R*,3*S*)-iodoreboxetine; potential SPECT imaging agents for the noradrenaline transporter†

Nicola K. Jobson,[a] Rosemary Spike,[b] Andrew R. Crawford,[b] Deborah Dewar,[b] Sally L. Pimlott[c] and Andrew Sutherland*[a]

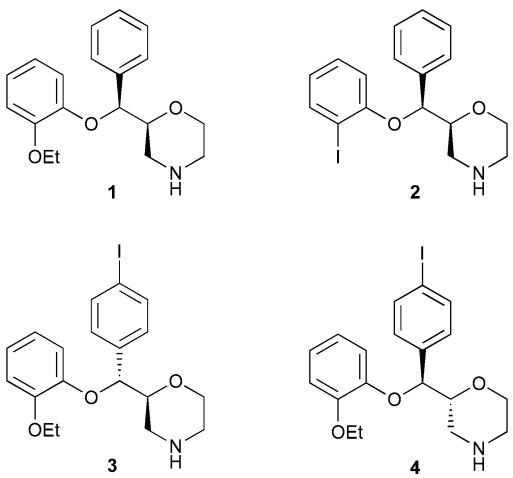
Received 19th February 2008, Accepted 25th March 2008
First published as an Advance Article on the web 30th April 2008
DOI: 10.1039/b802819b

With the aim of developing a new SPECT imaging agent for the noradrenaline transporter, a twelve-step stereoselective synthesis of iodinated analogues of (2*S*,3*R*)- and (2*R*,3*S*)-reboxetine has been achieved from 4-bromobenzaldehyde. The key steps involve a Sharpless asymmetric epoxidation to establish the stereogenic centres and a copper catalysed aromatic halogen exchange reaction to introduce the key iodine atom. *In vitro* testing of these compounds using a [^{3}H]nisoxetine displacement assay with homogenised rat brain shows both compounds to have significant affinity, with K_i values of 320.8 nM and 58.2 nM for (2*S*,3*R*)- and (2*R*,3*S*)-iodoreboxetine respectively.

Introduction

The noradrenaline transporter (NAT) is a transmembrane protein located at the pre-synaptic terminal of noradrenergic neurons. The principal physiological function of NAT is to regulate the amount of noradrenaline in the synaptic cleft *via* a re-uptake mechanism.[1,2] A change in the level of noradrenaline in the synapse results in a down-regulation in the level of NAT within the brain.[3] NAT has been implicated in the patho-physiology of numerous neuropsychiatric and neurodegenerative disorders including depression, attention-deficit/hyperactivity disorder, anxiety and Alzheimer's disease.[4–7]

To further probe the link between NAT and these disorders requires the development of a specific radioligand which could be used in association with single photon emission computed tomography (SPECT) or positron emission tomography (PET) for the non-invasive imaging of the receptor. Such a radioligand could be used to image changes in NAT density *in vivo*, resulting in a better understanding of these neuropsychiatric and neurodegenerative disorders. To achieve this goal a number of studies have investigated the radioiodination of compounds known to have high affinity with NAT for *in vivo* SPECT imaging.[8–10] A typical compound used in such studies is reboxetine, the well-known, selective noradrenaline re-uptake inhibitor.[11] Reboxetine is commercially available under the names Edronax, Prolift, Vestra and Norebox for the treatment of depressive disorder and, while the drug is marketed as a racemic mixture of (2*R*,3*R*)- and (2*S*,3*S*)-enantiomers, it is (2*S*,3*S*)-reboxetine **1** that has the best affinity and selectivity for NAT.[12] Using this information, the groups of Saji and Tamagnan have recently designed and synthesised a range of iodinated analogues of **1** for the SPECT imaging of NAT.[8,9] Testing of these compounds showed one compound in particular, iodo-analogue **2** to have excellent affinity (K_i 2.47 nM), good selectivity and significant potential as an imaging agent for NAT.[9]



While much research has been done to develop SPECT imaging agents based on (2*S*,3*S*)- and (2*R*,3*R*)-reboxetine, there have been no reports of investigations using the (2*S*,3*R*)- and (2*R*,3*S*)-stereoisomers. Moreover, little is known about the pharmacology of these stereoisomers of reboxetine. In this paper, we report the first stereoselective synthesis of iodinated analogues of (2*S*,3*R*)- and (2*R*,3*S*)-reboxetine (**3** and **4**), compounds which could be easily radioiodinated and used for SPECT imaging. We also report preliminary biological data showing the high affinity of these compounds for NAT.[13]

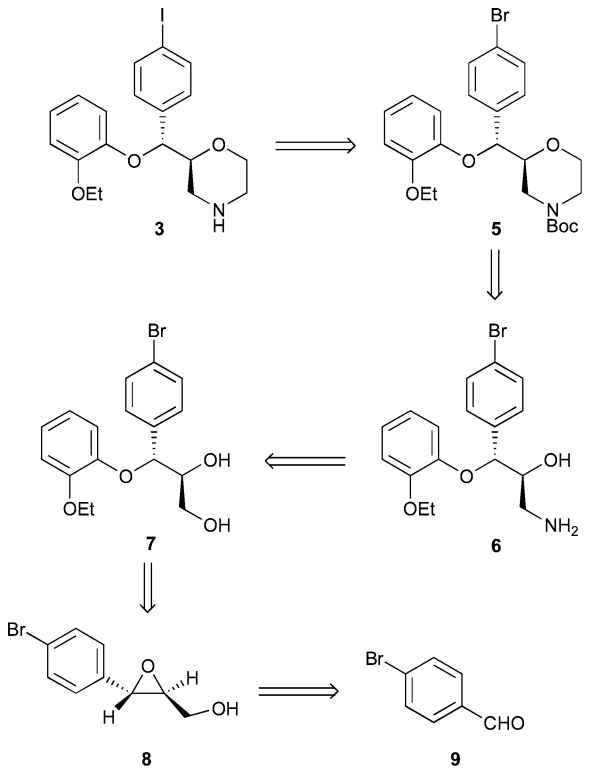
Results and discussion

A number of approaches have been developed for the preparation of single stereoisomers of reboxetine analogues, including resolution of enantiomers, the use of the chiral pool and catalytic asymmetric methods.[14] Our approach to these compounds is shown

[a]*WestChem, Department of Chemistry, The Joseph Black Building, University of Glasgow, Glasgow, UK G12 8QQ. E-mail: andrews@chem.gla.ac.uk; Fax: +44 (0)141 330 4888; Tel: +44 (0)141 330 5936*
[b]*Division of Clinical Neuroscience, Faculty of Medicine, University of Glasgow, UK G12 8QQ*
[c]*West of Scotland Radionuclide Dispensary, University of Glasgow and North Glasgow University Hospital NHS Trust, Glasgow, UK G11 6NT*

† Electronic Supplementary Information (ESI) available: ^{1}H and ^{13}C NMR spectra for compounds **7**, **6**, **12**, **13**, **5**, **14** and **3**. See DOI: 10.1039/b802819b

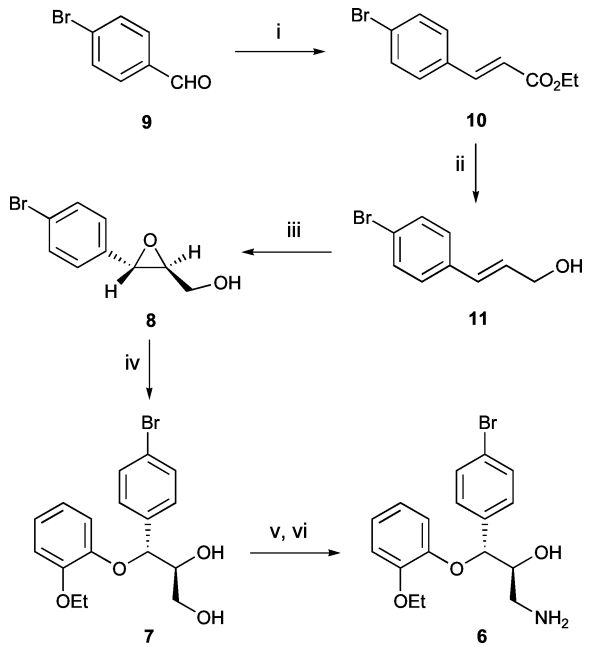
in Scheme 1. It was proposed that the key stereogenic centres of analogue **3** could be created by the synthesis of an *E*-allylic alcohol from 4-bromobenzaldehyde **9** which, following a Sharpless asymmetric epoxidation, would give epoxide **8**. Regioselective ring-opening of **8** with 2-ethoxyphenol would give diol **7** and subsequent selective functionalisation of the primary hydroxyl would give amino alcohol **6**. This would then be used to synthesise the morpholine ring and a halogen exchange reaction would allow incorporation of the key iodine atom to give the first target, (2*S*,3*R*)-iodoreboxetine **3**. We believed that such an approach would also allow the preparation of the (2*R*,3*S*)-stereoisomer **4** by using the opposite enantiomer of diisopropyl tartrate during the Sharpless asymmetric epoxidation step.



Scheme 1

The first stage of the synthesis of **3** involved the preparation of amino alcohol **6** (Scheme 2). 4-Bromobenzaldehyde **9** was subjected to a Horner–Wadsworth–Emmons reaction with triethyl phosphonoacetate under Masamune–Roush conditions, which gave *E*-α,β-unsaturated ester **10** in quantitative yield.[15] Reduction of **10** using DIBAL-H gave *E*-allylic alcohol **11**, again in quantitative yield, and this underwent a Sharpless asymmetric epoxidation with (+)-diisopropyl tartrate to give known epoxide **8** in 79% yield and >98% ee.[16] Introduction of the 2-ethoxyphenol group was achieved by regioselective ring-opening of epoxide **8** with the sodium salt of 2-ethoxyphenol, which gave diol **7** in 67% yield. Selective activation of the primary alcohol with *p*-toluenesulfonyl chloride and triethylamine gave the corresponding tosylate in 82% yield, and this was reacted with an aqueous solution of ammonia which gave amino alcohol **6** in a modest 49% yield.

Amino alcohol **6** was then converted to the desired (2*S*,3*R*)-iodoreboxetine **3** in a six-step sequence (Scheme 3). Acetylation of the amino group with chloroacetyl chloride gave compound **12**

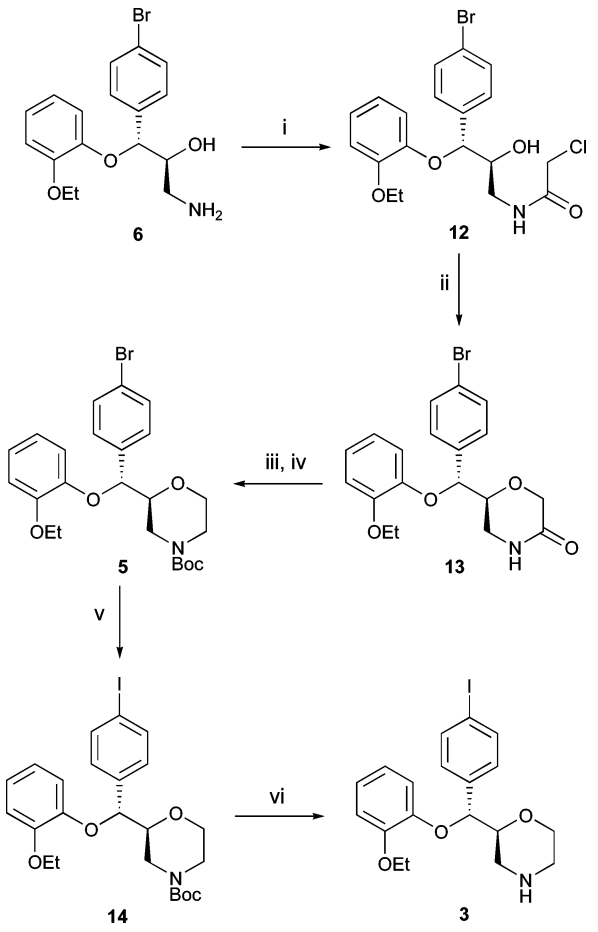


Scheme 2 *Reagents and conditions*: i. triethyl phosphonoacetate, DBU, LiCl, MeCN, 100%; ii. DIBAL-H (2.2 eq.), Et_2O, −78 °C to RT, 100%; iii. (+)-DIPT, $Ti(O^iPr)_4$, *t*-BuOOH, 4Å mol. sieves, CH_2Cl_2, 79%; iv. 2-ethoxyphenol, NaOH (aq.), 70 °C, 67%; v. TsCl, Et_3N, DMAP (cat.), Et_2O, 82%; vi. 25% NH_3 (aq.), MeCN, 49%.

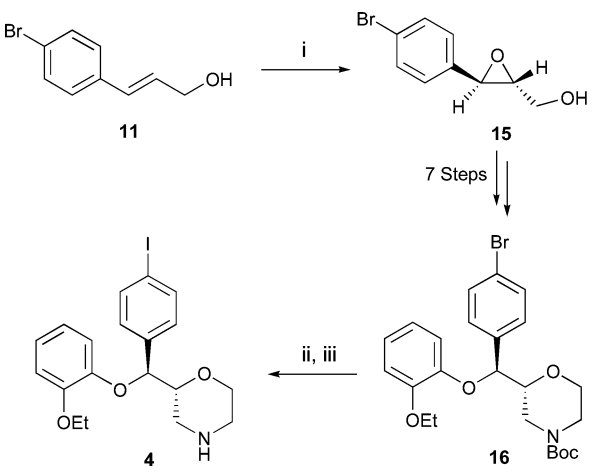
in 83% yield and this was then treated with sodium *tert*-butoxide to give morpholinone **13** in 79% yield.[14a] Reduction of **13** under mild conditions using borane·THF gave the corresponding amine in 73% yield and this was subsequently Boc-protected, which gave **5** in 75% yield. The last stage of the synthesis required the introduction of the key iodine atom and this was achieved using a copper catalysed aromatic halogen exchange reaction, previously described by Klapars and Buchwald.[17] Thus, reaction of bromide **5** with sodium iodide, 1,3-diaminopropane and catalytic amounts of copper iodide gave iodide **14** in 49% yield. Finally, deprotection of the amino group using TFA gave (2*S*,3*R*)-iodoreboxetine **3** in 57% yield.

Having developed a synthetic route to (2*S*,3*R*)-iodoreboxetine **3**, the second target compound, (2*R*,3*S*)-iodoreboxetine **4** was synthesised using the same approach. Thus, allylic alcohol **11** was subjected to a Sharpless asymmetric epoxidation using (−)-diisopropyl tartrate to give (2*R*,3*R*)-[3-(4-bromophenyl)oxiranyl]methanol **15** in 79% yield and this was converted to (2*R*,3*S*)-iodoreboxetine **4** in the same nine-step sequence as described for the (2*S*,3*R*)-stereoisomer **3** (Scheme 4). During the preparation of the second stereoisomer **4**, attempts were made to optimise some of the steps. For example, during the synthesis of **3**, iodination using 1,3-diaminopropane as a ligand for the copper iodide catalysed halogen exchange reaction required a reaction time of 48 hours at 130 °C and only gave the iodide **14** in 49% yield after substantial purification. However, on screening a range of diamine ligands, we found the use of *N*,*N*′-dimethylethylenediamine to be more useful, allowing the reaction to be carried out in 24 hours at 120 °C, giving the corresponding iodide more cleanly in 52% yield.[17,18]

On successful preparation of both stereoisomers, these were tested with homogenised rat brain using a [^{3}H]nisoxetine



Scheme 3 *Reagents and conditions*: i. chloroacetyl chloride, Et_3N, MeCN, 83%; ii. sodium *tert*-butoxide, *t*-BuOH, 79%; iii. BH_3·THF, THF, 73%; iv. Boc_2O, Et_3N, DMAP (cat.), CH_2Cl_2, 75%; v. CuI (cat.), NaI, 1,3-diaminopropane (cat.), 1,4-dioxane, 49%; vi. TFA, CH_2Cl_2, 57%.



Scheme 4 *Reagents and conditions*: i. (−)-DIPT, $Ti(O^iPr)_4$, *t*-BuOOH, 4Å mol. sieves, CH_2Cl_2, 79%; ii. CuI (cat.), NaI, *N*,*N*′-dimethylethylenediamine (cat.), 1,4-dioxane, 52%; iii. TFA, CH_2Cl_2, 57%.

displacement assay. Table 1 shows the K_i values for both compounds along with racemic reboxetine, used as a standard. As can be seen, both compounds **3** and **4** have substantial affinity with NAT, with the (2*R*,3*S*)-stereoisomer **4** the most potent, with a K_i value of 58.2 nM. While the iodinated (2*S*,3*R*)- and (2*R*,3*S*)-compounds do not have the same level of affinity as the parent compound, reboxetine, the (2*R*,3*S*)-iodo compound **4** in particular, still retains similar levels of affinity compared to the racemic mixtures of iodinated (2*S*,3*S*)-and (2*R*,3*R*)-analogues previously prepared.[9] Thus, these results show that, the development of more potent iodo-analogues around a (2*R*,3*S*)-morpholine backbone may yield compounds that can act as effective SPECT imaging agents for NAT.

Table 1 Binding affinity of reboxetine analogues with NAT

Entry	Compound	K_i/nM[a]
1	***rac-1***	6.9 ±1.6
2	**3**	320.8 ±9.0
3	**4**	58.2 ±9.4

[a] K_i values are the mean of 3 separate determinations.

Conclusions

In summary, we have developed a twelve-step synthesis of (2*S*,3*R*)- and (2*R*,3*S*)-iodoreboxetine using a Sharpless asymmetric epoxidation to establish the stereogenic centres, and a copper-mediated aromatic halogen exchange reaction to incorporate the iodine atom. Preliminary testing of these compounds shows the (2*R*,3*S*)-compound **4** to be particularly potent, with similar levels of affinity for the NAT in brain tissue to the more studied (2*S*,3*S*)- and (2*R*,3*R*)-iodoreboxetine analogues. The work described here represents the first generation of compounds from our study. Work is currently underway on the synthesis of further analogues of iodoreboxetine with the aim of discovering compounds with greater affinity, for the eventual development of a SPECT imaging agent for NAT.

Experimental

All reactions were performed under a nitrogen atmosphere unless otherwise noted. Reagents and starting materials were obtained from commercial sources and used as received. THF and diethyl ether were distilled from sodium and benzophenone. Lithium chloride was oven dried (100 °C) for at least 12 h before use. Brine refers to a saturated solution of sodium chloride. Flash column chromatography was carried out using Fisher Matrex silica 60. Macherey-Nagel aluminium backed plates pre-coated with silica gel 60 (UV_{254}) were used for thin layer chromatography and were visualised by staining with $KMnO_4$. ^1H NMR and ^{13}C NMR spectra were recorded on a Bruker DPX 400 spectrometer with chemical shift values in ppm relative to residual chloroform (δ_H 7.28 & δ_C 77.2) as standard. Infrared spectra were recorded using sodium chloride plates on a JASCO FTIR 410 spectrometer and mass spectra were obtained using a JEOL JMS-700 spectrometer. Optical rotations were determined as solutions irradiating with the sodium D line (λ = 589 nm) using an AA series Automatic polarimeter. $[\alpha]_D$ Values are given in units 10^{-1} deg cm^2 g^{-1}.

Ethyl (*E*)-4-bromocinnamate 10[19]

Lithium chloride (2.7 g, 65 mmol) was dissolved in acetonitrile (40 mL). 1,8-Diazobicyclo[5.4.0]undec-7-ene (9.7 mL, 65 mmol) and triethyl phosphonoacetate (12.9 mL, 65 mmol) were added sequentially with stirring. A solution of 4-bromobenzaldehyde (10.0 g, 54 mmol) in acetonitrile (60 mL) was added and the mixture was allowed to stir at room temperature for 18 h. The reaction mixture was concentrated *in vacuo* and the resulting residue dissolved in ethyl acetate (150 mL). The solution was then washed with water (4 × 100 mL) and the organic layer dried ($MgSO_4$). The filtrate was concentrated *in vacuo* to give ethyl (*E*)-4-bromocinnamate **10** as a yellow oil (13.7 g, 100%). δ_H (400 MHz, $CDCl_3$) 1.33 (3H, t, *J* 7.2 Hz, OCH_2CH_3), 4.27 (2H, q, *J* 7.2 Hz, OCH_2CH_3), 6.42 (1H, d, *J* 16.0 Hz, 2-H), 7.36–7.40 (2H, m, 2 × Ar H), 7.49–7.53 (2H, m, 2 × Ar H), 7.61 (1H, d, *J* 16.0 Hz, 3-H); δ_C (100 MHz, $CDCl_3$) 14.3 (CH_3), 60.7 (CH_2), 119.0 (CH), 124.5 (C), 129.5 (2 × CH), 132.2 (2 × CH), 133.4 (C), 143.2 (CH), 166.8 (C); *m*/*z* (EI) 253.9939 (M^+. $C_{11}H_{11}O_2{}^{79}Br$ requires 253.9942), 209 (89%), 183 (23), 181 (21), 102 (100).

(2*E*)-3-(4-Bromophenyl)prop-2-en-1-ol 11[20]

Ethyl (*E*)-4-bromocinnamate **10** (7.0 g, 27.5 mmol) was dissolved in diethyl ether (120 mL) and the solution cooled to −78 °C. DIBAL-H (60.4 mL, 60.4 mmol) was added to the solution dropwise. After 1 h, the reaction mixture was allowed to warm to room temperature and left to stir for 18 h. The yellow solution was cooled to 0 °C, before the reaction was quenched using a saturated solution of ammonium chloride (30 mL). The white solution was filtered through a pad of Celite® using diethyl ether and the filtrate concentrated *in vacuo.* The white solid was recrystallised using ethyl acetate–petroleum ether (40–60 °C) to give (2*E*)-3-(4-bromophenyl)prop-2-en-1-ol **11** as a colourless solid (5.8 g, 100%). Mp 63–65 °C (from ethyl acetate), lit.[20] 63–65 °C; δ_H (400 MHz, $CDCl_3$) 1.48 (1H, t, *J* 6.0 Hz, OH), 4.33 (2H, dt, *J* 6.0, 1.6 Hz, 1-H_2), 6.36 (1H, dt, *J* 16.0, 1.6 Hz, 2-H), 6.56 (1H, d, *J* 16.0 Hz, 3-H), 7.23–7.28 (2H, m, 2 × Ar H), 7.42–7.47 (2H, m, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 63.6 (CH_2), 121.5 (C), 128.0 (2 × CH), 129.3 (CH), 129.8 (CH), 131.7 (2 × CH), 136.0 (C); *m*/*z* (EI) 211.9840 (M^+. $C_9H_9O^{79}Br$ requires 211.9837), 133 (100%), 115 (46), 83 (80).

(2*S*,3*S*)-[3-(4-Bromophenyl)oxiranyl]methanol 8[16]

Dichloromethane (150 mL) and activated 4 Å molecular sieves (3.0 g) were added to a round-bottomed flask and the solvent cooled to −20 °C using an acetone–ice bath. (+)-Diisopropyl tartrate (0.18 mL, 0.8 mmol) and titanium isopropoxide (0.2 mL, 0.7 mmol) were added sequentially with stirring. Anhydrous *tert*-butyl hydroperoxide solution (3.2 mL, 35 mmol, 5.5 M in hexanes) was added dropwise and the mixture was stirred at −20 °C for 0.5 h. A solution of (2*E*)-3-(4-bromophenyl)prop-2-en-1-ol **11** (3.0 g, 14 mmol) dissolved in dichloromethane (10 mL) was added to the mixture dropwise over a period of 0.25 h. The mixture was left to stir at −20 °C for 2.5 h. The flask was allowed to warm to 0 °C and the reaction quenched using distilled water (15 mL). The mixture was left to stir for 0.5 h while allowing it to warm to room temperature. A solution of 30% sodium hydroxide saturated with sodium chloride (75 mL) was added and the mixture stirred for 0.3 h. Dichloromethane (100 mL) was added to the mixture and left to stir for a further 0.25 h. The mixture was transferred to a separating funnel and the water layer removed. The organic layer was dried ($MgSO_4$) and filtered through a pad of Celite® which was then washed with diethyl ether. The yellow solution was concentrated *in vacuo.* Purification was carried out by flash column chromatography and elution with 60 : 40 ethyl acetate–petroleum ether (40–60 °C) gave (2*S*,3*S*)-[3-(4-bromophenyl)oxiranyl]methanol **8** as a colourless solid (2.55 g, 79%). Mp 61–62 °C (from petroleum ether–ethyl acetate) lit.[16] 67–68 °C (from hexane–ethyl acetate); $[a]_D^{22}$ −38.6 (*c* 1.0, $CHCl_3$), lit.[16] $[a]_D^{25}$ −35.2 (*c* 1.0, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.74 (1H, dd, *J* 7.8, 5.0 Hz, OH), 3.17 (1H, dt, *J* 3.6, 2.2 Hz, 2-H), 3.81 (1H, ddd, *J* 12.8, 7.8, 3.6 Hz, 1-*H*H), 3.91 (1H, d, *J* 2.2 Hz, 3-H), 4.05 (1H, ddd, *J* 12.8, 5.0, 2.2 Hz, 1-H*H*), 7.14–7.18 (2H, m, 2 × Ar H), 7.46–7.50 (2H, m, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 54.9 (CH), 61.0 (CH_2), 62.4 (CH), 122.0 (C), 127.4 (2 × CH), 131.7 (2 × CH), 134.9 (C); *m*/*z* (EI) 227.9787 (M^+. $C_9H_9O_2{}^{79}Br$ requires 227.9786), 212 (10%), 185 (34), 89 (75), 83 (100).

(2*R*,3*R*)-[3-(4-Bromophenyl)oxiranyl]methanol 15[16]

The reaction was carried out as described above except using (−)-diisopropyl tartrate. On a 20 mmol scale this gave (2*R*,3*R*)-[3-(4-bromophenyl)oxiranyl]methanol **15** (3.56 g, 79%) as a colourless solid. Mp 61–62 °C (from petroleum ether–ethyl acetate) lit.[16] 67–68 °C (from hexane–ethyl acetate); $[a]_D^{22}$ +31.7 (*c* 1.0, $CHCl_3$); spectroscopic data as described for **8**.

(2*S*,3*R*)-3-(4-Bromophenyl)-3-(2-ethoxyphenoxy)propane-1,2-diol 7

2-Ethoxyphenol (1.10 g, 7.9 mmol) was added to an aqueous sodium hydroxide solution (0.30 g, 6.6 mmol in 70 mL water) and the mixture was heated to 70 °C until the solid had dissolved. After 1 h of stirring, (2*S*,3*S*)-[3-(4-bromophenyl)oxiranyl]methanol **8** (1.50 g, 6.6 mmol) was added and the mixture left to stir for 4 h. The flask was then allowed to cool to room temperature and acidified to pH 2–3 using 2 M hydrochloric acid. The bulk solvent was decanted and the remaining solid suspended in water, and then extracted using dichloromethane (2 × 100 mL) followed by ethyl acetate (3 × 100 mL). The organic layers were combined, dried ($MgSO_4$) and the filtrate concentrated *in vacuo.* The solid was recrystallised from diethyl ether to give (2*S*,3*R*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propane-1,2-diol **7** as a colourless solid (1.50 g, 67%). Mp 77–78 °C (from diethyl ether); ν_{max}/cm^{-1} (NaCl) 3345 (OH), 2943 (CH), 1500 (C=C), 1246, 732; $[a]_D^{22}$ −57.0 (*c* 1.0, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.51 (3H, t, *J* 7.2 Hz, OCH_2CH_3), 3.13 (1H, dd, *J* 10.0, 3.4 Hz, 1-OH), 3.26 (1H, d, *J* 8.0 Hz, 2-OH), 3.65 (1H, ddd, *J* 11.8, 10.0, 4.3, Hz, 1-*H*H), 3.84–3.89 (1H, m, 2-H), 3.95 (1H, dt, *J* 11.8, 3.4 Hz, 1-H*H*), 4.11 (2H, q, *J* 7.2 Hz, OCH_2CH_3), 5.22 (1H, d, *J* 4.3 Hz, 3-H), 6.59 (1H, dd, *J* 8.0, 1.6 Hz, 1 × Ar H), 6.71–6.75 (1H, m, 1 × Ar H), 6.87–6.93 (2H, m, 2 × Ar H), 7.27–7.31 (2H, m, 2 × Ar H), 7.49–7.52 (2H, m, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 14.8 (CH_3), 62.1 (CH_2), 64.2 (CH_2), 74.2 (CH), 85.6 (CH), 112.5 (CH), 116.4 (CH), 120.8 (CH), 122.2 (C), 122.8 (CH), 128.2 (2 × CH), 131.9 (2 × CH), 137.1 (C), 146.8 (C), 149.2 (C); *m*/*z* (EI) 366.0465 (M^+.

$C_{17}H_{19}O_4{}^{79}Br$ requires 366.0467), 169 (11%), 138 (100), 110 (50), 83 (58), 47 (12).

(2*R*,3*S*)-3-(4-Bromophenyl)-3-(2-ethoxyphenoxy)propane-1,2-diol

The reaction was carried out as described above, using (2*R*,3*R*)-[3-(4-bromophenyl)oxiranyl]methanol **15** (3.38 g, 14.8 mmol). This gave (2*R*,3*S*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propane-1,2-diol as a colourless solid (4.31 g, 79%). Mp 76–78 °C (from diethyl ether); $[\alpha]_D^{25}$ +64.6 (*c* 1.0, $CHCl_3$); spectroscopic data as described for **7**.

(2*S*,3*R*)-1-Amino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol 6

(2*S*,3*R*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propane-1,2-diol **7** (0.55 g, 1.49 mmol) was dissolved in dichloromethane (50 mL) and triethylamine (0.31 mL, 2.24 mmol), 4-dimethylaminopyridine (0.004 g, 0.04 mmol) and *p*-toluenesulfonyl chloride (0.34 g, 1.79 mmol) were added sequentially with stirring. The reaction mixture was stirred for 3 h before being diluted with diethyl ether (20 mL) and washed with 2 M hydrochloric acid (30 mL). The organic layer was dried ($MgSO_4$) and the filtrate concentrated *in vacuo* to give (2*S*,3*R*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)-1-(toluenesulfonyloxy)propan-2-ol as a colourless oil (0.64 g, 82%). (2*S*,3*R*)-3-(4-Bromophenyl)-3-(2-ethoxyphenoxy)-1-(toluenesulfonyloxy)propan-2-ol (0.64 g, 1.23 mmol) was dissolved in acetonitrile (40 mL) and 25% aqueous ammonia solution (50 mL) was added. The reaction mixture was left to stir for 96 h in a sealed round-bottomed flask. The acetonitrile was removed *in vacuo* and the aqueous solution diluted with distilled water (100 mL). The aqueous solution was then extracted with ethyl acetate (3 × 100 mL), dried ($MgSO_4$) and the filtrate concentrated *in vacuo*. Purification was carried out by flash chromatography and elution with 90 : 10 ethyl acetate–methanol gave (2*S*,3*R*)-1-amino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol **6** as a colourless solid (0.22 g, 49%). Mp 60–62 °C (from diethyl ether); ν_{max}/cm^{-1} (neat) 3338, 3303, 2901 (CH), 2115, 1593 (C=C); $[\alpha]_D^{22}$ −68.8 (*c* 1.0, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.47 (3H, t, *J* 7.0 Hz, OCH_2CH_3), 2.92 (1H, dd, *J* 12.8, 4.6 Hz, 1-*H*H), 3.03 (1H, dd, *J* 12.8, 5.6 Hz, 1-H*H*), 3.99 (1H, m, 2-H), 4.06 (2H, q, *J* 7.0 Hz, OCH_2CH_3), 5.11 (1H, d, *J* 4.6 Hz, 3-H), 6.63 (1H, dd, *J* 8.0, 1.4 Hz, 1 × Ar H), 6.72 (1H, dt, *J* 8.0, 1.4 Hz, 1 × Ar H), 6.85–6.93 (2H, m, 2 × Ar H), 7.27 (2H, d, *J* 8.2 Hz, 2 × Ar H), 7.47 (2H, d, *J* 8.2 Hz, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 14.7 (CH_3), 42.3 (CH_2), 64.3 (CH_2), 74.4 (CH), 84.5 (CH), 113.2 (CH), 117.7 (CH), 120.9 (CH), 121.9 (C), 122.8 (CH), 128.7 (2 × CH), 131.5 (2 × CH), 137.8 (C), 147.2 (C), 149.7 (C); *m*/*z* (CI) 366.0699 (MH^+ $C_{17}H_{21}O_3N^{79}Br$ requires 366.0705), 365 (3%), 138 (100), 171 (12), 110 (77), 60 (14).

(2*R*,3*S*)-1-Amino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol

The tosylation reaction was carried out as described above, using (2*R*,3*S*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propane-1,2-diol (3.26 g, 8.88 mmol). This gave (2*R*,3*S*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)-1-(toluenesulfonyloxy)propan-2-ol as a colourless oil (2.42 g, 44%). The aminolysis reaction was carried out as described above using (2*R*,3*S*)-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)-1-(toluenesulfonyloxy)propan-2-ol (2.42 g, 4.6 mmol). This gave (2*R*,3*S*)-1-amino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol as a colourless oil (0.97 g, 57%). $[\alpha]_D^{22}$ +71.5 (*c* 1.1, $CHCl_3$); spectroscopic data as described for **6**.

(2*S*,3*R*)-1-Chloroacetylamino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol 12

(2*S*,3*R*)-1-Amino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol **6** (0.15 g, 0.42 mmol) was dissolved in acetonitrile (6.5 mL) and the mixture cooled to −10 °C using an acetone–ice bath. Triethylamine (0.07 mL, 0.50 mmol) and chloroacetyl chloride (0.04 mL, 0.46 mmol) were added sequentially and the solution was allowed to stir at −10 °C for 1 h. The reaction mixture was then warmed to room temperature and left to stir for 18 h. The reaction mixture was concentrated *in vacuo*. Purification was carried out by flash column chromatography and elution with 50 : 50 ethyl acetate–petroleum ether (40–60 °C) gave (2*S*,3*R*)-1-chloroacetylamino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol **12** as a colourless oil (0.15 g, 83%). ν_{max}/cm^{-1} (NaCl) 3423, 3020 (CH), 1660 (CO), 1215, 770; $[\alpha]_D^{22}$ −105.4 (*c* 1.0, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.51 (3H, t, *J* 7.1 Hz, OCH_2CH_3), 3.35 (1H, ddd, *J* 14.0, 7.4, 4.2 Hz, 1-*H*H), 3.73 (1H, ddd, *J* 14.0, 7.2, 3.6 Hz, 1-H*H*), 3.98–4.00 (1H, m, 2-H), 4.03 (2H, s, CH_2Cl), 4.12 (2H, q, *J* 7.1 Hz, OCH_2CH_3), 4.97 (1H, d, *J* 5.2 Hz, 3-H), 6.73–7.00 (4H, m, 4 × Ar H), 7.18 (1H, br s, NH), 7.33 (2H, d, *J* 8.4 Hz, 2 × Ar H), 7.52 (2H, d, *J* 8.4 Hz, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 14.9 (CH_3), 41.9 (CH_2), 42.6 (CH_2), 64.5 (CH_2), 73.1 (CH), 85.2 (CH), 113.4 (CH), 119.5 (CH), 121.2 (CH), 122.3 (C), 123.8 (CH), 128.8 (2 × CH), 131.8 (2 × CH) 137.0 (C), 147.2 (C), 149.9 (C), 166.9 (C); *m*/*z* (CI) 444.0399 (MH^+. $C_{19}H_{22}O_4N^{35}Cl^{81}Br$ requires 444.0400), 306 (100%), 304 (74), 226 (62), 192 (19), 139 (80).

(2*R*,3*S*)-1-Chloroacetylamino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol

The reaction was carried out as described above, using (2*R*,3*S*)-1-amino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol (1.50 g, 4.10 mmol). This gave (2*R*,3*S*)-1-chloroacetylamino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol as a colourless oil (1.11 g, 62%). $[\alpha]_D^{22}$ +114.6 (*c* 1.1, $CHCl_3$); spectroscopic data as described for **12**.

(2*S*,3*R*)-2-[(4-Bromobenzyl)-(2-ethoxyphenoxy)methyl]morpholin-5-one 13

A solution of (2*S*,3*R*)-1-chloroacetylamino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol **12** (0.35 g, 0.79 mmol) in *tert*-butanol (4 mL) was added dropwise to a solution of potassium *tert*-butoxide (0.19 g, 2.0 mmol) in *tert*-butanol (1.0 mL) at 40 °C. The reaction mixture was allowed to stir for 3 h at this temperature. The solution was acidified to pH 2–3 by the addition of 2 M hydrochloric and then concentrated *in vacuo*. The residue was suspended in water (50 mL) and the aqueous solution extracted with ethyl acetate (3 × 50 mL), dried ($MgSO_4$), and the filtrate concentrated *in vacuo*. Purification was carried out by dry flash chromatography and elution with 100% ethyl acetate gave (2*S*,3*R*)-2-[(4-bromobenzyl)-(2-ethoxyphenoxy)methyl]morpholin-5-one **13** as a

colourless oil (0.25 g, 79%). ν_{max}/cm^{-1} (NaCl) 3410 (NH), 3019 (CH), 2400, 1679 (CO), 1500, 1216; $[\alpha]_D^{25}$ −79.5 (c 0.6, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.46 (3H, t, *J* 7.0 Hz, OCH_2CH_3), 3.66–3.80 (2H, m, 3-H_2), 3.99–4.02 (1H, m, 2-H), 4.04–4.29 (4H, m, 6-H_2, OCH_2CH_3), 5.04 (1H, d, *J* 7.6 Hz, 2-CH), 6.27 (1H, br s, NH), 6.64–6.94 (4H, m, 4 × Ar H), 7.29–7.32 (2H, m, 2 × Ar H), 7.46–7.51 (2H, m, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 15.1 (CH_3), 43.9 (CH_2), 64.3 (CH_2), 67.8 (CH_2), 76.0 (CH), 81.5 (CH), 113.6 (CH), 118.4 (CH), 120.7 (CH), 122.4 (C), 123.2 (CH), 129.0 (2 × CH), 131.6 (2 × CH), 137.3 (C), 146.5 (C), 149.9 (C), 168.5 (C); *m/z* (EI) 407.0562 (M^+. $C_{19}H_{20}O_4N^{81}Br$ requires 407.0558), 269 (73%), 267 (73), 171 (86), 169 (87), 138 (100).

(2*R*,3*S*)-2-[(4-Bromobenzyl)-(2-ethoxyphenoxy)methyl]morpholin-5-one

The reaction was carried out as described above, using (2*R*,3*S*)-1-chloroacetylamino-3-(4-bromophenyl)-3-(2-ethoxyphenoxy)propan-2-ol (1.07 g, 2.42 mmol). This gave (2*R*,3*S*)-2-[(4-bromobenzyl)-(2-ethoxyphenoxy)methyl]morpholin-5-one as a colourless oil (0.89 g, 90%). $[\alpha]_D^{22}$ +80.0 (*c* 0.9, $CHCl_3$); spectroscopic data as described for **13**.

(2*S*,3*R*)-2-[(4-Bromophenyl)-(2-ethoxyphenoxy)-methyl]morpholine 17

(2*S*,3*R*)-2-[(4-Bromobenzyl)-(2-ethoxyphenoxy)methyl]morpholin-5-one **13** (0.20 g, 0.5 mmol) was dissolved in THF (0.5 mL) and the solution cooled to 0 °C. Borane·THF complex (1.10 mL, 1.1 mmol) was added to the solution dropwise with stirring. After 0.5 h, the solution was heated under reflux for 4 h. After cooling in an ice bath, the excess borane was destroyed by the addition of distilled water (1 mL) and the solution was concentrated *in vacuo*. The residue was dissolved in 6 M hydrochloric acid (5 mL) and allowed to stir for 0.25 h. The solution was then concentrated *in vacuo* and the residue dissolved in 2 M sodium hydroxide (5 mL). The aqueous solution was extracted using chloroform (3 × 5 mL) and the organic layers combined, dried ($MgSO_4$), and the filtrate concentrated *in vacuo* to give (2*S*,3*R*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]morpholine **17** as a colourless oil (0.12 g, 73%). ν_{max}/cm^{-1} (NaCl) 3413 (NH), 2925 (CH), 2345, 1638, 1593; $[\alpha]_D^{25}$ −67.5 (*c* 0.2, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.45 (3H, t, *J* 7.0 Hz, OCH_2CH_3), 2.81–2.94 (3H, m, 5-H_2, 3-*H*H), 3.37 (1H, br d, *J* 12.4 Hz, 3-H*H*), 3.53 (1H, td, *J* 11.2, 2.8 Hz, 6-*H*H) 3.75–3.80 (1H, m, 2-H), 3.86 (1H, br d, *J* 11.2 Hz, 6-H*H*), 4.05 (2H, q, *J* 7.0 Hz, OCH_2CH_3), 4.98 (1H, d, *J* 6.8 Hz, 2-CH), 6.64–6.89 (4H, m, 4 × Ar H), 7.29 (2H, d, *J* 8.4 Hz, 2 × Ar H), 7.45 (2H, d, *J* 8.4 Hz, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 15.1 (CH_3), 44.6 (CH_2), 46.4 (CH_2), 64.4 (CH_2), 66.5 (CH_2), 78.1 (CH), 82.2 (CH), 113.7 (CH), 118.1 (CH), 120.7 (CH), 122.1 (C), 122.8 (CH), 129.1 (2 × CH), 131.5 (2 × CH), 137.5 (C), 146.9 (C), 149.9 (C); *m/z* (EI) 393.0756 (M^+. $C_{19}H_{22}O_3N^{81}Br$ requires 393.0765), 255 (86%), 253 (86), 169 (27), 86 (99), 84 (100).

(2*R*,3*S*)-2-[(4-Bromophenyl)-(2-ethoxyphenoxy)-methyl]morpholine

The reaction was carried out as described above, using (2*R*,3*S*)-2-[(4-bromobenzyl)-(2-ethoxyphenoxy)methyl]morpholin-5-one (0.85 g, 2.1 mmol). This gave (2*R*,3*S*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]morpholine as a colourless oil (0.09 g, 11%). $[\alpha]_D^{22}$ +46.8 (*c* 1.0, $CHCl_3$); spectroscopic data as described for **17**.

(2*S*,3*R*)-2-[(4-Bromophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine 5

(2*S*,3*R*)-2-[(4-Bromophenyl)-(2-ethoxyphenoxy)methyl]morpholine (0.14 g, 0.4 mmol) was dissolved in dichloromethane (3 mL). Triethylamine (0.055 mL, 0.4 mmol), dimethylaminopyridine (0.01 g, 0.07 mmol) and di-*tert*-butyl dicarbonate (0.09 g, 0.4 mmol) were added sequentially with stirring and the reaction mixture allowed to stir at room temperature for 18 h. The reaction mixture was concentrated *in vacuo*. Purification was carried out using flash column chromatography and elution with 20 : 80 ethyl acetate–petroleum ether (40–60 °C) gave (2*S*,3*R*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine **5** as a colourless oil (0.13 g, 75%). ν_{max}/cm^{-1} (NaCl) 2925 (CH), 1695 (CO), 1500 (C=C), 1259, 1106; $[\alpha]_D^{24}$ −34.8 (*c* 0.5, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.44–1.47 (12H, m, OCH_2CH_3, $C(CH_3)_3$), 2.94–3.00 (2H, m, 3-*H*H, 5-*H*H), 3.46 (1H, td, *J* 11.6, 2.8 Hz, 5-H*H*), 3.71–3.74 (1H, m, 2-H), 3.86 (2H, dd, *J* 11.6, 2.8 Hz, 6-H_2), 3.97–4.06 (2H, m, OCH_2CH_3), 4.34 (1H, d, *J* 13.2 Hz, 3-H*H*), 5.02 (1H, br m, 2-CH), 6.70–6.91 (4H, m, 4 × Ar H), 7.30 (2H, d, *J* 8.4 Hz, 2 × Ar H), 7.45 (2H, d, *J* 8.4 Hz, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 14.0 (CH_3), 27.4 (3 × CH_3), 42.2 (CH_2), 46.1 (CH_2), 63.4 (CH_2), 65.7 (CH_2), 78.1 (CH), 79.0 (C), 81.7 (CH), 113.9 (CH), 117.2 (CH), 120.7 (CH), 121.0 (C), 121.6 (CH), 129.2 (2 × CH), 130.3 (2 × CH), 136.6 (C), 146.1 (C), 148.9 (C), 153.8 (C); *m/z* (EI) 491.1302 (M^+. $C_{24}H_{30}O_5N^{79}Br$ requires 491.1307), 298 (90%), 254 (99), 138.0 (89), 57 (100).

(2*R*,3*S*)-2-[(4-Bromophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine 16

The reaction was carried out as described above, using (2*R*,3*S*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]morpholine (0.06 g, 0.18 mmol). This gave (2*R*,3*S*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine **16** as a colourless oil (0.04 g, 53%). $[\alpha]_D^{26}$ +38.8 (*c* 1.1, $CHCl_3$); spectroscopic data as described for **5**.

(2*S*,3*R*)-2-[(4-Iodophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine 14

A Schlenk tube was charged with copper iodide (0.001 g, 0.005 mmol) and sodium iodide (0.01 g, 0.14 mmol), and the tube evacuated and back filled with argon. 1,3-Diaminopropane (0.0005 g, 0.007 mmol) and (2*S*,3*R*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine **5** (0.03 g, 0.07 mmol) dissolved in 1,4-dioxane (1 mL) were added under argon. The Schlenk tube was sealed using a Teflon valve and the reaction mixture stirred at 130 °C for 48 h. The resulting suspension was cooled to room temperature and diluted with 25% aqueous ammonia solution (2 mL) and poured onto water (20 mL). The aqueous solution was washed with dichloromethane (3 × 15 mL) and the organic layers combined, dried ($MgSO_4$) and

the filtrate concentrated *in vacuo*. Purification was carried out using column chromatography and elution with 20 : 80 ethyl acetate–petroleum ether (40–60 °C) gave (2*S*,3*R*)-2-[(4-iodophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine **14** as a colourless oil (0.02 g, 49%). ν_{max}/cm^{-1} (NaCl) 2976 (CH), 2357, 1695 (CO), 1502 (C=C), 1254; $[\alpha]_D^{25}$ −37.1 (*c* 2.5, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.44–1.47 (12H, m, OCH_2CH_3, $(CH_3)_3$), 2.90–3.01 (2H, m, 3-*H*H, 5-*H*H), 3.46 (1H, td, *J* 11.6, 2.4 Hz, 5-H*H*), 3.70–3.74 (1H, m, 2-H), 3.86 (2H, dd, *J* 11.6, 2.4 Hz, 6-H_2), 4.05–4.11 (2H, m, OCH_2CH_3), 4.42 (1H, br d, *J* 13.2 Hz, 3-H*H*), 5.00 (1H, br m, 2-CH), 6.70–6.88 (4H, m, 4 × Ar H), 7.16 (2H, d, *J* 8.2 Hz, 2 × Ar H), 7.65 (2H, d, *J* 8.2 Hz, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 14.0 (CH_3), 27.4 (3 × CH_3), 42.3 (CH_2), 44.1 (CH_2), 63.4 (CH_2), 65.8 (CH_2), 77.2 (C), 78.1 (CH), 80.7 (CH), 112.9 (CH), 117.1 (CH), 119.7 (CH), 121.6 (CH), 128.4 (2 × CH), 130.4 (C), 136.3 (2 × Ar C), 137.3 (C), 146.1 (C), 148.9 (C), 153.8 (C); *m*/*z* (EI) 539.1166 (M^+. $C_{24}H_{30}O_5NI$ requires 539.1169), 346 (93%), 302 (94), 217 (44), 138 (73), 57 (100).

(2*R*,3*S*)-2-[(4-Iodophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine

A Schlenk tube was charged with copper iodide (0.01 g, 0.05 mmol) and sodium iodide (0.027 g, 0.18 mmol), and the tube evacuated and back filled with argon. *N*,*N*′-Dimethylethylenediamine (0.01 mL, 0.10 mmol) and a solution of (2*R*,3*S*)-2-[(4-bromophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine **16** in butan-1-ol (0.04 g, 0.09 mmol in 3 mL) were added under argon. The Schlenk tube was sealed using a Teflon valve and the reaction mixture stirred at 120 °C for 24 h. The resulting suspension was concentrated *in vacuo* and the crude material dissolved in diethyl ether (20 mL), and washed with ammonia solution (1 mL of 30% $NH_{3\,(aq)}$ in 20 mL water) followed by water (2 × 20 mL). The organic fractions were combined, dried ($MgSO_4$) and the filtrate concentrated *in vacuo* to give (2*R*,3*S*)-2-[(4-iodophenyl)-(2-ethoxyphenoxy)methyl]-*N*-tert-butoxycarbonylmorpholine as a colourless oil (0.03 g, 52%). $[\alpha]_D^{22}$ +43.2 (*c* 2.2, $CHCl_3$); spectroscopic data as described for **14**.

(2*S*,3*R*)-2-[(4-Iodophenyl)-(2-ethoxyphenoxy)methyl]morpholine 3

Trifluoroacetic acid (1 mL, 0.06 mmol) was added to a solution of (2*S*,3*R*)-2-[(4-iodophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine **14** (0.02 g, 0.04 mmol) in dichloromethane (5 mL). The reaction mixture was allowed to stir at room temperature for 4 h. The reaction mixture was concentrated *in vacuo* and the crude material redissolved in dichloromethane (10 mL) and washed with a saturated sodium hydrogen carbonate solution (10 mL). The organic layer was separated, dried ($MgSO_4$) and the filtrate concentrated *in vacuo* to give (2*S*,3*R*)-2-[(4-iodophenyl)-(2-ethoxyphenoxy)methyl]morpholine **3** as a colourless oil (0.01 g, 57%). ν_{max}/cm^{-1} (NaCl) 3421 (NH), 2084 (CH), 1639 (C=C), 1500, 1255; $[\alpha]_D^{23}$ −38.0 (*c* 0.9, $CHCl_3$); δ_H (400 MHz, $CDCl_3$) 1.45 (3H, t, *J* 7.0 Hz, OCH_2CH_3), 1.93 (1H, br s, NH), 2.78–2.94 (3H, m, 3-*H*H, 5-H_2,), 3.37 (1H, br d, *J* 11.6 Hz, 3-H*H*), 3.53 (1H, br t, *J* 12.4 Hz, 6-*H*H), 3.77 (1H, m, 2-H), 3.87 (1H, br d, *J* 12.4 Hz, 6-H*H*), 4.06 (2H, qd, *J* 7.0, 2.0 Hz, OCH_2CH_3), 4.97 (1H, d, *J* 7.0 Hz, 2-CH), 6.64–6.73 (2H, m, 2 × Ar H), 6.84–6.89 (2H, m, 2 × Ar H), 7.16 (2H, d, *J* 8.4 Hz, 2 × Ar H), 7.65 (2H, d, *J* 8.4 Hz, 2 × Ar H); δ_C (100 MHz, $CDCl_3$) 14.0 (CH_3), 45.0 (CH_2), 47.9 (CH_2), 64.5 (CH_2), 67.8 (CH_2), 77.3 (CH), 82.3 (CH), 113.7 (CH), 117.6 (CH), 120.8 (CH), 122.4 (CH), 129.3 (2 × CH), 130.3 (C), 136.3 (2 × CH), 137.8 (C), 146.3 (C), 148.7 (C); *m*/*z* (EI) 439.0642 (M^+. $C_{19}H_{22}O_3NI$ requires 439.0644), 301 (99%), 220 (96), 138 (61), 85 (77), 56 (100).

(2*R*,3*S*)-2-[(4-Iodophenyl)-(2-ethoxyphenoxy)methyl]morpholine 4

The reaction was carried out as described above, using (2*R*,3*S*)-2-[(4-iodophenyl)-(2-ethoxyphenoxy)methyl]-*N*-*tert*-butoxycarbonylmorpholine (0.02 g, 0.04 mmol). This gave (2*R*,3*S*)-2-[(4-iodophenyl)-(2-ethoxyphenoxy)methyl]morpholine 4 as a colourless oil (0.01 g, 57%). $[\alpha]_D^{22}$ +38.3 (*c* 0.9, $CHCl_3$); spectroscopic data as described for **3**.

[^{3}H]Nisoxetine competition binding assays

Whole brains, excluding the cerebellum, were obtained from male adult Sprague-Dawley rats and homogenised using a Polytron in ice-cold 50 mM Tris-HCl, pH 7.4, containing 300 mM NaCl and 5 mM KCl (1 : 10 volumes). Homogenates were centrifuged at 25 400 g for 0.25 h at 4 °C and the resulting pellet was washed (3×) by resuspension and centrifugation then stored at −50 °C until use. For determination of K_i values, aliquots of membrane suspensions (750–850 μg of protein) were incubated for 4 h at 4 °C in 50 mM Tris-HCl, pH 7.4, containing 300 mM NaCl and 5 mM KCl with 1.2 nM [^{3}H]nisoxetine (71 Ci/mmol, GE Healthcare) in the presence or absence of 14–16 concentrations of the competitor (range 1 pM–200 μM). The total incubation volume was 0.5 mL and non-specific binding was determined in the presence of 10 μM reboxetine. The reaction was terminated by rapid filtration through Whatman GF/B glass fibre filters presoaked in 0.5% polyethylenimine using a Brandel cell harvester. Filters were washed three times rapidly in ice-cold Tris buffer and the radioactivity determined by liquid scintillation counting. For each compound, 3 independent competition curves were performed with triplicate samples. K_i Values were derived from nonlinear regression analysis using GraphPad Prism Version 4 (GraphPad Software Inc.) and a K_d value for [^{3}H]nisoxetine (1.7 nM) determined under the same assay conditions.

Acknowledgements

Financial support from the EPSRC (DTA award to NKJ), BBSRC (Industrial Studentship with GSK to ARC) and Medical Research Scotland is gratefully acknowledged.

Notes and references

1 S. G. Amara and M. J. Kuhar, *Annu. Rev. Neurosci.*, 1993, **16**, 73.
2 R. D. Blakely and A. L. Bauman, *Curr. Opin. Neurobiol.*, 2000, **10**, 328.
3 M.-Y. Zhu and G. A. Ordway, *J. Neurochem.*, 1997, **68**, 134.
4 M. K. Spillmann, A. J. W. Van der Does, M. A. Rankin, R. D. Vuolo, J. E. Alpert, A. A. Nierenberg, J. F. Rosenbaum, D. Hayden, D. Schoenfeld and M. Fava, *Psychopharmacology*, 2001, **155**, 123.
5 J. Biederman and T. Spencer, *Biol. Psychiatry*, 1999, **46**, 1234.
6 S. Hisamichi, A. Y. Moon, A. Aoyagi, T. Hara, K. Abe, R. Yamazaki, Y. Kumagae, S. Naruto, K. Koyama, S. Marumoto, K. Tago, N. Toda, K. Takami, N. Yamada, M. Ori, H. Kogen and T. Kaneko, *J. Pharmacol. Sci. (Tokyo, Jpn)*, 2003, **93**, 95.

7 S. M. Tejani-Butt, J. X. Yang and H. Zaffar, *Brain Res.*, 1993, **631**, 147.
8 N. Kanegawa, Y. Kiyono, H. Kimura, T. Sugita, S. Kajiyama, H. Kawashima, M. Ueda, Y. Kuge and H. Saji, *Eur. J. Nucl. Med. Mol. Imaging*, 2006, **33**, 639.
9 G. D. Tamagnan, E. Brenner, D. Alagille, J. K. Staley, C. Haile, A. Koren, M. Early, R. M. Baldwin, F. I. Tarazi, R. J. Baldessarini, N. Jarkas, M. M. Goodman and J. P. Seibyl, *Bioorg. Med. Chem. Lett.*, 2007, **17**, 533.
10 B. Lakshmi, M.-P. Kung, B. Lieberman, J. Zhao, R. Waterhouse and H. F. Kung, *Nucl. Med. Biol.*, 2008, **35**, 43.
11 M. Hajos, J. C. Fleishaker, J. K. Filipiak-Reisner, M. T. Brown and E. H. F. Wong, *CNS Drug Rev.*, 2004, **10**, 23.
12 E. H. F. Wong, S. Ahmed, R. C. Marshall, R. McArthur, D. P. Taylor, L. Birgerson and P. Cetera, Patent WO 2001 001 973, 2001.
13 Part of this work has been previously reported as conference proceedings of the Ninth International Symposium on the Synthesis and Applications of Isotopically Labelled Compounds, Edinburgh, 2006: R. Bischoff, D. J. Hamilton, N. K. Jobson and A. Sutherland, *J. Labelled Compd. Radiopharm.*, 2007, **50**, 323.
14 (*a*) P. Melloni, A. Della Torre, E. Lazzari, G. Mazzini and M. Meroni, *Tetrahedron*, 1985, **41**, 1393; (*b*) E. Brenner, R. M. Baldwin and G. Tamagnan, *Org. Lett.*, 2005, **7**, 937; (*c*) W. W. Harding, M. Hodge, Z. Wang, W. L. Woolverton, D. Parrish, J. R. Deschamps and T. E. Prisinzano, *Tetrahedron: Asymmetry*, 2005, **16**, 2249; (*d*) S. A. Siddiqui, U. C. Narkhede, R. J. Lahoti and K. V. Srinivasan, *Synlett*, 2006, 1771; (*e*) T.-X. Métro, D. Gomez Pardo and J. Cossy, *J. Org. Chem.*, 2008, **73**, 707.
15 M. A. Blanchette, W. Choy, J. T. Davis, A. P. Essenfeld, S. Masumune, W. R. Roush and T. Sakai, *Tetrahedron Lett.*, 1984, **25**, 2183.
16 Y. Gao, R. M. Hanson, J. M. Klunder, S. Y. Ko, H. Masamune and B. K. Sharpless, *J. Am. Chem. Soc.*, 1987, **109**, 5765.
17 A. Klapars and S. L. Buchwald, *J. Am. Chem. Soc.*, 2002, **124**, 14844.
18 L. Stevenson, S. L. Pimlott and A. Sutherland, *Tetrahedron Lett.*, 2007, **48**, 7137.
19 A. Costa, C. Najera and J. M. Sansano, *J. Org. Chem.*, 2002, **67**, 5216.
20 E. A. Couladouros, I. C. Soufli, V. I. Moutsos and R. K. Chadha, *Chem.–Eur. J.*, 1998, **4**, 33.

