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Enantioselectivity of Some 1-(Benzofuran-2-yl)-1-(1-*H*-imidazol-1-yl) Alkanes as Inhibitors of $P450_{Arom}$

G. ALI KHODARAHMI, H. JOHN SMITH, PAUL J. NICHOLLS AND MASOUD AHMADI

Welsh School of Pharmacy, Cardiff University, Cathays Park, Cardiff CF1 3XF, UK

Abstract

The low stereospecificity of the enantiomers of 1-[(benzofuran-2-yl)-4-chlorophenylmethyl]imidazole (**6**, R = H, R′ = 4′-Cl) and the corresponding 4-fluoro compound as inhibitors of aromatase ($P450_{Arom}$) has been explored using 1-(5,7-dichlorobenzofuran-2-yl)-1-(1*H*-imidaz-1-yl)ethane (**7**, $R_1 = R_2 = Cl$, $R = CH_3$), -propane (**7**, $R_1 = R_2 = Cl$, $R = C_2H_5$), and the corresponding 5,7-dibromo compounds resolved as their dibenzoyl-D (or -L) tartrates.

Low enantioselectivity ratios of 4·8 (5,7-diCl) and 12·6 (5,7-diBr) were shown for the ethanes. The values for the corresponding propanes were 8·3 and 5·2, respectively, and for these compounds the stereoselectivity was reversed.

The stereochemistry of enzyme inhibitors with chiral centres is usually important in determining their in-vitro potency against a specific enzyme, although in-vivo the situation can be less clear because of enantiomer interconversion of an enzymic (Kaye 1991) or non-enzymic nature (Pepper et al 1994).

The final step in the biosynthesis of oestrogens from cholesterol is catalysed by aromatase ($P450_{Arom}$). In postmenopausal women androstenedione produced by the adrenal steroidogenic pathway is converted by peripheral and breast $P450_{Arom}$ to oestrone and then by the action of ubiquitous 17β-hydroxysteroid dehydrogenase to the potent oestrogen, oestradiol. Aromatase has been the target for the design of inhibitors as agents in the treatment of breast cancer in postmenopausal women after relapse on the oestrogen-antagonist tamoxifen (Brodie 1994). Inhibition of $P450_{Arom}$ reduces plasma oestrogen levels and consequently the stimulus to growth of oestrogen-dependent metastases.

Aminoglutethimide (Figure 1, **1**) has long been established as a non-steroidal, reversible inhibitor of $P450_{Arom}$; it is used clinically as the racemate for the treatment of breast cancer. The activity lies in the (+)-*R* form of **1** which is approximately 38-fold more potent than the (−)-*S* form (Graves & Salhanick 1979). Rogletimide (pyridoglutethimide; Figure 1, **2**), an analogue of aminoglutethimide, was at one time in clinical trials; the inhibitory potency of this compound resides mainly in the (+)-*R* form (20-fold more potent than the (−)-*S* form) (McCague et al 1989). 1-Alkylation of rogletimide, especially with longer chains, increases potency, but the activity is inverted in the most active member of the series, the 1-octyl derivative, and resides in the (−)-*S* form (Laughton et al 1990). This change is attributable to an alternative mode of binding to the active site. In the 3-(4-aminophenyl)pyrrolidine-2,5-dione series (Figure 1, **3**), ring-contracted analogues of aminoglutethimide, activity lies almost completely in the 1-methyl- and 1,3-dimethyl-substituted and the unsubstituted compounds with the (+)-*R* configuration. Inversion of activity occurs in the 1-pentyl and 1,3-dipentyl analogues where it resides almost completely in the (−) form, argued to have the *S* configuration, in a comparable manner to that noted for 1-alkyl rogletimide derivatives (Pepper et al 1995). The activity of 3-cyclohexylamino-glutethimide resides in the (+)-*S* form (same configuration as *R*-aminoglutethimide) which is 30-fold more potent than the (−)-*R* form (Hartmann et al 1990).

The non-selective imidazole antifungals ketoconazole and econazole are stereospecific towards $P450_{Arom}$. The activity of these compounds resides, respectively, in the *trans*-2*S*,4*S* form (9·6 times the activity of the 2*R*,4*R* form; Rotstein et al 1992) and in the (+) form (30 times the activity of the (−) form; Pepper et al 1995).

Within the second and third generation of $P450_{Arom}$ inhibitors used clinically the (−)-*S* form of fadrozole (Figure 1, **4**) (±) = 400 × aminoglutethimide) is 200 times more active than the (+)-*R* form (Furet et al 1993) and for the

Correspondence: H. J. Smith, Welsh School of Pharmacy, Cardiff University, Cathays Park, Cardiff CF1 3XF, UK.

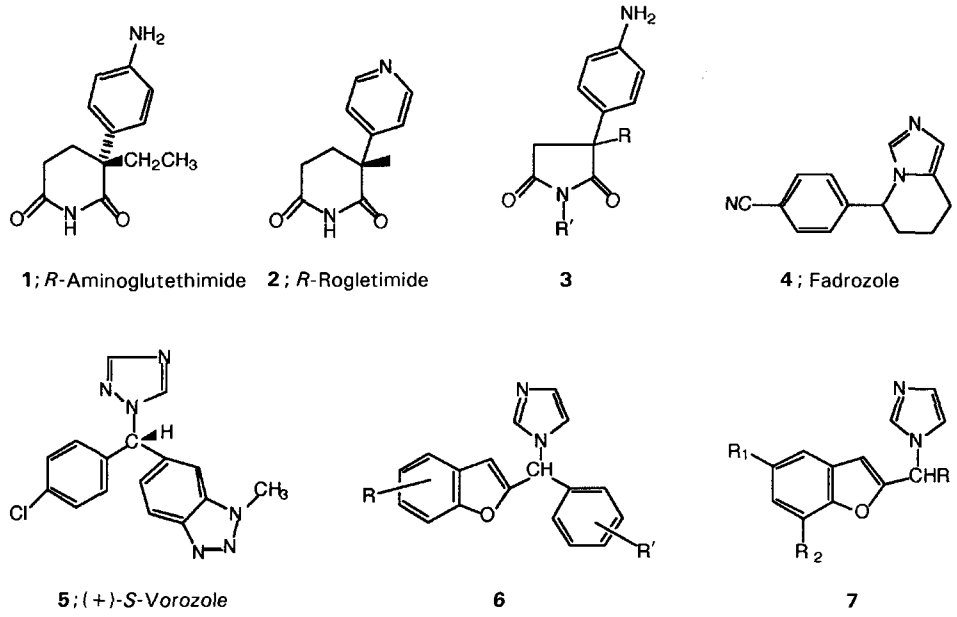


Figure 1. The structures of compounds investigated.

resolved triazole (Figure 1, **5**) the (+)-*S* vorozole (1000 × aminoglutethimide) is 32 times more active than the (−)-*R* form (Vanden Bossche et al 1990). Potent triazoles now licensed (i.e. letrozole, arimidex) are achiral.

We have found that a series of (±)-1-[(benzofuran-2-yl)phenylmethyl]imidazoles (Figure 1, **6**) are potent inhibitors of placental microsomal $P450_{Arom}$, being 80–1000 times more potent than (±)-aminoglutethimide. Resolution of two of these compounds unexpectedly resulted in reduced stereoselectivity in that both enantiomers of the 4′-chloro compound (**6**, R = H, R′ = 4′-Cl) had identical potencies (IC50 = 8·4 nM) and the (+) and (−) forms of the 4′-fluoro compound (6, R = H, R′ = 4′-F) had IC50 values of 5·3 and 65·0 nM, respectively. In view of this unusual reduced stereoselectivity within highly potent inhibitors of $P450_{Arom}$ we have studied the effect on stereoselectivity of replacement of the phenyl group by methyl and ethyl functions within the related 1-(benzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)alkanes (Figure 1, **7**).

Materials and Methods

Chemistry

The reagents used were either general purpose or analytical grade and from Aldrich (Gillingham, Dorset, UK), BDH (Poole, Dorset, UK) or Lancaster Synthesis (Morecombe, Lancs, UK). Solvents for resolution studies were from Fisher Scientific (Loughborough, UK).

Melting points were determined with an Electrothermal instrument and infrared spectra were determined with a Perkin-Elmer 681 infrared spectrophotometer. Optical rotation was measured with a Bellingham and Stanley polarimeter. Thin-layer chromatography (TLC) was performed on aluminium-backed silica gel plates with fluorescent indicator. ^{1}H NMR spectra were determined with a Perkin-Elmer R32 (90 MHz) spectrophotometer; TMS was used as internal standard. Mass spectra were acquired by the SERC Mass Spectrometry Centre, University of Wales College of Swansea, UK. Elemental analyses were determined by the School of Pharmacy, University of London, UK.

Synthesis

Imidazoles **8–14** were synthesized by the general method shown in Figure 2.

1-(Benzofuran-2-yl)-1-(1-H-imidazol-1-yl)ethane HCl (8)

1-(Benzofuran-2-yl)ethan-1-ol (21). 1-(Benzofuran-2-yl)ethanone (Aldrich; 10 g, 62·4 mmol) was dissolved in dioxane (75 mL). At a temperature below 20°C, sodium borohydride (2·63 g, 62·4 mmol) was added and the resulting solution was stirred at room temperature overnight. Excess sodium borohydride was neutralized with 2% HCl and the mixture evaporated under vacuum to small volume. Water (100 mL) was added and the resulting oil extracted with ether (2 × 50 mL). The combined organic phases were washed with water (2 × 50 mL) and dried ($MgSO_4$). The solvent was evaporated to leave a red-brown oil (9 g). Crystallization from *n*-hexane gave the alcohol (**21**) as a cream solid (8·6 g, 85%) mp 39–40°C. (Elliot 1951, mp 41°C). ν_{max}: 3350 (br, OH), 3150 (Ar,

CH), 2980 and 2930 (CH), 1600 (Ar) cm^{-1}. δ_H: 7·5 (4H, m benzofuran 4, 5, 6, 7-H), 6·65 (1H, s benzofuran 2-H), 5·05 (1H, q, J = 7 Hz C*H*–OH), 2·5 (1H, s, CH–O*H*), 1·6 (3H, d, J = 7 Hz, CH_3).

*1-(Benzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane HCl (8).* Thionyl chloride (5·93 g, 50 mmol) in acetonitrile (20 mL) was added dropwise to a solution of imidazole (13·58 g, 200 mmol) in acetonitrile (70 mL) while stirring at a temperature below 10°C. The resulting precipitate was left at 10°C for 1 h and then a solution of 1-(benzofuran-2-yl)ethan-1-ol (**21**) (8·12 g, 50 mmol) in acetonitrile (50 mL) and potassium carbonate (6·9 g, 50 mmol) was added. The suspension was stirred at room temperature for 4 days. The solution was filtered and the solvent evaporated under vacuum to leave an orange oil which was extracted with ether (3 × 100 mL). The extracts were combined and washed with NaOH (2%, 3 × 100 mL) and then water (2 × 100 mL). The organic phase was extracted with HCl (5%; 2 × 100 mL) and the combined aqueous solutions were then made alkaline with NaOH (5%) and the oil which separated was extracted with ether (3 × 50 mL). The combined ether extracts were washed with water (2 × 50 mL) and dried ($MgSO_4$). The solvent was evaporated to leave an orange oil (3·6 g). The oil was dissolved in ether (20 mL) and HCl gas was bubbled through the solution to yield a sticky semi-solid. The solvent was removed and the residue crystallized from acetone to yield a white crystalline solid (1·93 g, 15·5%) mp 155–155·5°C. Found: C, 62·7; H, 5·48; N, 11·13%; M^+, 212·0950. $C_{13}H_{12}N_2O$ requires C, 62·77; H, 5·27; N, 11·26%; M^+, 212·0957. ν_{max}: 3000 (Ar, CH), 2910 (CH), 3200–2500 (HCl salt), 1600 (Ar) cm^{-1}. δ_H: 9·96 (1H, s, imidazole 2-H), 7·63 (1H, d, J = 7·7 Hz, benzofuran 7-H), 7·48 (1H, d, J = 8·2 Hz, benzofuran 4-H), 7·45–7·20 (4H, m, benzofuran 5, 6-H and imidazole 4, 5-H), 7·02 (1H, s, benzofuran 3-H), 6·40 (1H, q, J = 6·9 Hz, C*H*–CH_3), 2·1 (3H, d, J = 7 Hz, CH–C*H*$_3$).

*1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-*H*-imidazole-1-yl)ethane HCl (9) 1-(5,7-Dichlorobenzofuran-2-yl)ethanone (15).*
3,5-Dichlorosalicylaldehyde (10 g, 52·4 mmol) in methanol (50 mL) was added dropwise to a solution of sodium methoxide (3·68 g, 68·1 mmol) in methanol (100 mL). A solution of chloroacetone (6·64 g, 68·1 mmol) in methanol (30 mL) was added and the solution heated under reflux for 3 h. The solvent was evaporated to leave a red oil which was dissolved in ether (200 mL). The ether layer was washed with water (6 × 50 mL), NaOH (5%,

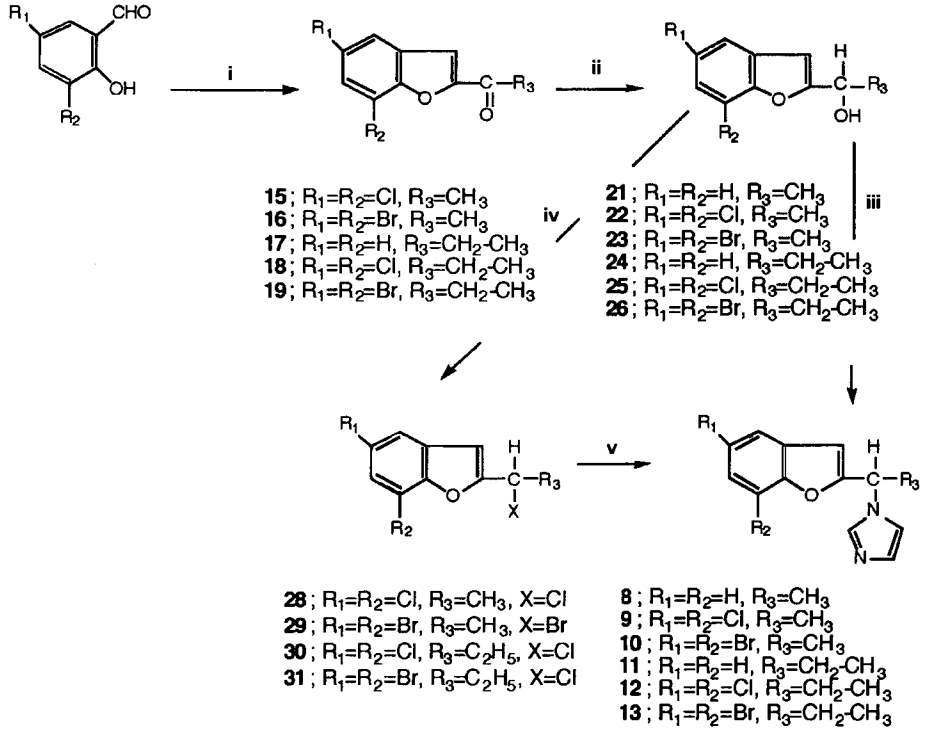


Figure 2. The synthetic pathway. Reagents: (i) CH_3COCH_2Cl, $NaOCH_3$, CH_3OH, reflux, 3 h or $CH_3CH_2COCH_2Br$, NaH, DMF, 110–120°C, 1 h, $NaOCH_3$, 110–120°C, 2 h; (ii) $NaBH_4$, dioxane, room temperature; (iii) $SOCl_2$, imidazole, K_2CO_3, CH_3CN, room temperature, 4 days; (iv) $SOCl_2$ or $SOBr_2$, cyclohexane–CH_2Cl_2, 60–80°C, 3–10 h; (v) imidazole, CH_3CN, reflux, 20 h, HCl gas.

3 × 100 mL) and again with water (4 × 50 mL) and then dried ($MgSO_4$). The solvent was evaporated to leave the ketone (**15**) as a solid which was recrystallized from ethanol to give needle-shaped cream crystals (4·7 g, 39%) mp 124°C (Binon et al 1966, mp 124°C). ν_{max}: 3182 (Ar CH), 3097 and 3079 (CH) 1686 (C=O), 1576 (Ar) cm^{-1}. δ_H: 7·8–7·5 (3H, m, benzofuran), 2·65 (3H, s, CH_3).

*1-(5,7-Dichlorobenzofuran-2-yl)ethan-1-ol (**22**).* 1-(5,7-Dichlorobenzofuran-2-yl)ethanone (**15**) with sodium borohydride using the general method described for **21** gave a pale-yellow oil, mp 78–79°C from *n*-hexane (92·6%). Found: C, 52·05; H, 3·63%; M^+, 229·99014. $C_{10}H_8Cl_2O_2$ requires C, 51·96; H, 3·49%; M^+, 229·99014. ν_{max}: 3300 (OH), 3084 (Ar CH), 2994 and 2922 (CH), 1612 and 1592 (Ar) cm^{-1}. δ_H: 7·50 (1H, d, J = 2 Hz, benzofuran 6-H), 7·4 (1H, d, J = 2 Hz, benzofuran 4-H), 6·7 (1H, s, benzofuran 3-H), 5·15 (1H, q, J = 7 Hz, −C*H*−OH), 2·4 (1H, br s, CH−O*H*), 1·65 (3H, d, J = 7 Hz, CH−CH_3).

*Crude 1-chloro-1-(5,7-dichlorobenzofuran-2-yl) ethane (**28**).* Thionyl chloride (8·54 g, 71·8 mmol) in dichloromethane (20 mL) was added dropwise to a solution of 1-(5,7-dichlorobenzofuran-2-yl)ethan-1-ol (**22**) (4·15 g, 17·95 mmol) in cyclohexane-dichloromethane (2 : 1, v/v, 150 mL) and the resulting solution was heated at 60°C for 3 h. The solvent was evaporated to leave a pale-grey oil. The residue was shown by TLC to contain at least two products chromatographing together and some of the starting material. The residual oil was passed down a silica gel column; use of petroleum ether-ether, 19 : 1 and then 9 : 1, as eluents enabled the elution of the first spot in small quantity and the elution of the remainder as a mixture. The first spot, upon evaporation of the solvent gave a white solid, mp 138–139°C. Found: C, 53·94; H, 3·38%; M^+, 441·9697. $C_{20}H_{14}Cl_4O_3$ requires C, 54·07; H, 3·18%; M^+, 441·9697. ν_{max}: 3082 (Ar CH), 2978 and 2931 (CH), 1613 (Ar) cm^{-1}. δ_H: 7·55 (2H, d, J = 2 Hz, 2 × benzofuran 6-H), 7·42 (2H, d, J = 2 Hz, 2 × benzofuran 4-H), 6·77 (2H, s, 2 × benzofuran 3-H), 4·85 (2H, q, J = 7 Hz, 2 × −O−C*H*−CH_3), 1·65 (6H, d, J = 7 Hz, 2 × −O−CH−CH_3).

*1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane HCl (**9**).* The crude oil from the previous step in acetonitrile (50 mL) was added to a hot mixture of imidazole (3·51 g, 51·6 mmol) and potassium carbonate in acetonitrile (100 mL) and the resulting mixture was heated under reflux for 20 h. The solution was filtered and the solvent removed to leave an oil which was worked up as described for **8** to give an orange oil. 1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane HCl (**9**) was prepared by bubbling HCl gas through an ethereal solution of the oil. The gum-like residue formed was crystallized from acetone to give white crystals (210 mg, 0·03%) mp 176–177°C. Found: C, 48·03; H, 3·66; N, 8·64%; M^+, 280·0170 (free base). $C_{13}H_{11}Cl_3N_2O$ requires C, 47·78; H, 3·70; N, 8·57%; M^+, 280·0180 (free base). ν_{max}: 3500–2600 (HCl salt), 3098 (Ar CH), 3000 (CH), 1615 (Ar) cm^{-1}. δ_H: 10·7 (1H, s, imidazole 2-H), 7·6–7·45 (4H, m, benzofuran 4, 6-H and imidazole 4, 5H), 7·2 (1H, s, benzofuran 3-H), 6·65 (1H, q, J = 7 Hz, −C*H*−CH_3), 2, 12 (3H, d, J = 7 Hz, −CH−CH_3).

*1-(5,7-Dibromobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane (**10**)*

*1-(5,7-Dibromobenzofuran-2-yl)ethanone (**16**).* 3,5-Dibromosalicylaldehyde with chloroacetone according to the general method described for **15** gave **16** as a white solid which was crystallized from methanol (39·3%) mp 152–153°C (Binon et al 1966, mp 150°C). ν_{max}: 3094 (Ar CH), 1693 (C=O) 1568 (Ar) cm^{-1}. δ_H: 7·92 (2H, s, benzofuran 4, 6-H), 7·6 (1H, s, benzofuran 3-H), 2·7 (3H, s, CH_3).

*1-(5,7-Dibromobenzofuran-2-yl)ethan-1-ol (**23**).* 1-(5,7-Dibromobenzofuran-2-yl)ethanone (**16**) with sodium borohydride according to the general method described for **21** gave **23** as a solid which was recrystallized from cyclohexane to give a pale-cream solid (88·7%) mp 102·5–103°C. Found: M+NH_4^+ 337·9225. $C_{10}H_8Br_2O_2{}^-NH_4^+$ requires M+NH_4^+ 337·9220. ν_{max}: 3302 (OH), 2991 and 2919 (CH), 1606 and 1592 (Ar) cm^{-1}. δ_H: 7·70 (1H, d, J = 2 Hz, benzofuran 6-H), 7·65 (1H, d, J = 2 Hz, benzofuran 4-H), 6·7 (1H, s, benzofuran 3-H), 5·1 (1H, q, J = 7·8 Hz, −C*H*−CH_3), 2·4 (1H, br s, OH), 1·65 (3H, d, J = 7·8 Hz, −CH_3).

*1-Bromo-1-(5,7-dibromobenzofuran-2-yl)ethane (**29**).* 1-(5,7-Dibromobenzofuran-2-yl)ethan-1-ol (**23**) (1·69 g, 5·28 mmol) in cyclohexane-dichloromethane (1 : 1 v/v, 30 mL) was added to a stirred solution of thionyl bromide (2·2 g, 10·56 mmol) in cyclohexane (20 mL) and the solution heated at 70–80°C for 10 h. The solvent was then evaporated to leave an oil which solidified when washed with petroleum ether (40–60°C) to give the bromo derivative (**29**) as a dark-cream solid (1·51 g, 74·8%) mp 94–95°C. ν_{max}: 3100 (Ar CH), 2967 (CH), 1586 (Ar) cm^{-1}. δ_H: 7·73 (2H, s, benzofuran

4, 6-H), 6·82 (1H, s, benzofuran 3-H), 5·4 (1H, q, J = 7·8 Hz, $-CH-CH_3$), 2·15 (3H, d, J = 7·8 Hz, $CH-CH_3$).

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-imidazol-1-yl)ethane HCl (10). 1-Bromo-1-(5,7-dibromobenzofuran-2-yl)ethane (**29**) with imidazole using the general method described for **9** gave an orange oil (430 mg, 32%) which gave the HCl salt as a pale-cream solid, mp 170–171°C. Found: C, 38·35; H, 2·89; N, 6·63%; M^+, 367·9160 (free base). $C_{13}H_{10}Br_2N_2O.HCl$ requires C, 38·41; H, 2, 73; N, 6·89%; M^+, 367·9170 (free base). ν_{max}: 3084 (ArCH), 2921 (CH), 1607 (Ar) cm^{-1}. δ_H: 10·75 (1H, s, imidazole 2H), 7·8-7·4 (4H, m, benzofuran 4, 6-H and imidazole 4, 5-H), 7·2 (1H, s, benzofuran 3-H), 6·65 (1H, q, J = 7·8 Hz, $CH-CH_3$), 2·05 (3H, d, J = 7·8 Hz, CH_3).

1-(Benzofuran-2-yl)-1-(1-H-imidazol-1-yl)propane (11) 1-(Benzofuran-2-yl)propan-1-one (17). Salicyl-aldehyde (2·83 g, 23·1 mmol) in dimethylformamide (DMF) (20 mL) was added dropwise to a stirred solution of oil-free sodium hydride (0·96 g, 40 mmol) in DMF (30 mL). This was left at room temperature for 20 min to liberate hydrogen and gave a bright yellow solution of the sodium salt. 1-Bromo-2-butanone (3·5 g, 23·1 mmol) in DMF (30 mL) was then added dropwise and the resulting solution was heated at 110–120°C for 1 h. Sodium methoxide (0·45 g, 8·27 mmol) was then added and the solution was heated for another 2 h at 110–120°C. The solvent was evaporated, water (50 mL) was added and the residue was extracted with ether (4 × 30 mL). The combined extracts were washed with water (3 × 20 mL) and dried ($MgSO_4$). The solvent was evaporated to leave an orange oil. The oil was passed down a dry silica gel column, using petroleum ether–ether (1 : 1) as eluent. The eluate, upon concentration under vacuum, gave the ketone (**17**) as a pale-yellow solid (2·8 g, 69·6%) mp 48–49°C. Found: C, 72·24; H, 6·06. $C_{11}H_{10}O_2.0·5H_2O$ requires C, 72·12; H, 6·05%. ν_{max}: 2980 and 2930 (C–H), 1685 (C=O), 1615 (Ar) cm^{-1}. δ_H: 7·76 (1H, d, J = 7·9 Hz, benzofuran 7-H), 7·64 (1H, d, J = 8·5 Hz, benzofuran 4-H), 7·56 (1H, s, benzofuran 3-H), 7·53 (1H, t, J = 7·45 Hz, benzofuran 6-H), 7·36 (1H, t, J = 7·2 Hz, benzofuran 5-H), 2·5 (2H, q, J = 7·23 Hz, CH_2-CH_3), 1·29 (3H, t, J = 7·26, $-CH_2-CH_3$).

1-(Benzofuran-2-yl)propan-1-ol (24). 1-(Benzofuran-2-yl)propan-1-one (**17**) (2·35 g, 13·5 mmol) with sodium borohydride (0·92 g, 24 mmol) using the general method described for **21** gave a pale-brown oil. The oil was passed down a column of dry silica gel, using petroleum ether–ether (1 : 1) as eluent; the eluate was evaporated to give **24** as a pale-yellow oil (90·8%). Found: C, 71·54; H, 6·91. $C_{11}H_{12}O_2.0·5H_2O$ requires C, 71·33; H, 7·07%. ν_{max}: 3450 (OH), 2970 and 2940 (CH), 1610 (Ar) cm^{-1}. δ_H: 7·61 (1H, d, J = 8·3 Hz, benzofuran 7-H), 7·52 (1H, d, J = 8 Hz, benzofuran 4-H), 7·33 (1H, t, J = 8·1 Hz, benzofuran 6-H), 7·27 (1H, t, J = 7·7 Hz, benzofuran 5-H), 6·67 (1H, s, benzofuran 3-H), 4·8 (1H, t, J = 6·6 Hz, $CH-OH$), 2·45 (1H, br s, $CH-OH$), 2·1–1·9 (2H, m, $-CH-CH_2-CH_3$), 1·08 (3H, t, J = 7·4 Hz, $-CH_2-CH_3$).

1-(Benzofuran-2-yl)-1-(1-H-imidazol-1-yl)propane (11). 1-(Benzofuran-2-yl)propan-1-ol (**24**), thionyl chloride and imidazole using the general method described for **8** gave a pale-yellow oil. The oil was passed down a silica gel column using chloroform–methanol (29 : 1) as eluent. Evaporation of the eluate gave **11** as a pale-yellow oil (29·7%). Found: C, 74·01; H, 6·39; N, 12·26%; M^+, 226·1106. $C_{14}H_{14}N_2O$ requires C, 74·31; H, 6·23; N, 12·38%; M^+, 226·1116. ν_{max}: 3111 (Ar CH), 2970 and 2940 (CH), 1610 and 1586 (Ar) cm^{-1}. δ_H: 7·75 (1H, s, imidazole 2-H), 7·6 (1H, d, J = 7·15 Hz, benzofuran 7-H), 7·5 (1H, d, J = 7·4 Hz, benzofuran 4-H), 7·4–7·2 (2H, m, benzofuran 5, 6-H), 7·14 (1H, br s, imidazole 5-H), 7·10 (1H, br s, imidazole 4-H), 6·67 (1H, s, benzofuran 3-H), 5·2 (1H, t, J = 7·7 Hz $-CH-CH_2-$), 2·5–2·2 (2H, m, $-CH-CH_2-$), 1·0 (3H, t, J = 7·4 Hz, $-CH_2-CH_3$).

1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-H-imidazol-1-yl)propane (12) 1-(5,7-Dichlorobenzofuran-2-yl)propan-1-one (18).
3,5-Dichlorosalicylaldehyde with 1-bromo-2-butanone using the method described for **17** gave a brownish oil which was crystallized from ethanol to give **18** as a cream solid (30·5%) mp 75–76°C. Found: C, 54·05; H, 3·36%; M^+, 241·9901. $C_{11}H_{14}Cl_2O_2$ requires C, 54·33; H, 3·22%; M^+, 241·9901. ν_{max}: 3090 (Ar CH), 2988 and 2940 (CH), 1684 (C=O), 1616 and 1580 (Ar) cm^{-1}. δ_H: 7·63 (1H, d, J = 1·7 Hz, benzofuran 6-H), 7·52 (1H, d, J = 1·7 Hz, benzofuran 4-H), 7·5 (1H, s, benzofuran 3-H), 3·10 (2H, q, J = 7·3 Hz, $-CH_2-CH_3$), 1·32 (3H, t, J = 7·3 Hz, $-CH_3$).

1-(5,7-Dichlorobenzofuran-2-yl)propan-1-ol (25). 1-(5,7-Dichlorobenzofuran-2-yl)propan-1-one (**18**) with sodium borohydride using the general method described for **21** gave the alcohol **25** as an orange oil (6·7%). Found: C, 52·20; H, 4·39.

$C_{11}H_{10}Cl_2O_2$ $0{\cdot}5H_2O$ requires C, 51·98; H, 4·36%. ν_{max}: 3361 (OH), 2966 and 2878 (CH), 1610 and 1595 (Ar) cm^{-1}. δ_H: 7·5 (1H, d, J = 1·9 Hz, benzofuran (6-H), 7·37 (1H, d, J = 1·9 Hz, benzofuran 4-H), 6·71 (1H, s, benzofuran 3-H), 4·89 (1H, t, J = 6·6 Hz, $-CH-CH_2-$), 2·52 (1H, br s, $-CH-OH$), 2·2–1·9 (2H, m, $-CH-CH_2-$), 1·50 (3H, t, J = 7·4 Hz, $-CH_3$).

1-Chloro-1-(5,7-dichlorobenzofuran-2-yl)propane (30). 1-(5,7-Dichlorobenzofuran-2-yl)propan-1-ol (**26**) with thionyl chloride using the general method for **28** gave a brownish oil which was used directly for the next step without purification. ν_{max}: 3083 (Ar CH), 2973 and 2936 (CH), 1612 and 1593 (Ar) cm^{-1}.

1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-H-imidazol-1-yl)propane (12). 1-Chloro-1-(5,7-dichlorobenzofuran-2-yl)propane (**30**) with imidazole using the general method described for **9** gave **12** as a pale-brown oil (37·3%). Found: C, 56·76; H, 4·21; N, 9·24%; M^+, 294·0327. $C_{14}H_{12}C_{12}N_2O$ requires C, 56·95; H, 4·1; N, 9·49%; M^+, 294·0337. ν_{max}: 3111 (Ar CH), 2970 and 2931 (CH), 1616 and 1575 (Ar) cm^{-1}. δ_H: 7·75 (1H, s, imidazole 2-H), 7·45 (1H, d, J = 1·9 Hz, benzofuran 6-H), 7·30 (1H, d, J = 1·8 Hz, benzofuran 4-H), 7·18 (1H, br s, imidazole 5-H), 7·12 (1H, br s, imidazole 4-H), 6·60 (1H, s, benzofuran 3-H), 5·30 (1H, t, J = 7·7 Hz, $-CH-CH_2-$), 2·6–2·2 (2H, m, $CH-CH_2-$), 1·1 (3H, t, J = 7 Hz, CH_3).

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-imidazol-1-yl)propane (13) 1-(5,7-Dibromobenzofuran-2-yl)propan-1-one (19). 3,5-Dibromosalicylaldehyde with 1-bromo-2-butanone using the general method described for **17** gave a red oil which was crystallized from methanol to give **19** as a pale-orange solid (27·8%) mp 107°C. Found: M^+ 329·8891. $C_{11}H_8Br_2O_2$ requires M^+ 329·8891. ν_{max}: 3100 (Ar CH), 2985 and 2940 (CH), 1686 (C=O), 1556 (Ar) cm^{-1}. δ_H: 7·85 (1H, d, J = 1·8 Hz, benzofuran 6-H), 7·81 (1H, d, J = 1·8 Hz, benzofuran 4-H), 7·52 (1H, s, benzofuran 3-H), 3·1 (2H, q, J = 7·3 Hz, CH_2-CH_3), 1·32 (3H, t, J = 7·3 Hz, CH_3).

1-(5,7-Dibromobenzofuran-2-yl)propan-1-one (26). 1-(5,7-Dibromobenzofuran-2-yl)propan-1-one (**19**) with sodium borohydride using the general method described for **21** gave the alcohol **26** as an orange oil (98%). ν_{max}: 3355 (OH), 3075 (Ar CH), 2966 and 2933 (CH), 1605 and 1566 (Ar) cm^{-1}. δ_H: 7·78 (1H, s, benzofuran 6-H), 7·75 (1H, s, benzofuran 4-H), 6·7 (1H, s, benzofuran 3-H), 4·85 (1H, t, J = 6·8 Hz, $-CH-OH$), 2·3 (1H, br s, $-CH-OH$), 2·2–1·8 (2H, m, $-CH_2-CH_3$), 1·07 (3H, t, J = 7 Hz, $-CH_2-CH_3$).

1-Chloro-1-(5,7-dibromobenzofuran-2-yl)propane (31). 1-(5,7-Dibromofuran-2yl)propan-1-ol (**26**) with thionyl chloride by the general method for **28** gave a dark-orange oil which was used for the next step without further purification. ν_{max}: 3076 (Ar CH), 2970 and 2926 (CH), 1606 (Ar) cm^{-1}.

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-imidazol-1-yl)propane (13). 1-Chloro-1-(5,7-dibromobenzofuran-2-yl)propane (**31**) with imidazole using the general method described for **9** gave 1-(5,7-dibromobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl) propane (**13**) as a pale-yellow oil (33%). Found: C, 43·73; H, 3·23; N, 7·11%; M^+, 381·9316. $C_{14}H_{12}Br_2N_2O$ requires C, 43·78; H, 3·15; N, 7·29%; M^+, 381·9327. ν_{max}: 3111 (Ar CH), 2970 and 2936 (CH), 1606 and 1566 (Ar) cm^{-1}. δ_H: 7·75 (1H, s, imidazole 2-H), 7·63 (1H, d, J = 1·7 Hz, benzofuran 6-H), 7·60 (1H, d, J = 1·6 Hz, benzofuran 4-H), 7·18 (1H, br s, imidazole 5-H), 7·13 (1H, br s, imidazole 4-H), 6·63 (1H, s, benzofuran 3-H), 5·27 (1H, t, J = 7·7 Hz, $-CH-CH_2-$), 2·55–2·1 (2H, m, $-CH_2-CH_3$), 1·05 (3H, t, J = 7·3 Hz, $-CH_2-CH_3$).

Resolution studies

1-(Benzofuran-2-yl)-1-(1-H-imidazol-1-yl)ethane dibenzoyl-D-(+)-tartrate, (+)-8. 1-(Benzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane HCl (**8**) (1·8 g, 7·23 mmol) was mixed with NaOH (5%, 100 mL) and the resulting solution extracted with ether (3 × 40 mL), washed with water (3 × 50 mL) and dried ($MgSO_4$). The solvent was evaporated to give the free base as a pale-cream oil (1·48 g, 6·97 mmol). A solution of dibenzoyl-D-(+)-tartaric acid (2·5 g, 6·97 mmol) in ethanol (8 mL) was added dropwise to a hot solution of the free base (1·48 g, 6·97 mmol) in ethanol (8 mL) and the solution was heated on a boiling-water bath for 5 min. The solution was then cooled on ice, scratched for 5 min and left overnight at room temperature to give a white solid. The solid was removed by filtration (2·24 g) and recrystallized a further six times from ethanol, at room temperature, to give the pure salt (+)-**8** as fine white crystals (390 mg, 9·8%), mp 133–134°C. $[\alpha]^D_{25}$ (1% w/v, methanol) = +79·12° (dibenzoyl-D-(+)-tartaric acid salt). $[\alpha]^D_{25}$ (1% w/v, chloroform) = +28·48° (released free base). The recrystallization steps were monitored after each recrystallization by high-performance liquid chromatography (HPLC) of the released base.

Attempts to isolate (−)-**8** as the dibenzoyl-L-(−) tartrate from the mother liquors were unsuccessful. The use of D-(−)-tartaric acid and 3-bromocamphor-8-sulphonate as resolving agents for **8** did not give crystalline material.

1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)ethane dibenzoyl-L*-(−)-tartrate, (−)-***9***. Dibenzoyl-L-(−)-tartaric acid (102 mg, 0·28 mmol) in ether (2 mL) was added to a solution of the free base (80 mg, 0·28 mmol) of 1-(5,7-dichlorobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane HCl (**9**) in ether (2 mL) to give a white solid. The solid was recrystallized twice from ethanol to give (−)-**9** as a white crystalline solid (30 mg, 16·5%), mp 138·8–139·1°C. $[\alpha]^{D}_{25}$ (1% w/v, methanol) = −63·19° (dibenzoyl-L-(−)-tartaric acid salt).

1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)ethane dibenzoyl-D*-(+)-tartrate, (+)-***9***. The mother liquors of (−)-**9** after the first and second recrystallization from ethanol were evaporated and the residue was reacted with a solution of K_2CO_3 (5%, 20 mL) and the mixture extracted with ether (3 × 20 mL), washed with water (3 × 20 mL), dried ($MgSO_4$) and the solvent evaporated to leave a pale-yellow oil (50 mg). Dibenzoyl-D-(+)-tartaric acid (64 mg, 0·18 mmol) in ether (2 mL) was added to a solution of the oil (50 mg, 0·18 mmol) in ether (3 mL) to give a white solid. The solid was recrystallized twice from ethanol to give (+)-**9** as white crystals (20 mg, 17·6%) mp 141–141·5°C. $[\alpha]^{D}_{25}$ (1% w/v, methanol) = +63·85° (dibenzoyl-D-(+)-tartaric acid salt).

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)ethane dibenzoyl-D*-(+)-tartrate, (+)-***10***. Dibenzoyl-D-(+)-tartaric acid (87 mg, 0·24 mmol) in ether (3 mL) was added to a solution of the free base from 1-(5,7-dibromobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)ethane HCl (**10**) (90 mg, 0·24 mmol) in ether (4 mL) to give a white solid. The solid was suspended in methanol (8 mL) and heated under reflux until dissolution was complete. The solution left to cool to room temperature and stand overnight at room temperature. The precipitate, a white solid, was removed by filtration and recrystallized from methanol to yield (+)-**10** as needle-shaped white crystals (40 mg, 22·6%) mp 151–151·5°C. $[\alpha]^{D}_{30}$ (1% w/v, methanol) = +78° (dibenzoyl-D-(+)-tartaric acid salt).

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)ethane dibenzoyl-L*-(−)-tartrate, (−)-***10***. The mother liquors from recrystallization of (+)-**10** were processed in the usual manner, and dibenzoyl-L-(−)-tartaric acid (48 mg, 0·14 mmol) in ether (3 mL) was added to a solution of the oil (50 mg, 0·14 mmol) obtained in ether (3 mL) to give a white solid. The solid was recrystallized from methanol to give (−)-**10** as white needle-shaped crystals (41 mg, 41·7%) mp 150·5–150·7°C. $[\alpha]^{D}_{30}$ (1% w/v, methanol) = −76·4° (dibenzoyl-L-(−)-tartaric acid salt).

1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)propane dibenzoyl-L*-(−)-tartrate, (−)-***12***. Dibenzoyl-L-(−)-tartaric acid (133 mg, 0·37 mmol) in ether (3 mL) was added to a solution of the base 1-(5,7-dichlorobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)propane (**12**; 110 mg, 0·37 mmol) in ether (4 mL) to give a white solid. The solid was recrystallized twice from ethanol to give (−)-**12** as white crystals (50 mg, 20·7%) mp 136·4–136·7°C. $[\alpha]^{D}_{25}$ (1% w/v, methanol) = −43·49°.

1-(5,7-Dichlorobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)propane dibenzoyl-D*-(+)-tartrate, (+)-***12***. The mother liquor from the first and second recrystallizations of (−)-**12** was processed in the usual manner and dibenzoyl-D-(+)-tartaric acid (85 mg, 0·24 mmol) in ether (2 mL) was added to a solution of the oil (70 mg, 0·24 mmol) in ether (3 mL) to yield a white precipitate. The solid was recrystallized from ethanol to give (+)-**12** as a fine crystalline solid (45 mg, 18·7%) mp 136·4–137°C. $[\alpha]^{D}_{25}$ (1% w/v, methanol) = +42·9° (dibenzoyl-D-(+)-tartaric acid salt).

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)propane dibenzoyl-D*-(+)-tartrate, (+)-***13***. Dibenzoyl-D-(+)-tartaric acid (103 mg, 0·29 mmol) in ether (2 mL) was added to a solution of the base 1-(5,7-dibromobenzofuran-2-yl)-1-(1-*H*-imidazol-1-yl)propane (**13**) (110 mg, 0·29 mmol) in ether (4 mL) to give a white solid. The solid was dissolved in ethanol (2 mL) and the solution was heated on a boiling water bath for 5 min and then cooled to room temperature and scratched for 5 min. On standing overnight at room temperature a semi-solid of (+)-**13** was deposited as fine white crystals (70 mg, 32·9%) mp 133–133·5°C. $[\alpha]^{D}_{25}$ (1% w/v, methanol) = +39·07° (dibenzoyl-D-(+)-tartaric acid salt).

1-(5,7-Dibromobenzofuran-2-yl)-1-(1-H-*imidazol-1-yl)propane dibenzoyl-L*-(−)-tartrate, (−)-***13***. The mother liquor of (+)-**13** was processed in the usual manner and dibenzoyl-L-(−)-tartaric acid (65 mg, 0·18 mmol) in ether (2 mL) added to a solution of the oil (70 mg, 0·18 mmol) in ether (2 mL) to give a white solid which was

recrystallized (ethanol) in the same manner as for (+)-**13** to give (−)-**13** as fine white crystals (60 mg, 44·3%) mp 134·5–135°C. $[\alpha]^{D}_{25}$ (1% w/v, methanol) = −37·45° (dibenzoyl-L-(−)-tartaric acid salt).

Determination of enantiomeric purity by HPLC

HPLC was performed with a Milton Roy LC system comprising a Model 3000 constantametric pump, a Rheodyne injection unit and a model 3100 variable-wavelength spectromonitor. A model CL-4100 computing integrator was used to process HPLC data. The amylose-coated silica gel HPLC column (4·6 mm × 250 mm) and precolumn (4·6 × 50 mm) were both packed with identical material (amylose tris(3,5-dimethylphenylcarbamate; Chiralpak AD; Daicel). Injection on to the column was achieved with a Hamilton syringe (50 μL) into a Rheodyne 20-μL loop.

Before connecting the column the pump was washed separately for 10 min with water, methanol and the mobile phase at a flow rate of 5 mL min^{-1}. The guard column and analytical column were fitted and washed with the solvent system using a flow rate of 0·5 mL min^{-1} to obtain a constant absorbance on the detector and a constant pressure from the pump.

*1-(Benzofuran-2-yl)-1-(1-H-imidazol-1-yl)ethane dibenzoyl-D-(+)-tartrate, (+)-**8**.* A solution of potassium carbonate (5%, 10 mL) was added to a sample of (+)-**8** and the free base was extracted with dichloromethane (2 × 10 mL). The organic phase was washed with water (2 × 10 mL) and dried ($MgSO_4$). The solvent was evaporated and the residue was dissolved in the mobile phase (1 mL) and injected (20 μL) into the HPLC. The HPLC conditions were: mobile phase, *n*-hexane–2-propanol–diethylamine, 80 : 20 : 0·1; flow rate, 0·7 mL min^{-1}; pressure, 130–140 psig.

Conditions used for the other racemates and enantiomers were as follows. (±)-, (+)- and (−)-**9**: *n*-hexane–2-propanol–diethylamine, 85 : 15 : 0·1; 0·45 mL min^{-1}, 50–6 psig. (±)-, (+)- and (−)-**10**: *n*-hexane–2-propanol–diethylamine, 90 : 10 : 0·1; 0–20 min 0·7 mL min^{-1} and then 0·4 mL min^{-1}; 130 psig and then 50 psig. (±)-, (+)- and (−)-**12**: *n*-hexane–2-propanol–diethylamine, 93 : 7 : 0·1; 0·45 mL min^{-1}; 50–60 psig. (±)-, (+)- and (−)-**13**: *n*-hexane–2-propanol–diethylamine, 93 : 7 : 0·1; 0·4 mL min^{-1}; 40–50 psig.

The purity of the compounds in respect of the other isomer was calculated from the integration to be: (+)-**8**, 99·28%; (+)-**9**, 99·00%; (−)-**9**, 98·50%; (+)- and (−)-**10**, **12** and **13**, 99·00%.

Biochemistry

Materials

D-Glucose-6-phosphate (monophosphate salt) and NADP (monosodium salt) were from Sigma. D-Glucose-6-phosphate dehydrogenase was from Boehringer-Mannheim. [1β-^{3}H]Androstenedione (40·00 Ci $mmol^{-1}$, 1 mCi (37 MBq) in 1 mL ethanol) was from Dupont (UK). All unlabelled laboratory reagents were AnalaR grade from BDH, Poole, Dorset. Centrifugation was undertaken with a MSE Europa 65M Ultracentrifuge. Radioactivity was determined with a LKB Wallac 1217 Rackbeta liquid scintillation counter. Scintillation fluid was Optiphase Hisafe 3, from FSA Laboratory Supplies, Loughborough, UK.

Protein concentration was determined with a BCA Protein Assay Reagent obtained from Pierce, IL.

NADPH-generating system consisted of glucose-6-phosphate (28·2 mg), NADPH (8·6 mg) and glucose-6-phosphate dehydrogenase (15 μL, 10 international units) in phosphate buffer (pH 7·4, 50 mM, 1 mL).

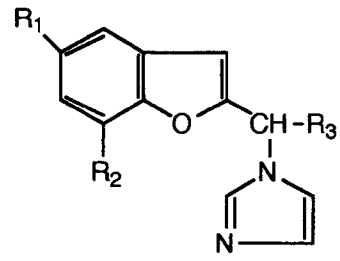
Assay of $P450_{Arom}$

Placental microsomes were prepared by the method of Thompson & Siiteri (1974) and stored at −20°C until required for use. A mixture of [1β-^{3}H]androstenedione and unlabelled androstenedione in propylene glycol (10 μL, 0·6 μM final concentration), NADPH-generating system (50 μL), placental microsomes (10 μL, 0·26 mg protein mL^{-1} final concentration) and phosphate buffer (50 mM, pH 7·4, 430 μL) was incubated, in triplicate, at 37°C for 7 min. Samples (300 μL) were removed and added to 1% charcoal suspension (900 μL) and 0·1 mM mercuric chloride solution (300 μL). The tubes were mixed thoroughly, left to stand on ice for 20 min and centrifuged at 2000 *g* (3000 rev min^{-1}) for 15 min. Samples (500 μL) of the supernatant were removed and transferred to counting vials containing scintillation fluid (3 mL, Hisafe Optiphase III), mixed thoroughly and the [^{3}H]H_2O formed was counted with a liquid-scintillation counter for 5 min to determine the extent of aromatization.

Enzyme inhibition

The compounds (100 μM) in ethanol (10 μL) at a range of concentrations were added to the assay mixture. Control incubations were performed in the absence of inhibitor but with addition of ethanol (10 μL). Aminoglutethimide was used for comparative purposes. IC50 values were calculated from a plot of (% activity remaining) against the logarithm

Table 1. IC50 values of racemates and enantiomers of some substituted 1-(benzofuran-2-yl)-1-(1-*H*-imidazol-1-yl) ethanes and propanes as inhibitors of $P450_{Arom}$.



Compound	R_1	R_2	R_3	IC50 (μM)*
(±)-**8** hydrochloride	H	H	CH_3	4·67
(+)-**8** dibenzoyl-D-(+)-tartaric acid salt	H	H	CH_3	5·62
(±)-**9** hydrochloride	Cl	Cl	CH_3	0·905
(+)-**9** dibenzoyl-D-(+)-tartaric acid salt	Cl	Cl	CH_3	0·49
(−)-**9** dibenzoyl-L-(−)-tartaric acid salt	Cl	Cl	CH_3	2·33
(±)-**10** hydrochloride	Br	Br	CH_3	1·305
(+)-**10** dibenzoyl-D-(+)-tartaric acid salt	Br	Br	CH_3	0·835
(−)-**10** dibenzoyl-L-(−)-tartaric acid salt	Br	Br	CH_3	10·54
(±)-**11** base	H	H	C_2H_5	2·02
(±)-**12** base	Cl	Cl	C_2H_5	0·335
(+)-**12** dibenzoyl-D-(+)-tartaric acid salt	Cl	Cl	C_2H_5	2·32
(−)-**12** dibenzoyl-L-(−)-tartaric acid salt	Cl	Cl	C_2H_5	0·28
(±)-**13** base	Br	Br	C_2H_5	0·91
(+)-**13** dibenzoyl-D-(+)-tartaric acid salt	Br	Br	C_2H_5	1·51
(−)-**13** dibenzoyl-L-(−)-tartaric acid salt	Br	Br	C_2H_5	0·29
Aminoglutethimide	–	–	–	11·00

* Androstenedione, 0·6 μM. Mean of triplicate estimations for which the spread was < 2% of the mean.

of the inhibitor concentration for the racemates and enantiomers (Table 1) using Cricket Graph.

Results and Discussion

The series of (±)-1-[(benzofuran-2-yl)phenylmethyl] imidazoles (**6**) are potent inhibitors of $P450_{Arom}$ having 80–1000 times the inhibitory activity of (±)-aminoglutethimide (Whomsley et al 1993). The achiral substituted 1-(benzofuran-2-yl methyl) imidazoles (**7**, R = H) are much weaker inhibitors of the enzyme than the phenyl analogues (**6**) being only 3–7 times more potent than (±)-aminoglutethimide. It would seem that removal of the hydrophobic phenyl group radically reduces the overall binding of **7** (R = H).

It is unusual for high potency to lie in both enantiomers of a $P450_{Arom}$ inhibitor and the reduced stereoselectivity observed for the enantiomers of **6** was further explored for a series of chiral 1-(benzofuran-2-yl)-1-(1-*H*-imidazol-1-yl) alkanes (**7**) with the phenyl ring in **6** replaced by the less hydrophobic methyl and ethyl functions. The (±)-methyl-substituted compounds (**7**, $R = CH_3$) were 2·36–12·0 times more potent than (±)-aminoglutethimide and, as expected with an increase in hydrophobicity, the (±)-ethyl substituted compounds were 5·4–33 times more potent; the 5,7-dichloro compounds (**9** and **12**) were the most potent in each series.

The resolved dibenzoyl-D-(+)-tartrate salt of the (+) form of the methyl-substituted compound and its enantiomeric dibenzoyl-L-(−)-tartrate salt of the (−) form had stereoselective ratios (IC50 of the (−) form/IC50 of the (+) form) of 4·8 (**9**) and 12·6 (**10**), and increased potency compared with (±)-aminoglutethimide of 22·4- and 13·2-fold for the more active (+) isomer.

In the ethyl-substituted series the (−)-dibenzoyl-L-(−)-tartaric acid salts were the more active, the stereoselective ratios being 8·3 and 5·2 for (**12**) and (**13**), with both having 39-fold greater potency than (±)-aminoglutethimide.

The stereoselective inversion towards $P450_{Arom}$ for the ethyl-substituted series relative to the methyl series is reasonably established on the basis of the following arguments, in the absence of confirmatory absolute configuration data from X-ray crystallography which was unsuccessfully attempted on the two series and on the phenyl-substituted compounds (**6**). In the methyl-substituted series the (+)-dibenzoyl-D-(+)-tartaric acid

salts consist of (+) base and (+) acid and the (−)-dibenzoyl-L-(−)-tartaric acid salts of (−) base and (−) acid, defined as p+ (or p−) enantiomers, as shown by the near identical (but opposite) signs of specific rotation. In support of this the (+)-dibenzoyl-D-(+)-tartaric acid salt of **8** gave the (+) base on release with alkali. In the phenyl-substituted series p+ (or p−) enantiomers were obtained and the (−)-dibenzoyl-L-(−)-tartaric acid salt of the 4′-fluoro compound (**6**, R = H, R′ = 4′-F) gave the (−) base with alkali. Furthermore the (+)-dibenzoyl-D-(+)-tartaric acid salts of the 4′-methyl (**6**, R = H, R′ = 4′-CH_3) and 4-fluoro (**6**, R = H, R′ = 4′-F) compounds were the more potent isomers.

In the ethyl-substituted series the specific rotations of the enantiomeric salts are lower than those of the methyl- and phenyl- substituted series, average values for the (+)-dibenzoyl-D-(+)-tartaric acid salts being +41, +73·70 and +63·1°, respectively. However, the value of +41° is not sufficiently low for the [(−) base-(+)-dibenzoyl-D-(+)-tartaric acid salt] of the alternative pair of n+ (or n−) enantiomers i.e. [(+) base-(−)-dibenzoyl-L-(−)-tartaric acid] and [(−) base-(+)-dibenzoyl-D-(+)-tartaric acid] which would have accounted for the stereoselective inversion through inversion of configuration in the isolated forms.

This study demonstrates that within moderately potent inhibitors of $P450_{Arom}$ the high enantioselectivity previously encountered in inhibitors can be reduced so that both forms have activity although one form is favoured in the methyl and ethyl series (**7**) studied here. Stereoselectivity and potency usually go hand in hand but here for the moderately potent methyl and ethyl series (**7**) enantioselectivity is similar to that observed for the potent phenyl series (**6**). It might be that three-point attachment of an azole function to the Fe^{3+}-haem and two additional non-specific hydrophobic binding groups to the enzyme surface in the three series gives some scope for the interchange of the non-specific binding groups at the active site.

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