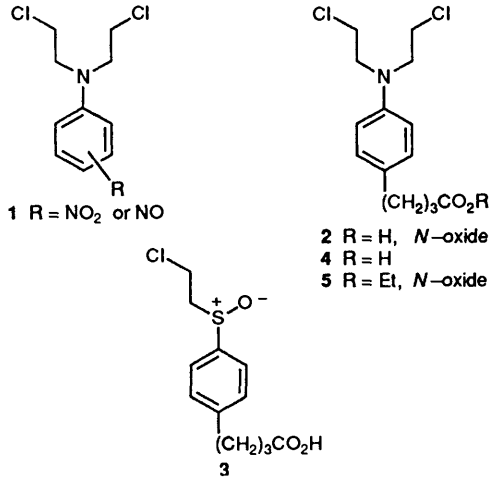
Synthesis of Novel *N*- and *S*-Mustards as Potential Pro-drugs Activated by Bioreductive Processes

John Mann and Leroy A. Shervington
Department of Chemistry, Reading University, Whiteknights, Reading, Berkshire RG6 2AD, UK

We describe the synthesis of 4-{*p*-[bis-(2-chloroethyl)amino]phenyl}butanoic acid *N*-oxide, which is the *N*-oxide of the anti-cancer drug chlorambucil, and 4-[*p*-(2(chloroethylsulphinyl)phenyl]-butanoic acid. The 3-nitro and 3,6-dinitro derivatives of chlorambucil have also been prepared.

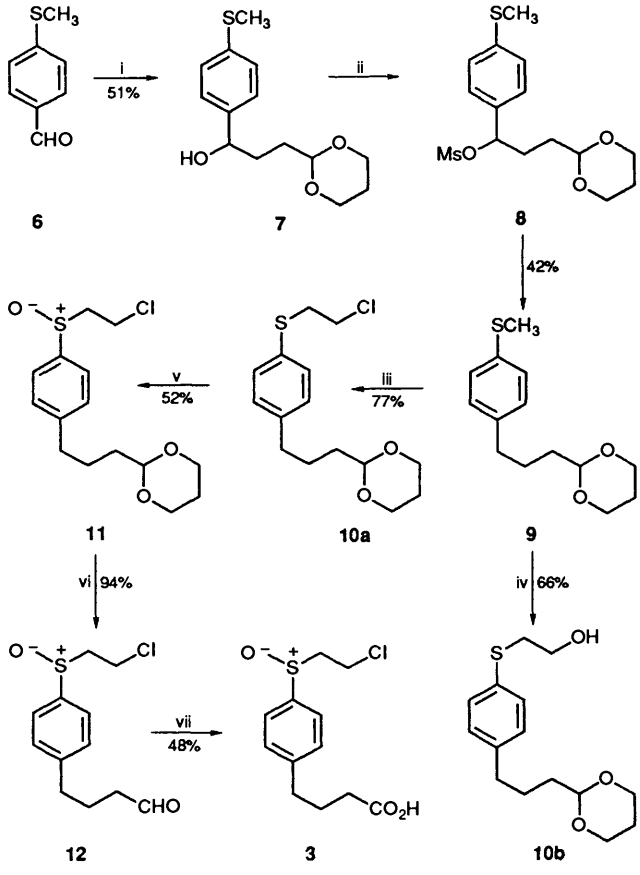
Once a tumour has reached a certain critical size, it often becomes refractory to chemotherapy. This is primarily due to restriction of the blood supply, with consequent oxygen deficiency; in fact these hypoxic cells often have reducing activity. One chemotherapeutic strategy that seeks to exploit this reductive potential involves the use of pro-drugs that are activated through bioreductive processes.[1] A good example of this approach is provided by the work of Denny *et al.*[2] who prepared nitro and nitroso derivatives of *N*,*N*-bis(2-chloroethyl)-anilines (nitro- and nitroso-arylmustards) **1**. Several of these relatively non-toxic compounds were converted into amino- and hydroxylamino-*N*-mustards with much enhanced cytotoxic properties following *in vivo* reduction.



Our initial efforts in this area have concentrated upon the synthesis of *N*- and *S*-mustard oxides, which we expected might be reduced to the corresponding *N*- and *S*-mustards within the tumour. We report here our syntheses of chlorambucil *N*-oxide **2** and the *S*-oxide analogue **3**. We chose to prepare analogues of chloroambucil because this is one of the most commonly used *N*-mustards, and is well-tolerated by patients. Like other mustards, it functions through alkylation of the *N*-7 of guanine residues on DNA, with subsequent reaction with other nucleophiles to form cross-linked DNA strands.[3,4]

Chloroambucil **4** was oxidised with peracetic acid to yield the *N*-oxide **2** directly (32% yield), and the corresponding ethyl ester **5** was prepared by esterification of chloroambucil followed by oxidation (50% overall). The ^{1}H NMR spectra of these *N*-oxides were of interest since the methylene hydrogens adjacent to the *N*-oxide were clearly non-equivalent, one set appearing as a triplet (*J* 6.5 Hz) at δ 4.0, and the other set appearing as a triplet (*J* 6.5 Hz) at δ 3.5. This is presumably due to restricted rotation about the N–C bonds.

A seven-stage synthesis of the sulphoxide **3** was developed commencing with 4-methylthiobenzaldehyde **6**, and this is shown in Scheme 1. Reaction of **6** with the Grignard reagent



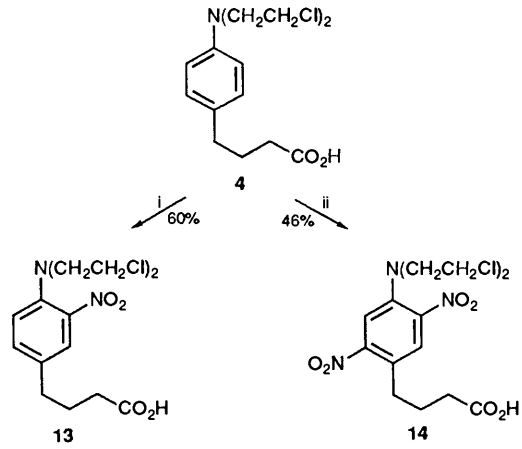
Scheme 1 *Reagents:* i, Mg/2-(2-bromoethyl)-1,3-dioxane in THF; ii, mesyl chloride/Et_3N then $LiAlH_4$; iii, MeSNa/HMPA then 1,2-dichloroethane; iv, MeSNa/HMPA then chloroethanol; v, $NaIO_4$ in MeOH(aq); vi, $H_3{}^+O$ in acetone; vii, $NaClO_2/Na_2H_2PO_4$ in *tert*-butyl alcohol and 2-methylbut-2-ene.

from 2-(2-bromoethyl)-1,3-dioxane provided the expected alcohol **7**, which was converted into the mesylate **8** prior to reduction with lithium aluminium hydride.[5] The overall yield of the resultant ketal **9** was *ca.* 25%. Conversion into the *S*-chloroethyl derivative **10a** was achieved in 77% yield by treatment of the ketal **9** with hot sodium thiomethoxide,[6] and reaction of the resultant thiophenoxide with 1,2-dichloroethane. The thiophenoxide could also be intercepted with chloroethanol to produce the *S*-hydroxyethyl derivative **10b** (66% yield), and the process seems to be of general utility for the *S*-functionalisation of thioanisoles.

The synthesis was completed by formation of the sulphoxide

11 (52%), dioxane hydrolysis, and oxidation with sodium chlorite to yield the desired analogue **3** (45% for the two steps).

Finally, the 3-nitro and 3,6-dinitro derivatives of chloroambucil **13** and **14** were prepared by treatment of chloroambucil with an excess of nitronium tetrafluoroborate [7] (Scheme 2). The



Scheme 2 *Reagents:* i, $NO_2^+BF_4^-/CH_3CN$ (2 equiv.); iii, $NO_2^+BF_4^-/CH_3CN$ (6 equiv.).

structure of mononitro derivative **13** was apparent from an NOE experiment in which there was a marked interaction between the benzylic hydrogens and two aryl hydrogens. Somewhat surprisingly, these two compounds had not been previously described, and proved to be the most interesting during biological evaluation.

Compounds **2**, **3**, **13** and **14** were assessed for their inhibitory activities on the growth of chinese hamster V79 cells maintained under an atmosphere of oxygen or nitrogen (to stimulate hypoxic conditions). All of the compounds were cytotoxic, but none of them had greater activity under hypoxic conditions. However, in further tests that employed a chlorambucil-resistant cell line (chinese-hampster ovary cells), the two nitro compounds **13** and **14** proved to have greater activity than chlorambucil, and this interesting result is being further investigated.

Experimental

Melting points were determined on a Kofler hot stage and are uncorrected. The IR spectra for solids were recorded as either a Nujol mull or a chloroform solution using a Perkin-Elmer 881 double beam grating spectrometer; ^1H NMR were recorded in deuteriated chloroform using Perkin-Elmer R34 (220 MHz) or Varian T60 (60 MHz) instruments, tetramethylsilane was used as internal standard, and *J*-values are given in Hz. ^{13}C NMR spectra were recorded on a JEOL FX-90Q FT (22.49 MHz) spectrometer. Mass spectra were recorded on a VG ZAB-Z high resolution spectrometer and micro-analysis were recorded on a CEC 240XA instrument. Solvents were distilled before use. Light petroleum refers to the fraction with b.p. 40–60 °C. Reactions were monitored, whenever possible, by TLC on silica gel plates (G_{254}). Column chromatography was performed using Crossfield Sorbsil C60 silica gel (40–60 μm).

4-{p-[*Bis*(2-*chloroethyl*)*amino*]*phenyl*}*butanoic Acid* N-*Oxide* **2**.—A solution of chlorambucil **4** (3 g, 9.90 mol dm^{-3}) in dichloromethane (30 cm^3) was treated with 40% (w/v) peracetic acid in acetic acid (10 cm^3), dropwise at 0 °C. The mixture was stirred at this temperature for 4 h. The dichloromethane and the excess of acetic acid were removed under reduced pressure at room temperature and the residue was dissolved in dichloromethane (100 cm^3). The solution was washed successively in water (4 × 60 cm^3), and dried over sodium sulphate. The crude product was chromatographed on a column of silica gel with ethyl acetate–light petroleum (4:3) as eluent to afford the *N*-oxide **2** as a light coloured oil (1.01 g, 32%) (Found: C, 52.2; H, 6.0; N, 4.3; Cl, 22.4. Calc. for $C_{14}H_{19}Cl_2NO_3$: C, 52.50; H, 6.00; N, 4.35; Cl, 22.15%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1710 (C=O), 2600–3500br (OH); δ_H(220 MHz; $CDCl_3$) 1.96 (2 H, m, $CH_2CH_2CO_2H$), 2.38 (2 H, t, *J* 7.5, CH_2CO_2H), 2.66 (2 H, t, *J* 7.5, $ArCH_2$), 3.49–4.03 [8 H, complex m, $N(CH_2CH_2Cl)_2$], 7.14 (4 H, 2 × d, *J* 8.8, C_6H_4) and 8.40 (1 H, br s, CO_2H); δ_C(22.49 MHz; $CDCl_3$) 26.2 ($CH_2CH_2CO_2H$), 33.2 and 34.2 ($CH_2CH_2CH_2$), 41.9 (2 × CH_2Cl), 72.9 (2 × H_2CN), 118.6 (aromatic, 2 × CH), 129.0 (aromatic, 2 × CH), 136.8 (aromatic, C–CH_2), 148.8 (aromatic, C–N) and 179.7 (CO_2H).

Ethyl 4-{p-[*Bis*(2-*chloroethyl*)*amino*]*phenyl*}*butanoate* N-*Oxide* **5**.—A mixture of chlorambucil (10 g, 33 mol dm^{-3}), ethanol (100 cm^3) and concentrated sulphuric acid (3 cm^3) was heated under reflux for 4 h. The ethanol was removed under reduced pressure and the residue was dissolved in dichloromethane (100 cm^3). The solution was washed successively with aqueous sodium hydrogen carbonate (3 × 50 cm^3) and water (3 × 50 cm^3), and dried over sodium sulphate. The dichloromethane was removed under reduced pressure to afford *ethyl* 4-{p-[*bis*(2-*chloroethyl*)*amino*]*phenyl*}*butanoate* as an oil (10.02 g, 92%); $\nu_{max}(CDCl_3)/cm^{-1}$ 1730 (C=O); δ_H(220 MHz; $CDCl_3$), 1.25 (3 H, t, CO_2CH_2Me), 1.92 (2 H, m, $CH_2CH_2CO_2Et$), 2.20 (2 H, t, *J* 7.5, CH_2CO_2Et), 2.55 (2 H, t, *J* 7.5, $ArCH_2$), 3.63 [8 H, s, $N(CH_2CH_2Cl)_2$], 4.09 (2 H, q, OCH_2Me), 6.54 [2 H, d, *J* 8, *ortho* to $N(CH_2CH_2Cl)_2$] and 7.01 [2 H, d, *J* 8, *meta* to $N(CH_2CH_2Cl)_2$].

A solution of the ethyl ester (4 g, 12.08 mmol dm^{-3}) in dichloromethane (20 cm^3) was treated with 40% (w/v) peracetic acid in acetic acid (12 cm^3), dropwise at 0 °C. The mixture was stirred at that temperature for 4 h, and then aqueous sodium hydrogen carbonate was added with vigorous stirring. The organic material was extracted with dichloromethane and chromatographed on a column of alumina using ethyl acetate–light petroleum (1:7) as eluent. This procedure afforded an oil that required further purification on a column of silica gel using dichloromethane–light petroleum (3:2) as eluent to afford the *N*-oxide **5** as a light yellow oil (2.29 g, 54%) (Found: C, 55.1; H, 6.95; N, 3.9; Cl, 20.15. $C_{16}H_{23}Cl_2NO_3$ requires C, 55.20; H, 6.65; N, 4.00; Cl, 20.35%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1740 (C=O); δ_H(220 MHz; $CDCl_3$) 1.25 (3 H, t, CO_2CH_2Me), 1.92 (2 H, m, $CH_2CH_2CO_2Et$), 2.30 (2 H, t, *J* 7.5, $CH_2CH_2CO_2Et$), 2.60 (2 H, t, *J* 7.5, $ArCH_2$), 3.48–4.05 [8 H, complex m, $N(CH_2CH_2Cl)_2$], 4.14 (2 H, q, CO_2CH_2Me) and 7.10 (4 H, s, C_6H_4); δ_C(22.49 MHz; $CDCl_3$) 14.2 (CH_2Me), 26.5 ($CH_2CH_2CH_2$), 33.6 and 34.4 ($CH_2CH_2CH_2$), 41.9 (2 × CH_2Cl), 60.2 (CO_2CH_2), 72.8 (2 × H_2CN), 118.2 (aromatic, 2 × CH), 129.0 (aromatic, 2 × CH), 137.0 (aromatic, C–CH_2), 148.7 (aromatic, C–N) and 173.4 (CO_2CH_2); *m/z* (EI) 347.

2-[3-*Hydroxy*-3-(p-*methylthiophenyl*)*propyl*]-1,3-*dioxane* **7**.—Dried magnesium ribbon (3.24 g, 135 mmol dm^{-3}) was treated with a solution of 2-(2-bromoethyl)-1,3-dioxane (25.6 g, 133 mmol dm^{-3}) in THF (80 cm^3) at 0 °C under argon, and after 10 min complete conversion into the Grignard reagent was achieved. A solution of 4-methylthiobenzaldehyde **6** (20 g, 133 mmol dm^{-3}) in THF (40 cm^3) was added to the Grignard reagent with sufficient rapidity to maintain the reaction mixture at gentle reflux, and the mixture was then stirred at 0 °C for 2 h. The mixture was poured into saturated aqueous ammonium chloride (100 cm^3), and the organic material was extracted with ether (4 × 60 cm^3), washed with water (4 × 60 cm^3) and dried over magnesium sulphate to give a crude product which was chromatographed on a silica column using ethyl acetate–light

petroleum (2:3) as eluent. This procedure afforded alcohol **7** as a white crystalline solid (18.25 g, 51%), m.p. 75–79 °C (Found: C, 62.65; H, 7.6; S, 11.95. $C_{12}H_{20}O_3S$ requires C, 62.65; H, 7.50; S, 11.95%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1055, 1090, 1110, 1140 and 1150 (C–O–C) and 3430br (O–H); δ_H(220 MHz; $CDCl_3$) 1.30–2.14 (6 H, complex m, OCH_2CH_2, CH_2CH, $ArCH_2CH_2$), 2.46 (3 H, s, SMe), 2.92 (1 H, br s, OH), 3.75 (2 H, t, *J* 13, $OCH_2CH_2CH_2O$, H_{ax}), 4.09 (2 H, dd, *J* 13 and 4, $OCH_2CH_2CH_2O$, H_{eq}), 4.64 (1 H, t, *J* 4.4, ArC*H*OH) and 7.22 (4 H, 2 × s, C_6H_4); δ_C(22.49 MHz; $CDCl_3$) 16.0 (SMe), 25.7 (OCH_2CH_2), 31.4 and 33.2 ($CHCH_2CH_2CH$), 66.9 (2 × OCH_2), 73.5 (CHOH), 101.9 (CH_2CH_2CH), 126.4 and 126.7 (aromatic, 4 × CH), 137.0 (aromatic, C–S) and 141.8 (aromatic C–CH_2); *m/z* (EI) 268.

2-[3-(p-*Methylthiophenyl*)*propyl*]-1,3-*dioxane* **9**.—Alcohol **7** (16.4 g, 61.2 mmol dm^{-3}) in dichloromethane (150 cm^3) was treated with triethylamine (9.3 g, 92 mmol dm^{-3}) in dichloromethane (50 cm^3) at 0 °C under argon and, after being stirred for 30 min, the mixture was stirred at room temperature for a further 30 min. The mixture was then cooled to −5 °C, methanesulphonyl chloride (8.41 g, 73.4 mmol dm^{-3}) in dichloromethane (50 cm^3) was added dropwise over 45 min and the mixture was stirred at −5 °C for 3 h. Lithium aluminium hydride (61.3 cm^3, 61.3 mmol dm^{-3}) in THF was added and the mixture was stirred at room temperature for 2 h. The mixture was cooled, poured into aqueous ammonium chloride (200 cm^3), and the organic material was extracted with ether (4 × 60 cm^3), washed with water (4 × 60 cm^3) and dried. The solvent was removed under reduced pressure to give an oil which was chromatographed on a column of silica using ethyl acetate–light petroleum (1:6) as eluent to afford methylsulphide **9** as an oil (6.50 g, 42%) (Found: C, 66.5; H, 8.15; S, 12.65. $C_{14}H_{20}O_2S$ requires C, 66.65; H, 8.00; S, 12.70%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1055, 1090, 1110, 1140 and 1150 (C–O–C); δ_H(220 MHz; $CDCl_3$) 1.25–2.17 (6 H, complex m, OCH_2CH_2, CH_2CH, $ArCH_2CH_2$), 2.48 (3 H, s, SMe), 2.61 (2 H, t, *J* 7, $ArCH_2$), 3.76 (2 H, t, *J* 13, $OCH_2CH_2CH_2O$, H_{ax}), 4.12 (2 H, dd, *J* 13 and 4, $OCH_2CH_2CH_2O$, H_{eq}), 4.54 (1 H, t, *J* 4.4, CH_2CH) and 7.09–7.24 (4 H, 2 × d, *J* 8.8 and 8.8, C_6H_4); δ_C(22.49 MHz; $CDCl_3$) 16.4 (SMe), 25.7 (2 × $CH_2CH_2CH_2$), 34.7 and 35.1 (CH_2CH_2-CH_2CH), 66.8 (2 × OCH_2), 102.1 (CH_2CH), 127.2 (aromatic, 2 × CH), 128.9 (aromatic, 2 × CH), 135.0 (aromatic, *C*–SMe) and 139.4 (aromatic, C–CH_2); *m/z* (EI) 252.

2-{3-[p-(2-*Hydroxyethylthio*)*phenyl*]*propyl*}-1,3-*dioxane* **10b**.—A solution of methylsulphide **9** (500 mg, 1.98 mmol dm^{-3}) in hexamethylphosphoramide (HMPA) (3 cm^3) was added dropwise with stirring to sodium thiomethoxide (555 mg, 7.92 mmol dm^{-3}) in HMPA (9 cm^3) and the mixture was heated to 100 °C under argon for 6 h, cooled to 50 °C, and then excess of chloroethanol was added rapidly. The mixture was stirred at 80 °C for a further 2 h, and then poured into water (100 cm^3). The organic material was extracted with ether (3 × 40 cm^3), washed with water (5 × 40 cm^3), dried and the solvent was removed under reduced pressure. The crude material was chromatographed on a column of silica with ethyl acetate–light petroleum as eluent to give the thioethanol **10b** as an oil (370 mg, 66%), $\nu_{max}(CHCl_3)/cm^{-1}$ 3450br (O–H); δ_H(200 MHz; $CDCl_3$) 1.26–2.18 (6 H, complex m, OCH_2CH_2, CH_2CH, $ArCH_2CH_2$), 2.30 (1 H, br s, OH), 2.61 (2 H, t, *J* 7, $ArCH_2$), 3.07 (2 H, t, *J* 6.6, CH_2OH), 3.68–3.84 (4 H, m, SCH_2 and $OCH_2CH_2CH_2O$, H_{ax}), 4.12 (2 H, dd, *J* 13 and 4, $OCH_2CH_2CH_2O$, H_{eq}), 4.53 (1 H, t, *J* 4.4, CH_2CH), 7.12 (2 H, d, *J* 8.8, *meta* to SCH_2CH_2OH) and 7.32 (2 H, d, *J* 8.8, *ortho* to SCH_2CH_2OH); δ_C(22.49 MHz; $CDCl_3$) 25.6 and 25.8 (2 × $CH_2CH_2CH_2$), 34.7 and 35.2 ($CH_2CH_2CH_2CH$), 37.8 (SCH_2), 60.2 (CH_2OH), 66.9 (2 × OCH_2), 102.0 (CH_2CH), 129.3 (aromatic, 2 × CH), 130.9 (aromatic, 2 × CH), 131.3 (aromatic, C–S) and 141.3 (aromatic, C–CH_2); *m/z* (EI) 282.

2-{3-[p-(2-*Chloroethylthio*)*phenyl*]*propyl*}-1,3-*dioxane* **10a**.—A solution of methylsulphide **9** (3.64 g, 14.4 mmol dm^{-3}) in HMPA (20 cm^3) was added dropwise with stirring to sodium thiomethoxide (1.21 g, 17.28 mmol dm^{-3}) in HMPA (20 cm^3) and the mixture was heated to 100 °C under argon for 6 h, cooled to 50 °C, and dichloromethane (14.3 cm^3, 144 mmol dm^{-3}) was rapidly added. The mixture was stirred at 80 °C for a further 2 h, and then poured into water (100 cm^3). The organic material was extracted with ether (3 × 60 cm^3) with care, washed with water (5 × 60 cm^3), dried over magnesium sulphate, and the solvent was removed under reduced pressure. The crude product was chromatographed on a column of silica with ethyl acetate–light petroleum (1:4) as eluent to give the chloroethylsulphide **10a** as an oil (3.36 g, 77%) (Found: C, 60.25; H, 7.15. $C_{15}H_{21}ClO_2S$ requires C, 59.90; H, 7.05%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1055, 1090, 1110, and 1140 and 1150 (C–O–C); δ_H(200 MHz; $CDCl_3$) 1.25–2.17 (6 H, complex m, OCH_2CH_2, CH_2CH, $ArCH_2CH_2$), 2.71 (2 H, t, *J* 7, $ArCH_2$), 3.16 (2 H, t, *J* 7, CH_2Cl), 3.58 (2 H, t, CH_2CH_2Cl), 3.75 (2 H, t, *J* 13, $OCH_2CH_2CH_2O$), 4.12 (2 H, dd, *J* 13 and 4, OCH_2CH_2-CH_2O), 4.53 (1 H, t, *J* 4.4, CH_2CH), 7.14 (2 H, d, *J* 8.8, *meta* to SCH_2CH_2Cl) and 7.32 (2 H, d, *J* 8.8, *ortho* to SCH_2CH_2Cl); δ_C(22.49 MHz; $CDCl_3$) 25.6 and 25.8 (2 × $CH_2CH_2CH_2$), 34.7 and 35.2 ($CH_2CH_2CH_2CH$), 36.7 (CH_2Cl), 42.4 (SCH_2), 66.9 (2 × OCH_2), 102.0 (CH_2CH), 129.3 (aromatic, 2 × CH), 130.8 (aromatic, C–S), 131.2 (aromatic, 2 × CH) and 141.7 (aromatic, C–CH_2); *m/z* (EI) 300.

2-{3-[p-(*Chloroethylsulphinyl*)*phenyl*]*propyl*}-1,3-*dioxane* **11**.—Chloroethylsulphide **10a** (3.26 g, 10.86 mmol dm^{-3}) in methanol (30 cm^3) was added dropwise with stirring to a solution of sodium periodate (2.57 g, 12 mmol dm^{-3}) in water (12 cm^3) and the mixture was stirred for 24 h at 0 °C. Dichloromethane (50 cm^3) was added and the sodium salt was filtered off and washed. The organic material was extracted with dichloromethane (4 × 50 cm^3), washed with water (4 × 40 cm^3), dried, and the solvent was removed under reduced pressure to give the crude product which was chromatographed on a column of silica using ethyl acetate as eluent to afford the sulphoxide **11** as a white crystalline solid (1.78 g, 52%), m.p. 49–51 °C (Found: C, 56.85; H, 6.7; Cl, 11.2; S, 9.85. $C_{15}H_{21}ClO_3S$ requires C, 56.85; H, 6.70; Cl, 11.20; S, 10.10%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1060 (S=O), 1090, 1110, 1140 and 1150 (C–O–C); δ_H(220 MHz; $CDCl_3$) 1.25–2.16 (6 H, complex m, OCH_2CH_2, CH_2CH, $ArCH_2CH_2$), 2.70 (2 H, t, *J* 7, $ArCH_2$), 3.14 (2 H, t, *J* 6, CH_2Cl), 3.66 (1 H, m, O=S–CH_aH_b), 3.75 (2 H, t, *J* 13, $OCH_2CH_2CH_2O$), 3.95 (1 H, m, O=S–CH_aH_b), 4.09 (2 H, dd, *J* 13 and 4, $OCH_2CH_2CH_2O$), 4.54 (1 H, t, *J* 4.4, CH_2CH), 7.35 (2 H, d, *J* 8.8, *meta* to O=S–CH_2) and 7.52 (2 H, d, *J* 8.8, *ortho* to O=S–CH_2); δ_C(22.49 MHz; $CDCl_3$) 25.4 and 25.8 (2 × CH_2-CH_2CH_2), 34.6 and 35.5 ($CH_2CH_2CH_2CH$), 36.7 (CH_2Cl), 59.3 (O=S–CH_2), 66.8 (2 × OCH_2), 101.9 (CH_2CH), 123.9 (aromatic, 2 × CH), 129.6 (aromatic, 2 × CH), 139.8 (aromatic, C–S=O) and 146.3 (aromatic, C–CH_2); *m/z* (EI) 316.

4-[p-(2-*Chloroethylsulphinyl*)*phenyl*]*butanal* **12**.—A mixture of chloroethylsulphoxide **11** (1.56 g, 4.93 mmol dm^{-3}) and hydrochloric acid (2 mol dm^{-3}; 15 cm^3) in acetone (30 cm^3), was heated to mild reflux for 2 d and then poured into water (50 cm^3). The organic material was extracted with dichloromethane (4 × 40 cm^3), washed with water (4 × 40 cm^3), dried over sodium sulphate, and the solvent was removed under reduced pressure to give the crude product which was chromatographed on a column of silica with ethyl acetate as eluent to afford the

aldehyde **12** as an oil (1.20 g, 94%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1740 (C=O); δ_H(220 MHz; $CDCl_3$) 1.98 (2 H, m, CH_2CH_2CHO), 2.49 (2 H, t, *J* 7, CH_2CHO), 2.73 (2 H, t, *J* 7, $ArCH_2$), 3.15 (2 H, m, *J* 6, CH_2Cl), 3.69 (1 H, m, $O{=}S{-}CH_aH_b$), 3.96 (1 H, m, $O{=}S{-}CH_aH_b$), 7.36 (2 H, d, *J* 8.8, *meta* to $O{=}S{-}CH_2$), 7.56 (2 H, d, *J* 8.8, *ortho* to $O{=}S{-}CH_2$) and 9.76 (1 H, s, CHO); δ_C(22.49 MHz; $CDCl_3$) 23.3 ($CH_2CH_2CH_2$), 34.8 ($ArCH_2$), 36.7 (CH_2Cl), 42.9 (CH_2CHO), 59.3 ($O{=}S{-}CH_2$), 124.1 (aromatic, 2 × CH), 129.6 (aromatic, 2 × CH), 140.3 (aromatic, C–S=O), 145.3 (aromatic, $C{-}CH_2$) and 201.6 (CHO); *m/z* (EI) 258 (Found: M^+, 258.0479. $C_{12}H_{15}ClO_2S$ requires *M*, 258.0478).

4-[p-(2-*Chloroethylsulphinyl*)*phenyl*]*butanoic Acid* **3**.—A solution of sodium chlorite (3.78 g, 41.76 mmol dm^{-3}) and sodium dihydrogen phosphate (3.62 g, 23.2 mmol dm^{-3}) in water (20 cm^3) was added dropwise with stirring to a mixture of aldehyde **12** (1.20 g, 4.6 mmol dm^{-3}), and 2-methylbut-2-ene (14 cm^3) in *tert*-butyl alcohol (50 cm^3). After the mixture had been stirred for 5 h at room temperature, water (60 cm^3) was added and the organic material was extracted with dichloromethane (3 × 40 cm^3), washed with water (3 × 40 cm^3) and dried over sodium sulphate. The crude product was chromatographed on a column of silica with ethyl acetate–light petroleum (2:1) as eluent to give the acid **3** as a clear oil (615 mg, 48%); $\nu_{max}(CHCl_3)$ 1060 (S=O), 1718 (C=O) and 2800–3500br (OH); δ_H(220 MHz; $CDCl_3$) 1.99 (2 H, m, $CH_2CH_2CO_2H$), 2.39 (2 H, t, *J* 7, CH_2CO_2H), 2.76 (2 H, t, *J* 7, $ArCH_2$), 3.21 (2 H, m, CH_2Cl), 3.68 (1 H, m, $O{=}S{-}CH_aH_b$), 3.96 (1 H, m, $O{=}S{-}CH_aH_b$), 7.38 (2 H, d, *J* 8.8, *meta* to $O{=}SCH_2$), 7.58 (2 H, d, *J* 8.8, *ortho* to $O{=}SCH_2$) and 8.90 (1 H, br, s, CO_2H); δ_C(22.49 MHz; $CDCl_3$) 26.0 ($CH_2CH_2CH_2$), 32.2 and 34.8 ($CH_2CH_2CH_2$), 36.7 (CH_2Cl), 59.1 ($CH_2S{=}O$), 124.2 (aromatic, 2 × CH), 129.7 (aromatic, 2 × CH), 139.6 (aromatic, C–S=O), 145.6 (aromatic, $C{-}CH_2$) and 177.7 (CO_2); *m/z* (EI) 274.

Preparation of the 3-*Nitrochlorambucil* **13**.—A solution of nitronium tetrafluoroborate (535 mg, 4.03 mmol dm^{-3}) in acetonitrile (20 cm^3) was stirred at 0 °C under argon, and after 15 min chlorambucil (614 mg, 2.02 mmol dm^{-3}) in acetonitrile (20 cm^3) was added dropwise over 15 min. After being stirred for an additional 30 min at 0 °C, the mixture was stirred at room temperature for 1 h and then poured into an excess of water. The organic material was extracted with dichloromethane and chromatographed on a column of silica gel with ethyl acetate–light petroleum (1:1) as eluent to give **13** as an oil (420 mg, 60%) (Found: C, 47.9; H, 5.25; N, 8.0; Cl, 20.05. $C_{14}H_{18}N_2Cl_2O_4$ requires C, 48.15; H, 5.20; N, 8.00; Cl, 20.30%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1720 (C=O) and 2800–3500br (OH); δ_H(220 MHz; $CDCl_3$) 2.07 (2 H, m, $CH_2CH_2CO_2H$), 2.51 (2 H, t, *J* 7.5, CH_2CO_2H), 2.80 (2 H, t, *J* 7.5, $ArCH_2$), 3.59 [8 H, 2 × s, $N(CH_2CH_2Cl)_2$], 7.30 (1 H, d, *J* 8.2, *meta* to NO_2, $C_6H_3NO_2$), 7.33 (1 H, dd, *J* 8.2 and 2.0, *para* to NO_2, $C_6H_3NO_2$) and 7.51 (1 H, d, *J* 2.0, *ortho* to NO_2, $C_6H_3NO_2$); δ_C(22.48; $CDCl_3$) 25.8 ($CH_2CH_2CO_2H$), 33.1 and 33.9 ($CH_2CH_2CH_2$), 41.5 (2 × CH_2Cl), 56.0 (2 × H_2CN), 124.8 [CH, *ortho* to $N(CH_2CH_2Cl)_2$], 126.8 (CH, *ortho* to NO_2), 133.3 (CH, *para* to NO_2), 138.5 ($C{-}CH_2$), 141.0 ($C{-}NO_2$), 146.7 (C–N) and 179.0 (CO_2H); *m/z* (EI) 348 (Found: M^+, 348.0645. $C_{14}H_{18}Cl_2N_2O_4$ requires *M*, 348.0655).

Preparation of the 3,6-*Dinitrochlorambucil* **14**.—A solution of nitronium tetrafluoroborate (2.62 g, 19.7 mmol dm^{-3}) in acetonitrile (30 cm^3) was stirred at 0 °C under argon and after 15 min chlorambucil (1.0 g, 3.29 mmol dm^{-3}) in acetonitrile (30 cm^3) was added dropwise over a period of 15 min. After being stirred for an additional 30 min, the mixture was stirred at room temperature for 1 h and then poured into an excess of water. The organic material was extracted with dichloromethane and chromatographed on a column of silica gel with ethyl acetate–light petroleum (2:1) as eluent to afford **14** as an oil (600 mg, 46%) (Found: C, 42.7; H, 4.25; N, 10.5; Cl, 17.8. $C_{14}H_{12}Cl_2N_3O_6$ requires C, 42.66; H, 4.35; N, 10.66; Cl, 17.99%); $\nu_{max}(CHCl_3/cm^{-1}$ 1718 (C=O) and 2800–3500br (OH); δ_H(220 MHz; $CDCl_3$) 2.01 (2 H, m, $CH_2CH_2CO_2H$), 2.49 (2 H, t, *J* 7.5, CH_2CO_2H), 2.92 (2 H, t, *J* 7.5, $ArCH_2$), 3.59 [8 H, 2 × s, 2 × (CH_2CH_2Cl)], 7.70 [1 H, s, *ortho* to $N(CH_2CH_2Cl)_2$, $C_6H_2(NO_2)_2$], 7.85 [1 H, s, *meta* to $N(CH_2CH_2Cl)_2$, $C_6H_2(NO_2)_2$] and 9.40–10.60 (1 H, br s, CO_2H); δ_C(22.48; $CDCl_3$) 25.8 ($CH_2CH_2CO_2H$), 31.1 and 33.3 ($CH_2CH_2CH_2$), 41.6 (2 × CH_2Cl), 54.7 (2 × H_2CN), 121.2 [CH, *ortho* to $N(CH_2CH_2Cl)_2$], 128.7 [CH, *meta* to $N(CH_2CH_2Cl)_2$], 130.8 ($C{-}CH_2$), 141.8 [$C{-}NO_2$, *ortho* to $N(CH_2CH_2Cl)_2$], 146.0 (C–N), 151.1 [$C{-}NO_2$, *meta* to $N(CH_2CH_2Cl)_2$] and 179.0 (CO_2H) (Found: M^+, 393.0526. $C_{14}H_{17}Cl_2N_3O_6$ requires *M*, 393.0505).

Acknowledgements

We thank the British Technology Group for a research fellowship to L. A. Shervington, the MRC Radiobiology Unit for preliminary biological results, and Oliver Howarth for high field NMR studies.

References

1 H. W. Moore, *Science*, 1977, **197**, 527.
2 B. D. Palmer, W. R. Wilson, S. M. Pullen and W. A. Denny, *J. Med. Chem.*, 1990, **33**, 112.
3 W. B. Pratt and R. W. Ruddon, *The Anticancer Drugs*, 1979, Oxford University Press, New York.
4 K. A. Kennedy, *Anti-Cancer Drug Design*, 1987, **2**, 181.
5 G. Büchi, W. Hofheinz and J. W. Paukstelis, *J. Am. Chem. Soc.*, 1969, **91**, 6473.
6 L. Testaferri, M. Tingoli and M. Tiecco, *Tetrahedron Lett.*, 1980, **21**, 3099.
7 S. J. Kuhn and G. A. Olah, *J. Am. Chem. Soc.*, 1961, **83**, 6564.

Paper 1/03522C
Received 11*th July* 1991
Accepted 21*st August* 1991