Applications of Biotransformations in Organic Synthesis: Preparation of Enantiomerically Pure Esters of *cis*- and *trans*-Epoxysuccinic Acid

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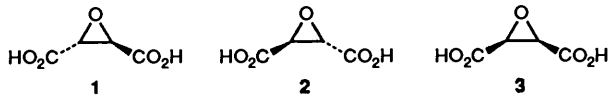
Chemical and enzymatic procedures have been investigated for the resolution of esters of *trans*-epoxysuccinic acid (*trans*-oxirane-2,3-dicarboxylic acid). A successful resolution was achieved by transesterification of the diethyl ester against heptanol catalysed by the lipase from *Rhizopus javanicus*. Asymmetrization of *cis*-epoxysuccinic acid was achieved by enantioselective and regiospecific hydrolysis of the racemic diethyl ester of 2-(toluene-*p*-sulfonyloxy)tartaric acid [(2*RS*,3*RS*)-2-hydroxy-3-(4-methylphenyl)sulfonyloxybutanoic acid], followed by esterification with 2-methylpropanol and oxirane formation.

The epoxysuccinic acids (oxirane-2,3-dicarboxylic acids) have attracted attention from several points of view. The (−)-(2*R*, 3*R*) acid **1** has been isolated from a range of microorganisms including *Penicillium vermiculatum*, *Paecilomyces varioti*,[1] *Aspergillus fumigatus*[2-4] and *Lentinus degener*.[5] The (+)-(2*S*,3*S*) acid **2** is a constituent of E-64, a specific inhibitor of thiol proteases.[6-8] The *trans*-acid and its esters have been applied in the synthesis, for example of a cysteine protease inhibitor,[9] penems and carbapanems,[10,11] carumonam[12] and other β-lactams,[13] malic acid and esters,[14] (2*S*,3*S*)-aziridine-2,3-dicarboxylic acid,[15] β-hydroxy-α-amino acids,[16] the insect pheromone multistriatin[17] and (+)-phyllanthocin, the aglycone of the anti-leukaemic glycoside (+)-phyllanthoside.[18]

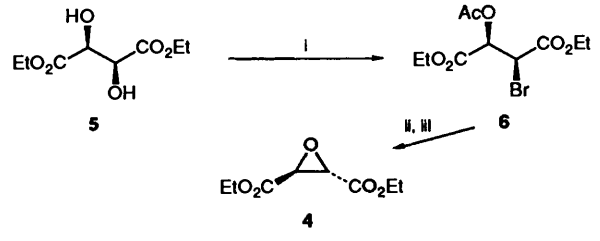


1 **2** **3**

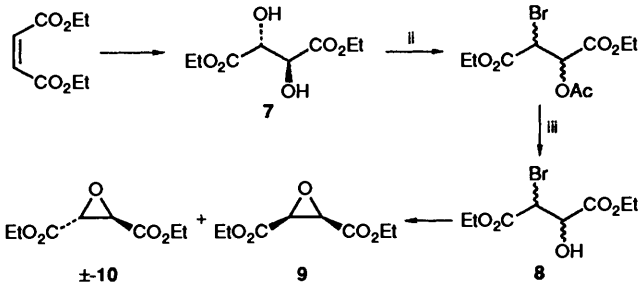
By contrast, the *cis*-acid, **3**, has not been used in synthesis, clearly because its symmetry is too high for it to be useful for the synthesis of chiral compounds.

trans-Epoxysuccinic acid has been resolved by classical crystallisation of diastereoisomeric salts.[19,20] However, the obvious precursors of optically active epoxysuccinates are the tartaric acids. Mori and Iwasawa[17] developed a double inversion procedure for the conversion of L-(+)-tartaric acids diesters (*i.e.* the 2*R*,3*R*-isomer) to corresponding diesters of L-(−)-epoxysuccinic acid (2*R*,3*R*). However, we describe below a direct enzyme-catalysed transesterification for the kinetic resolution of diethyl epoxysuccinate. We also describe a procedure for producing chiral unsymmetrical diesters of *cis*-epoxysuccinic acid from tartaric acids.

For the preparation of the enantiomers of esters of *trans*-epoxysuccinic acid, optically active standards were needed. Accordingly, the diethyl ester of the (2*S*,3*S*)-epoxysuccinate (**4**, Scheme 1) was prepared from diethyl (2*S*,3*S*)-tartrate **5** *via* the derivative **6** by a modification of the published method.[17] The



Scheme 1 *Reagents:* i, HBr, HOAc; ii, HBr, EtOH; iii, NaOEt



Scheme 2 *Reagents:* i, OsO_4, *N*-methylmorpholine *N*-oxide, 79%; ii, AcOH, HBr, 30%; iii, HBr, EtOH, 6%; iv, NaOEt, 54%

product had $[\alpha]_D^{27}$ 108.4 (lit.,[17] [α] = 105.5). To confirm the optical purity of the product it was examined by ^1H NMR spectroscopy (400 MHz) in the presence of (*S*)-(+)-2,2,2-trifluoro-1-(9-anthryl)ethanol. (This chiral solvating agent was used throughout this investigation and is referred to below as the 'chiral solvating agent'). Using the racemate, baseline splitting was observed for the signal attributable to the methine proton. When the product from diethyl (2*S*,3*S*)-tartrate was examined in the same way, only the downfield signal was observed, from which it was concluded that the product had ≥98% ee.

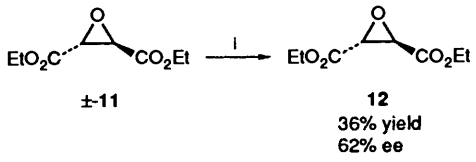
The procedure of Mori[7] (Scheme 1) was applied to diethyl *meso*-tartrate **7**, prepared from diethyl maleate. However, the bromohydrin **8** was obtained in very poor yield and gave on ring closure a mixture of *cis*- and *trans*-epoxy esters **9** and **10** in a ratio of 54:45 (Scheme 2).

For the enzymatic resolution of diethyl *trans*-epoxysuccinate **11**, the effectiveness of a hydrolytic procedure was questionable, as the ester was found to undergo spontaneous hydrolysis at 30 °C in phosphate buffer at pH 7. The rate of hydrolysis was rather variable, but generally, about 25% of the ester groups were hydrolysed within four hours at room temperature. Although this militated against the effective use of a hydrolytic system, an experiment was carried out with the lipase from *Rhizopus javanicus*, chosen because of its effectiveness in the transesterification reaction described below. In an isooctane–buffer (pH 7) medium (4:1, v/v), hydrolysis was allowed to proceed to 26% of all ester groups using an autotitrator. Unchanged (2*R*,3*R*)-diester **12** was recovered in 36% yield and with 62% ee (Scheme 3). The ratio of isooctane to water was crucial. With a ratio of 4:1, recovery of unchanged ester was reasonable; with a ratio of 1:4, recovery was much lower, owing, presumably to more favourable partitioning of the substrate into the aqueous phase with a correspondingly greater degree of spontaneous hydrolysis.

Table 1 Screening for stereocontrol in the enzyme-catalysed transesterification of diethyl *trans*-epoxysuccinate ±-**11**

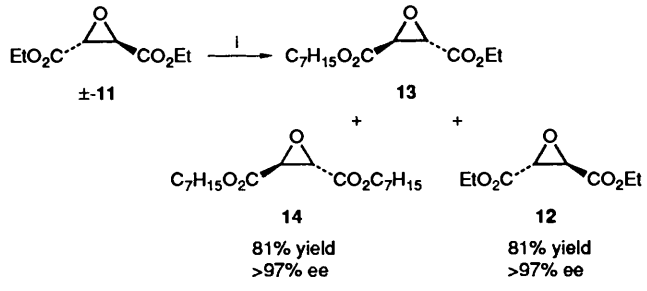
				Product ratio			% ee		
Enzyme[a]	Alcohol[b]	*T*/°C	*t*/d	**12**	**13**	**14**	**12**	**13**	**14**
CCL	A	RT	3	53	47	—	28	43	—
CCL	A	40	22	—	49	51	—	—	7
PPL	A	40	26	60	40	—	0	—	—
LP	A	50	9	—	47	53	—	21	21
LAP6	A	40	37	40	60	—	8	14	—
CCL	B	55	2	45	55	—	21	—	—
LF-AP15	A	40	48	25	38	37	≥97	—	≥97

[a] CCL = lipase from *Candida cylindracea*; PPL = porcine pancreatic lipase; LP = lipase LP from *Pseudomonas fluorescens*; LAP6 = lipase from *Aspergillus niger*; LF-AP15 = lipase from *Rhizopus javanicus*. [b] A = heptanol; B = 2-methylpropan-1-ol.



Scheme 3 *Reagent:* lipase LF-AP15, pH 7

Transesterification studies led to a much more favourable result. The system chosen was lipase–isooctane–heptanol or 2-methylpropan-1-ol. Five enzymes were investigated. The products were found to be unchanged diethyl ester **12**, the mixed ethyl heptyl diester **13** and the diheptyl ester **14** (Scheme 4). The



Scheme 4 *Reagent:* i, lipase LF-AP15, pH 7

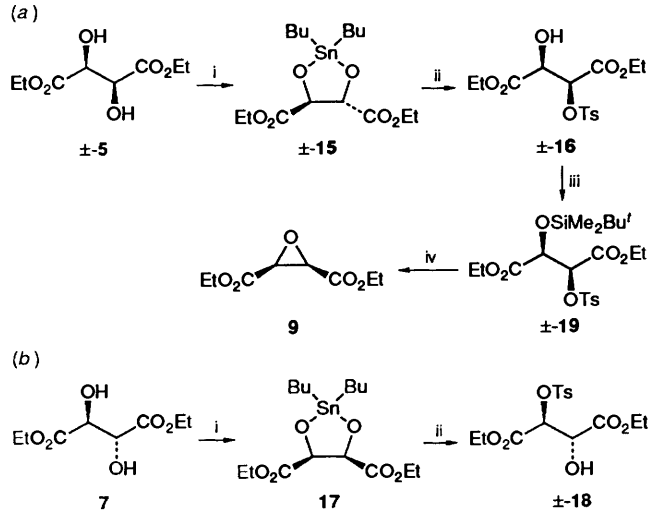
lipases from *Candida cylindracea*, porcine pancreas, *Pseudomonas fluorescens* and *Aspergillus niger* gave unsatisfactory results (Table 1). However, transesterification with the lipase from *Rhizopus javanicus* proceeded with high selectivity (Table 1). All of the enzymes, except that from *Aspergillus niger* catalysed transesterification preferentially of the (2*S*,3*S*)-diester.

Although the diethyl and diheptyl esters obtained using the lipase from *R. javanicus* were optically pure, reaction times were long. However, the rate of reaction was affected by the amount of water present. In addition, the use of an inert support (Biofix E) significantly increased the rate of transesterification. These factors were not investigated systematically, but it is clear that the time needed for complete transesterification could be significantly reduced.

In order to assign absolute configurations using the NMR/chiral solvating agent method, the transesterification of (2*S*,3*S*)-epoxysuccinate was carried out. Examination of the ^{1}H NMR spectrum of the diheptyl ester produced in the presence of the CSA, and re-examination after the addition of racemic ester, showed that the downfield signal attributable to the proton attached to the oxirane ring could be assigned to the (2*S*,3*S*)-enantiomer.

The obvious approach to chiral derivatives of *cis*-epoxysuccinic acid was by application of the '*meso*-trick' in which discrimination is effected between chiral centres in *meso* compounds. However, as with the *trans*-isomer, the success of a hydrolytic approach was rendered uncertain by the possibility of spontaneous hydrolysis. Thus 50% of the ester groups of diethyl *cis*-epoxysuccinate were hydrolysed in 12.5 hours at 37 °C. This propensity towards spontaneous hydrolysis no doubt explains the poor results obtained previously in enzymatic hydrolysis of *cis*-epoxysuccinic acid esters.[10,11,21] Only if a system could be found in which the catalysed reaction occurred at a much greater rate than the uncatalysed reaction could success be expected in a kinetic resolution. However, when (±)-diethyl *cis*-epoxsuccinate was hydrolysed by pig liver esterase at pH 7 and the reaction was stopped after 50% hydrolysis, the isolated ethyl hydrogen *cis*-epoxysuccinate had an ee of 21% as determined by ^{1}H NMR spectroscopy in the presence of (*R*)-(+)-α-methylbenzylamine. Accordingly, the hydrolytic approach was abandoned. Instead, a quite different strategy was adopted, based on the wide substrate tolerance of α-chymotrypsin.[22] The approach devised is illustrated in Scheme 5. It is based on the enantio- and regio-selective hydrolysis of a monosubstituted derivative of racemic diethyl tartrate.

Before the strategy of Scheme 5 could be realised in practice, some synthetic problems needed to be resolved. The first of these was the formation of a monotosylate of diethyl tartrate. Treatment of the diethyl ester **5** with an equimolar amount of toluene-*p*-sulfonyl chloride in pyridine–benzene led to a mixture of the unsaturated ester, the ditosyl derivative and, in 40% yield, the required monotosylate. Use of a lower temperature for the reaction and slow addition of the reagents led to no significant improvement and the desired product could only



Scheme 5 *Reagents:* i, Bu_2SnO; ii, TsCl, Et_4NCl; iii, Bu^tMe_2SiCl; iv, Bu_4NF

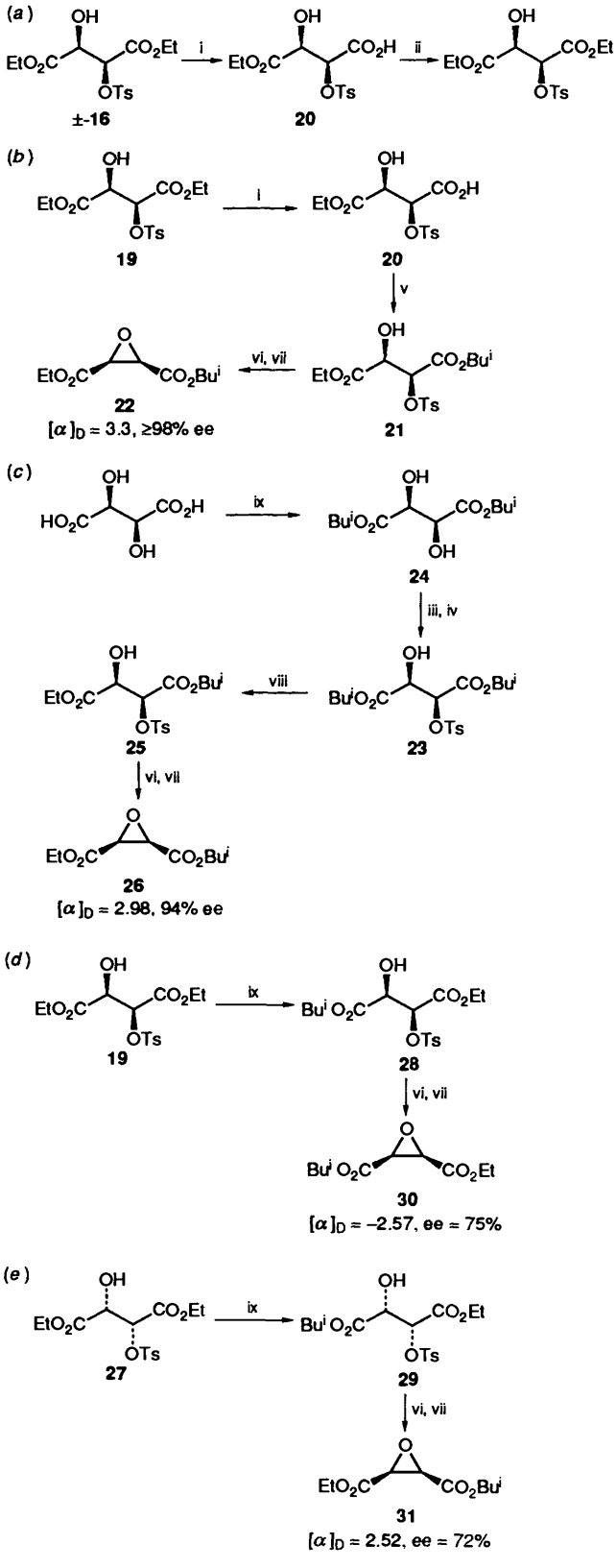
be obtained by tedious chromatographic separation. However, when the reaction was carried out on the cyclic stannylene **15** formed in quantitative yield from diethyl tartrate **5** and in dibutyltin oxide,[23] the required monotosylate **16** was formed cleanly and in 84% isolated yield (Scheme 5*a*). Similarly, the monotosylate **18** of diethyl *meso*-tartrate **7** was formed cleanly from the stannylene **17** [Scheme 5(*b*)].

The second problem concerned control of the stereoselectivity of ring closure of the monotosylate to the epoxide. When this was attempted under the usual conditions (NaOEt, EtOH) a mixture of *cis*- and *trans*-epoxides was obtained in a ratio of approximately 4:1 under various conditions, although the pure *cis*-ester could be obtained by distillation. This mixture presumably arises owing to epimerisation at the α-tosyl methine centre by reversible deprotonation. This problem was circumvented by a sequence of silylation and fluoride-mediated desilylation of the derivative **19** to generate the required alkoxide ion under essentially neutral conditions. The silylation proceeded in quantitative yield and ring closure in 87% yield based on isolated product **9** (Scheme 5).

Ethoxide-mediated ring closure of the monotosylate **18** gave the *trans*-epoxide **11** in 84% yield.

The monotosyl derivative **16** [Scheme 6(*a*)] was hydrolysed using α-chymotrypsin at pH 7.0 using an autotitrator and pH stat. Hydrolysis was regioselective; hydrolysis only of the ester group adjacent to the tosyl function was observed. The products were isolated and the hydrolysed product was reconverted into the diethyl ester using dicyclohexylcarbodiimide and hydroxybenzotriazole (Scheme 6*a*). Optical purities were determined by NMR spectroscopy using the chiral solvating agent. Surprisingly, splitting of the signal of the methyl group attached to the aromatic ring was marked, an effect possibly attributable to the formation of a charge transfer complex. This signal was conveniently used to measure enantiomeric purity. By using an authentic sample of the monotosylate of diethyl (2*S*,3*S*)-tartrate, the downfield and upfield components of the split signals were assigned to the (2*R*,3*R*)- and (2*S*,3*S*)-enantiomers respectively. From the data obtained, the enantiomeric ratio *E*[24] was found to be 8.8. This indicated that optically pure unhydrolysed (2*R*,3*R*) starting diester could be obtained if hydrolysis were allowed to proceed until approximately 40% of total ester groups had been hydrolysed (85% of the susceptible ester group).[23] In practice, after only 31% hydrolysis, the (2*R*,3*R*)-diester could be obtained in optically pure (>98% ee) form, which would correspond to an *E* value of ~15. The derivation of enantiomeric ratios, *E*, is based on the assumption of simple Michaelis–Menten kinetics with an irreversible final step and on the assumption that the kinetics do not change over the course of a reaction even though the conditions may vary continuously from beginning to end. That there should be a small deviation from a constant value is therefore not entirely unexpected.

Proof that hydrolysis had occurred at the ester function adjacent to the tosyl group was obtained by examining the product by ^{1}H NMR spectroscopy in the presence of 'proton sponge' (*N*,*N*,*N*′,*N*′-tetramethylnaphthalene-1,8-diamine). The signal (δ 5.40) attributable to the methine proton at the tosylated centre, was shifted upfield by 0.2 ppm; the other methine proton signal was not shifted. Based on now well-established procedures for amplification of optical activity, the optical purity of the hydrolysis product could be raised to any desired level by re-esterification and repetition of the hydrolysis.[24] Accordingly, to demonstrate the formation of chiral diesters of *cis*-epoxysuccinic acid, hydrolyses were carried out on the monotosyl derivative of diethyl (2*S*,3*S*)-tartrate [Scheme 6(*b*)]. On a 4 g scale, the diester **19** was hydrolysed in 92% yield to the mono acid **20**. Esterification with 2-methylpropanol gave the diester **21** in 48% yield after chromatographic purification. Silylation (quantitative) was followed by fluoride-promoted ring closure to give the chiral epoxy ester **22** in 81% yield. The product had $[\alpha]_D^{24}$ +3.33 (≥98% ee by NMR spectroscopy using the chiral solvating agent) [Scheme 6(*b*)].



Scheme 6 *Reagents:* i, α-chymotrypsin; ii, EtOH, DCCI, HOBT; iii, Bu_2SnO; iv, TsCl, Et_4NCl; v, Bu^iOH, DCCI, HOBT; vi, Bu^tMe_2SiCl; vii, Bu_4NF; viii, EtOH, TsOH; ix, Bu^iOH, TsOH

For chiral analysis, samples of both enantiomers of the chiral epoxide (as **22**) was required. The most convenient way to obtain these was by transesterification. This was carried out on the diisobutyl ester **23** of the monotosyl derivative of (2*S*,3*S*)-tartaric acid prepared from the diisobutyl ester **24** in the usual way [Scheme 6(*c*)]. Toluene-*p*-sulfonic acid-catalysed transesterification against ethanol gave the mixed diester **25** in 58% yield. This mixture was converted into the epoxy diester **26** $[\alpha]_D^{22}$ 2.98 (94% ee). The formation of this compound indicated that transesterification had occurred selectively at the ester group distal to the tosyl group [Scheme 6(*c*)]. The experiment was repeated using the monotosyl derivatives of diethyl D- and L-tartrate (**19** and **27** respectively) which were transesterified against isobutyl alcohol [Scheme 6(*d*, *e*)]. The products, **28** and **29** were converted into the corresponding epoxy diesters **30** and **31** respectively which had optical purities of 75 and 72%, respectively, indicating a lower selectivity for transesterification at the ester group distal to the tosyl function. Chiral analysis by ^{1}H NMR spectroscopy using the chiral solvating agent was carried out. Baseline splitting of all signals attributable to the protons of the ester groups was observed.

The present results provide a route to chiral esters of *cis*-epoxysuccinic acid from either racemic tartaric acid or from a single enantiomer. In spite of the marked differences between the diester **16** [Scheme 6(*a*)] and normal substrates of α-chymotrypsin, it is clear that the enzyme recognises the tosylate group as the P_2 unit of the peptide mimic,[25] and this directs the specificity of hydrolysis towards the adjacent ester function. The distal ester function mimics the hydrophobic side chain of the natural P_1 units (*e.g.* phenylalanine) and presumably binds in the hydrophobic pocket, completing the analogy with a phenylalanine, tyrosine or tryptophan peptide unit.

Recently, conversion of *meso*-tartaric acid into chiral building blocks has been achieved by an enzymatic route employing porcine pancreatic lipase.[26] The present route to chiral esters of *cis*-epoxysuccinic acid extends the range of available chiral and optically pure esters based on a *meso*-diacid derived from tartaric acid.

Experimental

Enzymes were obtained from the following sources: pig liver esterase (130 U mg^{-1}), Boehringer; porcine pancreatic lipase (Type II, 35–70 U mg^{-1}), Sigma; *Candida cylindracea* lipase (1,400–2,800 U mg^{-1}), lipase LAP 6 (from *Aspergillus niger*, 60 U mg^{-1}), lipase LF-AP 15 (from *Rhizopus javanicus* 150 U mg^{-1}) and lipase LP (from *Pseudomonas fluorescens*, 30 U mg^{-1}) from Amano. ^{1}H NMR spectra were determined at 400 MHz on a Bruker WH400 spectrometer or at 220 MHz on a Perkin-Elmer R34 spectrometer. Coupling constants are recorded in Hz. ^{13}C NMR spectra (proton-decoupled) were determined at 100.62 MHz on a Bruker WH400 NMR spectrometer for solutions in $CDCl_3$. *J* values are given in Hz. IR spectra were determined on a Perkin-Elmer 580-B spectrometer. Mass spectra were determined on a Kratos MS80 mass spectrometer. Optical rotations were determined on an AA-1000 polarimeter (Optical Activity Ltd.) using a 2 dm cell, $[\alpha]_D$ values are given in units of 10^{-1} deg cm^2 g^{-1}. Autotitrations were carried out using an RTS 882 recording titration system (Radiometer Ltd.). Flash chromatography was carried out using Kieselgel 60 (230–400 mesh) (Merck). Biofix beads were obtained from English China Clays.

Diethyl (2S,3S)-2,3-*Dihydroxybutane*-1,4-*dioate* [*Diethyl* (2S,3S)-*Tartrate*] **5**, *Diethyl* (2R,3R)-2,3-*Dihydroxybutane*-1,4-*dioate and Diethyl* (2RS,3RS)-2,3-*Dihydroxybutane*-1,4-*dioate*.—To a suspension of (2*S*,3*S*)-tartaric acid (19.8 g) in benzene (82 cm^3) was added ethanol (40 cm^3) and Dowex 50W-X8 ion exchange resin (H^+ form, 5.4 g). The mixture was boiled under reflux in an apparatus fitted with a Dean and Stark trap until no more water was trapped and all of the tartaric acid had gone into solution (≤24 h). The mixture was filtered and the resin was washed with benzene. The combined filtrate and washings were evaporated under reduced pressure. The residue was distilled (Kugelrohr; 135 °C/0.1 mmHg) to give diethyl (2*S*,3*S*)-tartrate,[27] 24.9 g (91%). Diethyl (2*R*,3*R*)-tartrate **27** was obtained in a similar manner in 89% yield, as was diethyl (±)-tartrate **16** in 82% yield. For the racemic ester ν_{max}/cm^{-1} 3480 (br s, OH) and 1750 (CO); δ_H(220 MHz; $CDCl_3$) 1.35 (t, 6 H, *J* 7.3, 2 × CH_2*Me*), 2.40–2.70 (br s, 2 H, 2 × OH), 4.37 (q, 4H, *J* 7.3, 2 × CH_2Me) and 4.60 (s, 2 H, CHCH); δ_C 13.95 (2 × CH_2Me), 62.25 (2 × CH_2Me), 71.95 (2 × HCOH) and 171.42 (2 × CO).

Diethyl (2RS,3RS)-2-*Acetoxy*-3-*bromosuccinate*.—To stirred and ice-cooled diethyl DL-tartrate (23.2 g) was added dropwise a solution of HBr (30%) in glacial acetic acid (68 cm^3). The mixture was stirred for 15 min at 0 °C and for 4 h in the dark at room temp. The mixture was poured into ice–water (75 cm^3) and the aqueous solution was extracted with several portions of ether. The combined ether extracts were washed with saturated brine, dried ($MgSO_4$) and evaporated under reduced pressure to give diethyl (2*RS*,3*RS*)-2-acetoxy-3-bromosuccinate[17] as a pale yellow oil that was used without further purification. ν_{max}/cm^{-1} 1752 and 1715sh; δ_H(220 MHz; $CDCl_3$) 1.31 (t, 6 H, *J* 7.3), 2.19 (s, 3 H, COMe), 4.20–4.40 (m, 4 H, 2 × CH_2Me), 4.82 (d, 1 H, *J* 5.4, CHBr) and 5.63 (d, 1 H, *J* 5.4, ArOC*H*).

Diethyl (2RS,3RS)-2-*Bromo*-3-*hydroxysuccinate*.—To a solution of diethyl (±)-2-acetoxy-3-bromosuccinate (30.5 g) in dry ethanol (260 cm^3) was added a solution of HBr (30%) in glacial acetic acid (8.5 cm^3). The solution was boiled under reflux under N_2 for 4 h and evaporated under reduced pressure. The residue was purified by distillation (88–90 °C/0.02 mmHg), flash chromatography (ethyl acetate–light petroleum b.p. 40–60 °C) and distillation (Kugelrohr, 130 °C/0.08 mmHg) to give diethyl (±)-3-bromo-2-hydroxysuccinate[17] as a colourless oil (14.4 g, 55%); δ_H(220 MHz; $CDCl_3$) 1.32 (t, 6 H, *J* 7.3, CH_2*Me*), 3.49 (d, 1 H, *J* 7.3, OH), 4.24–4.45 (m, 4 H, 2 × CH_2Me) and 4.64–4.80 (m, 2 H, CHCH).

Diethyl (2RS,3RS)-*Epoxysuccinate* (as **4**).—To a stirred solution prepared from sodium (0.28 g) in dry ethanol (6.6 cm^3) was added dropwise at 0 °C a solution of racemic diethyl 3-bromo-2-hydroxysuccinate (2.63 g) in dry ethanol (2.3 cm^3). The mixture was stirred at room temp. for 1 h, neutralised (HOAc) and evaporated under reduced pressure. The residue was taken up in ice–water and the solution was extracted with diethyl ether (3 × 35 cm^3). The ether extracts were washed with saturated brine, dried ($MgSO_4$) and evaporated. The residue was distilled (Kugelrohr, 130 °C/0.1 mmHg) to give diethyl (2*RS*,3*RS*)-epoxysuccinate (as **4**) (1.5 g, 84%). The product was further purified by flash chromatography [ethyl acetate–light petroleum (1:4)] to give the diester **1**, 1.09 g (59%); ν_{max}/cm^{-1} 1745 (CO); δ_H(220 MHz; $CDCl_3$) 1.32 (t, 6 H, *J* 7.3, 2 × CH_2*Me*), 3.68 (s, 2 H, CHO) and 4.29 (q, 4 H, 2 × CH_2Me).

Diethyl (2S,3S)-*Epoxysuccinate* **4**.—This was prepared as for the racemic diester in 81% yield $[\alpha]_D^{27}$ 93.9 (*c* 0.36, $CHCl_3$), $[\alpha]_D^{27}$ 108.4 (*c* 0.289, diethyl ether) [lit.,[17] $[\alpha]_D^{23}$ 105.5 (*c* 1.413, diethyl ether)]; δ_H(400 MHz; $CDCl_3$) 1.29 (t, 6 H, *J* 7.14, 2 × CH_2*Me*), 3.64 (s, 2 H, 2 × CHO), 4.22 (dq, 2 H, *J* 10.82, 7.14, 2 × C*H*HMe) and 4.26 (dq, 2 H, *J* 10.82, 7.14, 2 × CH*H*Me). Enantiomeric purity was determined from the ^{1}H NMR spectrum determined in $CDCl_3$ with [$^2H_6$]-benzene/[$^2H_5$]-benzene as internal standard and in the presence of three molar equivalents of (*S*)-(+)-2,2,2-trifluoro-1-

(9-anthryl)ethanol. A single peak for the epoxide methine proton was observed at δ 3.38. Upon addition of an equal amount of racemic material with the addition of three molar equivalents of the chiral solvating agent, a signal of one-third the intensity of the downfield peak appeared at δ 3.30 corresponding to the methine proton of diethyl (2*R*,3*R*)-epoxysuccinate.

Diethyl meso-*Tartrate* **7**.—Diethyl maleate (2.18 g) was added slowly to a mixture of *N*-methylmorpholine *N*-oxide (22.3 g), water (61 cm^3), acetone (25 cm^3) and osmium tetraoxide [80 mg, previously dissolved in *tert*-butanol (10 cm^3)]. The mixture was allowed to stand with stirring overnight under N_2. A slurry of sodium hydrogen sulfite (1 g) and magnesium silicate (12 g) in water (80 cm^3) was added. The mixture was filtered and the filtrate was brought to pH 7 (0.5 mol dm^{-3} H_2SO_4). The acetone was removed under reduced pressure and the residue was adjusted to pH 2. The solution was extracted with NaCl and extracted with ethyl acetate. The extracts were dried ($MgSO_4$) and extracted with ethyl acetate. The extracts were dried and evaporated under reduced pressure. The solid residue was recrystallised [diethyl ether–light petroleum (b.p. 40–60 °C)] to give diethyl *meso*-tartrate **7** (19.9 g, 79%), m.p. 56–57.5 °C (lit.,[28] m.p. 55 °C) (Found: M + 1, 207.0874. Calc. for $C_8H_{15}O_6$: 207.0868. Found: C, 46.65; H, 7.00. Calc. for $C_8H_{14}O_6$: C, 46.60; H, 6.85%); ν_{max}/cm^{-1} 3420, 1755 and 1740; δ_H(220 MHz; $CDCl_3$) 1.30 (t, 6 H, *J* 7.3, 2 × CH_2Me), 3.35 (br s, 1 H, OH), 4.20–4.42 (m, 4 H, 2 × CH_2Me) and 4.59 (s, 2 H, 2 × HCO); δ_C 13.91 (Me), 62.09 (CH_2Me), 72.80 (HCO) and 170.89 (CO); *m*/*z* (EI) (relative abundance) 207 [(M + 1)$^+$, 9.4%], 133 (49), 104 (87), 76 (52) and 69 (100). When an attempt was made to convert this ester into the corresponding epoxide following the procedure described above starting from diethyl (2*RS*,3*RS*)-tartrate, a mixture was obtained which, by ^1H NMR spectroscopy was found to consist of *cis*- and *trans*-epoxides **9** and **10** in the ratio 54:46 (see Scheme 2).

Hydrolysis of Diethyl (±)-*Epoxysuccinate by the Lipase from* Rhizopus javanicus.—Diethyl (±)-epoxysuccinate ±-**11** (135 mg, 0.72 mmol) was dissolved in isooctane (38 cm^3). To the stirred solution was added phosphate buffer (pH, 7, 1 mol dm^{-3}, 9.5 cm^3) containing lipase from *Rhizopus javanicus* (9600 units, 64 mg). Sodium hydroxide (0.1 mol dm^{-3}) was added from an autotitrator to maintain pH 7. When hydrolysis reached 26% of all ester groups the organic phase was separated and evaporated under reduced pressure to give unhydrolysed diethyl epoxysuccinate, 62% ee.

Enzyme-catalysed Transesterification of Diethyl (2RS,3RS)-2,3-*Epoxybutane*-1,4-*dioate*.—A typical transesterification was carried out as follows: to a solution of racemic diethyl (2*RS*,3*RS*)-2,3-epoxybutane-1,4-dioate ±-**11** (0.10 g, 0.54 mmol) and heptanol (0.5 cm^3) in isooctane (12 cm^3) was added lipase LP (25 mg, 750 U). The mixture was stirred at 50 °C. Periodically samples were analysed by gas chromatography [temperature programme 130 °C (10 min), 6 °C min^{-1} to 250 °C]. After 9 days no starting material was detectable and the mixed ester **13** and the diheptyl ester **14** were present in a ratio of 47:53. The mixture was filtered and the heptanol was removed by distillation (Kugelrohr). The esters were separated by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (95:5, v/v)] to give ethyl heptyl 2,3-epoxybutane-1,4-dioate (as **13**), (51 mg, 40%) and diheptyl 2,3-epoxybutane-1,4-dioate (as **14**), (78 mg, 40%) as colourless oils. Chiral analysis showed in each case that the 2*S*,3*S*-enantiomer was present in excess. This was determined by a repetition of the experiment using optically pure diethyl (2*S*,2*S*)-2,3-epoxybutane-1,4-dioate obtained from diethyl (2*S*,3*S*)-tartrate as described above. The mixed ester **13** had $[\alpha]_D$ 73.4 (*c* 0.54, $CHCl_3$) and the diheptyl ester **14** had $[\alpha]_D$ 59.3 (*c* 0.44, $CHCl_3$). The optical purity was determined by ^1H NMR spectroscopy ($CDCl_3$) in the presence of the chiral solvating agent. The epoxide protons of the mixed ester **13** gave an AB system δ 3.39, fully resolved (baseline) on addition of racemate and showing that the downfield component could be assigned to the (2*S*,3*S*)-enantiomer. For the diheptyl ester **14** a corresponding singlet was obtained δ 3.37, fully resolved (baseline) on addition of chiral solvating agent and showing that the downfield signal could be assigned to the (2*S*,3*S*)-enantiomer. Ethyl heptyl 2,3-epoxybutane-1,4-dioate: (Found: M + 1 259.1576. $C_{13}H_{23}O_5$ requires 259.1545. Found: C, 60.4; H, 8.8. $C_{13}H_{22}O$ requires C, 60.45; H, 8.58%); ν_{max}(thin film)/cm^{-1} 1750; δ_H(400 MHz; $CDCl_3$) 0.87 (3 H, t, *J* 6.6, $MeCH_2CH_2$), 1.2–1.4 [8 H, m, $Me(CH_2)_4$], 1.37 (3 H, t, *J* 7.2, $MeCH_2O$), 1.66 (2 H, m, OCH_2CH_2), 3.65 (2 H, m, HCOCH) and 4.12–4.32 (4 H, m, $MeCH_2O$, $CH_2CH_2CH_2O$); δ_C 13.89 (2 × Me), 22.40, 25.57, 28.28, 28.67 [$Me(CH_2)_4$], 31.51 ($CH_2CH_2CH_2O$), 51.92 (COC), 62.08 ($MeCH_2O$), 66.23 (CH_2CH_2O) and 166.66, 166.73 (2 × CO); *m*/*z* 161 (64%), 143 (12), 133 (23), 115 (24), 98 (54), 87 (44), 71 (26), 70 (63), 69 (37), 57 (100), 56 (62), 55 (52), 43 (56), 42 (27) and 41 (70). Diheptyl 2,3-epoxybutane-1,4-dioate [Found: (M + 1)$^+$ 329.2315. $C_{18}H_{33}O_5$ requires 329.2327. Found: C, 65.8; H, 9.9. $C_{18}H_{32}O_5$ requires C, 65.82; H, 9.82%]; ν_{max}/cm^{-1} 1760; δ_H(400 MHz; $CDCl_3$) 0.87 (3 H, t, *J* 7.0, $MeCH_2$), 1.2–1.4 [10 H, m, $(CH_2)_5Me$], 1.66 (4 H, m, 2 × CH_2O), 3.65 (2 H, s, CHOCH) and 4.13–4.24 (4 H, m, 2 × CH_2O); δ_C 13.85 (2 × Me), 22.39, 25.56, 28.27, 28.66 [$(CH_2)_5Me$], 31.50 (2 × CH_2CH_2O), 51.91 (COC), 66.20 (2 × CH_2O) and 166.73 (2 × CO); *m*/*z* 329 [(M + 1)$^+$, 0.5%], 271 (0.3), 257 (0.3), 231 (7), 185 (5), 143 (41), 133 (37), 115 (5), 99 (91), 87 (32), 70 (66) and 57 (100).

Transesterification with Lipase LF-AP15 *from* Rhizopus javanicus.—In a typical experiment the racemic diethyl 2,3-epoxybutane-1,4-dioate (0.32 g), heptanol (1.6 cm^3) and lipase (14 mg, 21, 150 U) were stirred in isooctane (40 cm^3) for 48 d. Unchanged diethyl ester **12** (56 mg, 36%) and diheptyl ester **14** (88 mg, 32%) were recovered as above, both of > 97% ee by ^1H NMR spectroscopy using the chiral solvating agent. By comparison with authentic compounds (above), the diethyl ester was shown to have the (2*R*,3*R*)-configuration, and the diheptyl ester the (2*S*,3*S*)-configuration.

Spontaneous Hydrolysis of Diethyl cis-2,3-*Epoxybutane*-1,4-*dioate* **9** *in Phosphate Buffer at* pH 7.—Diethyl *cis*-2,3-epoxybutane-1,4-dioate **9** (0.2 g) was suspended in phosphate buffer (0.1 mol dm^{-3}, 6 cm^3). The mixture was stirred vigorously with the addition of NaOH (0.3 mol dm^{-3}) from an autotitrator and pH stat at pH 7. When 3.75 cm^3 of NaOH solution had been added, the reaction mixture was extracted with ethyl acetate (4 × 20 cm^3). The aqueous phase was brought to pH 1 (5 cm^3 HCl) and re-extracted with ethyl acetate (4 × 20 cm^3). The combined extracts from the acidified solution were dried ($MgSO_4$) and evaporated to give ethyl hydrogen (2*RS*,3*SR*)-2,3-epoxybutane-1,4-dioate, 0.12 g, 98%. The crude product was purified by distillation (Kugelrohr, 150 °C, 0.01 mmHg) to give the half ester in 77% yield [Found: (M + 1)$^+$ 161.0433. $C_6H_9O_5$ requires *M*, 161.0450]; ν_{max}/cm^{-1} 1750; δ_H(220 MHz; $CDCl_3$) 1.33 (3 H, t, *J* 7.3, CH_2Me), 3.73 (2 H, s, HCOCH), 4.31 (2 H, q, CH_2Me) and 7.87 (1 H, s, OH); δ_C 13.77 (Me), 52.71 (COC), 62.70 (CH_2Me) and 166.28, 168.56 (CO); *m*/*z* (CI, NH_3) 178 [(M + NH_4)$^+$, 100%], 161 (3), 145 (4), 134 (31), 117 (25), 88 (3) and 35 (37).

Hydrolysis of Diethyl cis-2,3-*Epoxybutane*-1,4-*dioate* **9** *Catalysed by Pig Liver Esterase*.—To diethyl *cis*-2,3-epoxybutane-

1,4-dioate **9** (0.19 g) suspended in phosphate buffer (0.1 mol dm^{-3}, pH 7, 6 cm^3) was added pig liver esterase (260 U). The mixture was stirred and maintained at 37 °C with addition of NaOH solution (1 mol dm^{-3}) using an autotitrator and pH stat. When 1 mol mol^{-1} substrate had been added, the mixture was brought to pH 2 (5 mol dm^{-3} HCl) and extracted with ethyl acetate (4 × 20 cm^3). The combined extracts were dried and evaporated under reduced pressure to give the half ester (0.15 g, 92%). This was purified by distillation (Kugelrohr, 88 °C, 0.07 mmHg). The spectroscopic data were identical with those obtained for the non-enzymatically hydrolysed sample (above). Chiral analysis was carried out by ^{1}H NMR spectroscopy using (*R*)-(+)-α-methylbenzylamine (1 equiv.) in $CDCl_3$. Splitting of the signal attributable to the methyl group was observed, the downfield component having 1.65 times the intensity of the upfield signal (25% ee).

Dibutylstannylene Derivative of Diethyl (2RS,3RS)-*Tartrate* ±-**15**.—This was prepared as for the derivative of the *meso*-tartrate, below, in 95% yield, m.p. 138–139 °C (Found: C, 44.1; H, 7.15. $C_{16}H_{30}O_6Sn$ requires C, 43.97; H, 6.92%); ν_{max}/cm^{-1} 1755 and 1730; *m/z* 439 [(M + H)$^+$, 11%], 381 (36), 365 (29), 336 (55), 251 (15), 223 (60) and 177 (26).

Dibutylstannylene Derivative of Diethyl meso-*Tartrate* **17**.—A mixture of diethyl *meso*-tartrate **7** (14.38 g, 69.7 mmol) and dibutyltin oxide (17.4 g, 69.8 mmol) in benzene (115 cm^3) was boiled under reflux in an apparatus fitted with a Dean and Stark trap for 5 h. The mixture was concentrated to approximately 30 cm^3 and cooled. The stannylene **17** (29.5 g, 97%) was filtered off and dried under reduced pressure over paraffin wax. A portion was crystallised (benzene) m.p. 158 °C (decomp.) (Found: C, 44.1; H, 7.20. $C_{16}H_{30}O_6Sn$ requires C, 43.97; H, 6.92%); ν_{max}/cm^{-1} 1745 and 1760; *m/z* 439 [(M + H)$^+$, 7%], 381 (34), 365 (44), 336 (88), 251 (26), 223 (100) and 177 (52).

Diethyl (2RS,3RS)-2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate* ±-**16**.—This was prepared as described below and purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (1:1, v/v)] to give the ester ±-**16** in 91% yield, m.p. 62.5–64.0 °C (ether–light petroleum) [Found: (M + 1)$^+$ 361.0961. $C_{15}H_{21}O_8S$ requires: 361.0957; Found: C, 49.90; H, 5.52. $C_{15}H_{20}O_8S$ requires C, 49.99; H, 5.59%]; ν_{max}/cm^{-1} 3470br, 1775 and 1750; δ_H(220 MHz; $CDCl_3$) 1.22–1.35 (6 H, m, 2 × CH_2*Me*), 2.49 (3 H, s, Ar*Me*), 3.17 (1 H, d, *J* 9, OH), 4.07–4.42 (4 H, m, 2 × CH_2Me), 4.79 [1 H, dd, *J* 9, 3, CH(OH)], 5.41 (1 H, d, *J* 3, CHOTs), 7.44 (2 H, d, *J* 9, Ar *m*-H × 2) and 7.93 (2 H, d, *J* 9, Ar *o*-H × 2); δ_C 13.81 (2 × CH_2*Me*), 21.53 (Ar*Me*), 62.37, 62.92 (2 × CH_2Me), 71.15 (HOCH), 77.14 COTs), 128.07, 129.56, 133.09, 145.14 (Ar) and 165.65, 169.72 (CO); *m/z* 361 [(M + H)$^+$, 2%], 332 (0.2), 289 (2), 258 (7), 241 (5), 173 (9), 156 (23), 139 (6), 115 (6), 103 (19) and 91 (100).

Diethyl (2RS,3SR)-2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate* **18**.—A mixture of the dibutylstannylene **17** prepared from diethyl *meso*-tartrate (4.14 g, 9.47 mmol), toluene-*p*-sulfonyl chloride (1.82 g, 9.54 mmol) and tetraethylammonium chloride (1.75 g, 9.52 mmol) in acetonitrile (200 cm^3) was stirred at room temp. for 24 h. The solution was evaporated under reduced pressure and the residue was purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (7:3, v/v)] to give the ester **18** (3.23 g, 95%), as a colourless oil (Found: M$^+$ 361.0954. $C_{15}H_{21}O_8S$ requires *M*, 361.0975. Found: C, 50.3; H, 5.6. $C_{15}H_{20}O_8S$ requires C, 49.99; H, 5.59%); ν_{max}/cm^{-1} 3540br, 1755sh and 1750; δ_H(220 MHz; $CDCl_3$) 1.15–1.40 (6 H, m, 2 × Me), 2.47 (3 H, s, Ar*Me*), 3.17 (1 H, d, *J* 7, OH), 4.10–4.44 (4 H, m, 2 × CH_2Me), 4.63–4.72 [dd, *J* 3, 7, CH(OH)], 5.37 [1 H, d, *J* 3, CH(OTs)], 7.42 (2 H, d, *J* 10, Ar *m*-H × 2) and 7.92 (2 H, d, *J* 10, Ar *o*-H × 2); δ_C 13.80, 13.84 (CH_2*Me*), 21.56 (Ar*Me*), 62.20, 62.75 (2 × CH_2Me), 71.06 (HOCH), 73.87 (HCOTs), 128.00, 129.63, 133.14, 145.13 (Ar) and 165.22, 169.64 (CO); *m/z* (CI, NH_3) 361 [(M + H)$^+$, 12%], 287 (54), 258 (8), 188 (5), 155 (100), 108 (15), 103 (16) and 91 (52).

Diethyl (2RS,3RS)-*Epoxysuccinate* ±-**11**.—To a solution of sodium ethoxide prepared from sodium (0.23 g, 10 mmol) and dry ethanol (5.7 cm^3) was added dropwise at 0 °C a solution of diethyl (2*RS*,3*SR*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)-butane-1,4-dioate ±-**18** (2.88 g, 8 mmol) in dry ethanol (2 cm^3). The mixture was stirred for 1 h, neutralised (HOAc) and concentrated under reduced pressure. The residue was dissolved in ice–water and the solution was extracted with diethyl ether (3 × 35 cm^3). The combined ethereal extracts were washed with saturated NaCl solution, dried ($MgSO_4$) and evaporated. The residue was distilled (Kugelrohr, 90 °C/0.07 mmHg) to give the ester ±-**11** as a colourless oil (0.26 g, 84%). ^{1}H NMR spectroscopic data as for the material prepared by the method of Mori[17] above [Found: (M + 1)$^+$ 189.0808. $C_8H_{13}O_5$ requires 189.0763. Found: C, 51.0; H, 6.7. $C_8H_{12}O_5$ requires C, 51.06; H, 6.43%]; δ_C 13.91 (Me), 51.91 (C–O–C), 62.11 (CH_2) and 166.65 (CO); ν_{max}/cm^{-1} 1750br; *m/z* (CI, NH_3) 189 [(M + 1)$^+$, 40%], 161 (13), 143 (9), 114 (100), 99 (10), 89 (7), 71 (18) and 59 (12).

Diethyl cis-*Epoxysuccinate*.—Attempts to prepare this ester from diethyl (2*RS*,3*RS*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate ±-**16** by the method described above[17] led invariably to a mixture of the *cis*- and *trans*-epoxy esters in a ratio of 4:1. The *cis*-ester **9** was isolated and purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (85:15, v/v)] (24% yield). Its purity was confirmed by HPLC [Spherisorb C_{18} column, acetonitrile:water (1:1, v/v), UV detection (214 nm)]. The *cis*-epoxide was eluted first: spectral data as for the material prepared by the alternative route below.

Hydrolysis Catalysed by α-*Chymotrypsin of Diethyl* 2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate* ±-**16**.—In a typical procedure the racemic diester ±-**16** (2.07 g) in dichloromethane (2.5 cm^3) was mixed with phosphate buffer (pH 7, 0.1 mol dm^{-1}, 35 cm^3). To the stirred mixture at 37 °C was added α-chymotrypsin (210 mg, 10 710 U). The aqueous solution was maintained at pH 7 by the addition of NaOH solution (0.31 mol dm^{-3}) using an autotitrator and pH stat. After 22 h, the solution was brought to pH 2 (5 mol dm^{-3} HCl) to halt the reaction. The volume of NaOH solution added corresponded to the hydrolysis of 25% of all ester groups. The solution was brought to pH 7 (5 mol dm^{-3} NaOH) and extracted with ethyl acetate to recover unhydrolysed substrate. The aqueous solution was brought to pH 1 (5 mol dm^{-3} HCl) and extracted with ethyl acetate. The extracts were dried ($MgSO_4$) and evaporated under reduced pressure. The product was purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate, 8:2 v/v] and isolated as a colourless oil, 0.99 g, 48%. Chiral analysis was carried out using ^{1}H NMR spectroscopy in CCl_4 with the chiral solvating agent and [$^2H_5$]-benzene as reference. Except for signals attributable to the ethyl group, all signals showed baseline splitting. The signal attributable to the methyl group attached to the benzene ring was particularly well resolved and were used to determine optical purity (59% ee in this case). By comparison with the pure enantiomers prepared from the pure enantiomers of tartaric acid, the major isomer was found to have the (2*R*,3*R*)-configuration. (The downfield signal was attributable to this derivative). The spectroscopic data for the recovered diester were as reported above. The ^{1}H NMR spectrum of the monoacid was consistent with the proposed structure: δ_H(220

MHz; $CDCl_3$) 1.30 (3 H, t, *J* 6.1, CH_2Me), 2.45 (3 H, s, Ar*Me*), 4.14 (2 H, m, OCH_2Me), 4.78 (1 H, d, *J* 3, *H*COH), 5.40 (1 H, d, *J* 3, TsOC*H*), 7.39 (2 H, d, *J* 9.3, Ar-*H*) and 7.95 (2 H, d, *J* 9.3, Ar-*H*). On addition of proton sponge (1 mol mol^{-1}) the signal δ 5.40 was shifted upfield by 0.2 ppm. Full data for the optically pure product are given below.

Diethyl 2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate*.—A mixture of ethyl hydrogen 2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate, from the α-chymotrypsin-catalysed hydrolysis, above, (0.8 g, 2.4 mmol), 1-hydroxybenzotriazole hydrate (HOBT) (0.32 g, 2.4 mmol) and ethanol (1.1 cm^3) in dichloromethane (4.2 cm^3) was cooled to 0 °C and stirred under N_2. Dicyclohexylcarbodiimide (DCCI) (0.61 g, 2.9 mmol) was added. The mixture was left at 0 °C for 1 h, allowed to warm to room temp. overnight and filtered. The precipitate was washed with dichloromethane (3 × 4 cm^3). The combined filtrate and washings were washed with hydrochloric acid (5 mol dm^{-1}, 2 cm^3) and water, dried (Na_2SO_4) and evaporated under reduced pressure. The crude product was purified by flash chromatography to give diethyl (2*S*,3*S*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate (0.49 g, 57%, 66% ee).

Diethyl (2RS,3RS)-2-tert-*Butyldimethylsilyloxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate*.—To a solution of diethyl (2*RS*,3*RS*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate ±-**16** (7.08 g, 19.6 mmol) and imidazole (4.6 g, 67.6 mmol) in dimethylformamide (17.4 cm^3) was added, under N_2, *tert*-butylchlorodimethylsilane (5.12 g, 34 mmol). The mixture was stirred at 25 °C for 7.5 h. Ethyl acetate (300 cm^3) was added and the solution was washed with water (2 × 50 cm^3). The organic phase was dried ($MgSO_4$), the solvent was removed under reduced pressure and the product was purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (4:1, v/v)]. The derivative ±-**19** was obtained as a viscous oil (9.3 g, 100%) (Found: C, 53.25; H, 6.95. $C_{21}H_{34}O_8SSi$ requires C, 53.14; H, 7.22%); ν_{max}(thin film)/cm^{-1} 1750 and 1740; δ_H(220 MHz; $CDCl_3$) 0.00 and 0.09 (each 3 H, s, SiMe), 0.84 (9 H, s, CMe_3), 1.13–1.31 (6 H, m, 2 × CH_2Me), 2.43 (3 H, s, Ar*Me*), 3.91–4.29 (4 H, m, 2 × CH_2Me), 4.74 (1 H, d, *J* 4, HCOSi), 5.33 (1 H, d, *J* 4, HCOTs), 7.37 (2 H, d, *J* 9, Ar-H) and 7.85 (2 H, d, *J* 9, Ar-H); δ_C − 5.81 (SiMe), −5.02 (SiMe), 13.71, 13.84 (2 × CH_2Me), 18.09 (CMe_3), 21.52 (Ar*Me*), 25.38 (CMe_3), 61.69, 62.09 (2 × OCH_2Me), 72.72 (SiOC), 78.51 (COTs), 128.01, 129.44, 133.60, 144.73 (Ar) and 166.15, 168.81 (2 × CO); *m*/*z* (CI, NH_3) 492 [$(M + NH_4)^+$, 100%], 475 [$(M + H)^+$, 15], 459 (6), 417 (25), 229 (18), 189 (18), 155 (13), 108 (13) and 91 (20).

Diethyl cis-2,3-*Epoxybutane*-1,4-*dioate* **9**.—To a solution of the *tert*-butyldimethylsilyl ether ±-**19** of diethyl (2*RS*,3*RS*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate ±-**16** (0.31 g) in dry tetrahydrofuran (THF) was added under N_2 a solution of tetrabutylammonium fluoride in THF (0.8 cm^3; 1 mol dm^{-3}). After 2 h, an additional 0.4 cm^3 of the fluoride solution was added. After 3 h, the solution was concentrated under reduced pressure. The residue was dissolved in ice–water (10 cm^3) and the solution was extracted with diethyl ether (3 × 60 cm^3). The ethereal solution was dried ($MgSO_4$) and evaporated under reduced pressure. The residue was purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (85:15, v/v)], to give the ester **9** (109 mg, 87%) (Found: M^+, 188.0699. $C_8H_{12}O_5$ requires *M*, 188.0684. Found: C, 50.90; H, 6.45. $C_8H_{12}O_5$ requires C, 51.06; H, 6.43%); ν_{max}/cm^{-1} 1750; δ_H(220 MHz; $CDCl_3$) 1.31 (6 H, t, *J* 7.2, 2 × CH_2Me), 3.72 (2 H, s, HCO) and 4.29 (4 H, q, *J* 7.5, 2 × CH_2Me); δ_C 13.75 (2 × Me), 52.3 (COC), 61.67 (2 × CH_2Me) and 165.45 (2 × CO); *m*/*z* 188 (M^+, 8%), 161 (55), 143 (65), 115 (71), 99 (10), 87 (53), 70 (100), 59 (10) and 43 (35).

Diethyl (2S,3S)-2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)-*butane*-1,4-*dioate* **19**.—A solution of the cyclic stannylene from diethyl D-tartrate prepared as above (2.7 g, 6.2 mmol), toluene-*p*-sulfonyl chloride (1.2 g, 6.2 mmol) and tetramethylammonium chloride (1.14 g, 6.2 mmol) in acetonitrile (130 cm^3) was stirred at room temp. for 24 h. The solution was evaporated under reduced pressure and the residue was purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate (1.5:1, v/v)] to give diethyl (2*S*,3*S*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate **19** (0.912 g, 41%) [Found: $(M + 1)^+$, 361.0945. $C_{15}H_{21}O_8S$ requires 361.0957]; $[\alpha]_D^{26}$ −16.6 (*c* 10.1, $CHCl_3$); ν_{max}/cm^{-1} 1745; δ_H(400 MHz; $CDCl_3$) 1.24 (3 H, t, *J* 7.17, CH_2Me), 1.29 (3 H, t, *J* 7.15, CH_2Me), 2.44 (3 H, s, Ar*Me*), 4.1 (4 H, m, 2 × CH_2Me), 4.72 (1 H, d, *J* 2.1, *H*COH), 5.34 (1 H, d, *J* 2.1, HCOTs), 7.34, (2 H, d, *J* 8.19, ArH) and 7.82 (2 H, d, *J* 6.69, ArH); δ_C 13.82 (2 × CH_2CH_3), 21.56 (ArCH_3), 62.41, 62.94 (each CH_2Me), 71.14 (CH_2Me), 77.17 (HCOTs), 128.08, 129.60 (Ar), 133.03 ($C_{4r}Me$), 145.20 ($C_{4r}SO_2$) and 165.69, 169.74 (each CO); *m*/*z* 361 [$(M + 1)^+$, 6%], 287 (18), 258 (3), 173 (4), 155 (49), 91 (100) and 65 (20).

Ethyl 2-*Methypropyl* (2S,3S)-2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate* **21**.—Hydrolysis of diethyl (2*S*,3*S*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate **19** (4.07 g) was carried out as described above but at 37 °C and using α-chymotrypsin (210 mg, 10 710 U). A portion (1.02 g) of the monoacid **20** produced (3.44 g, 92%) was esterified as above using HOBT (417 mg), 2-methylpropan-1-ol (2.2 cm^3) and DCCI (783 mg) in dichloromethane (5.4 cm^3). The product, isolated as above was purified by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate, 8:2 v/v] to give ethyl 2-methylpropyl (2*S*,3*S*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate **21**, (0.58 g, 48%) [Found: $(M + 1)^+$ 389.1215. $C_{17}H_{25}O_8S$ requires 389.1270. Found: C, 52.7; H, 6.25. $C_{17}H_{24}O_8S$ requires C, 52.57; H, 6.23%]; ν_{max}/cm^{-1} 3500br, 1770sh and 1745; δ_H(220 MHz; $CDCl_3$) 0.93 (6 H, d, *J* 7.0, Me_2CH), 1.30 (3 H, t, *J* 7.6, $MeCH_2$), 1.95 (1 H, m, Me_2CH), 2.46 (3 H, s, Ar*Me*), 3.13 (1 H, d, *J* 9.3, OH), 3.99 (2 H, d, *J* 7.3, Me_2CHCH_2), 4.10, 4.27 (each 1 H, m, OCH_aH_bMe), 4.75 (1 H, app d, J_{app} 9.0, *H*COH), 5.39 (1 H, s_{app}, TsOC*H*), 7.39 (2 H, d, *J* 8.8, Ar-*H*) and 7.88 (2 H, d, *J* 8.8, Ar-*H*); δ_C 13.75 ($MeCH_2$), 18.70 (Me_2CH), 21.47 (Ar*Me*), 27.52 (Me_2CH), 62.83 ($MeCH_2$), 71.17 (HCOH), 72.15 (CH_2CHMe_2), 77.12 (HCOTs), 128.00, 129.54, 133.14 (Ar), 165.69 (CO) and 169.68 (CO); *m*/*z* (CI, NH_3), 406 [$(M + NH_4)^+$, 9%], 389 [$(M + 1)^+$, 28], 333 (11), 315 (35), 287 (17), 259 (28), 231 (10), 203 (9), 173 (43), 155, (100) and 133 (17).

Ethyl 2-*Methylpropyl* (2S,3S)-2-tert-*Butyldimethylsilyloxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate*.—Ethyl 2-methylpropyl (2*S*,3*S*)-2-hydroxy-3-(4-methylphenylsulfonyloxy)-butane-1,4-dioate (1.86 g) was silylated as described above for the corresponding diethyl ester to give ethyl 2-methylpropyl (2*S*,3*S*)-2-*tert*-butyldimethylsilyloxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate in 94% yield as a viscous oil after purification by flash chromatography [light petroleum (b.p. 40–60 °C)–ethyl acetate, 9:1 v/v] (Found: C, 54.9; H, 7.75. $C_{23}H_{38}O_8SSi$ requires C, 54.95; H, 7.62%); ν_{max}/cm^{-1} 1750 and 1740; δ_H(220 MHz; $CDCl_3$) 0.00 (3 H, s, SiMe), 0.09 (3 H, s, SiMe), 0.85 (9 H, s, Me_3CSi), 0.93 (6 H, d, *J* 7.3, Me_2CH), 1.23 (3 H, t, *J* 7.6, $MeCH_2$), 1.94 (1 H, m, Me_2CH), 2.46 (3 H, s, Ar*Me*), 3.93 (2 H, m, $MeCH_2$), 4.75 (1 H, d, *J* 3, SiOCH), 5.35 (1 H, d, *J* 3, *H*COTs), 7.37 (2 H, d, *J* 8, Ar-H) and 7.86 (2 H, d, *J* 8, Ar-*H*); δ_C −5.80 (SiMe), −5.00 (SiMe), 13.76 (CH_2Me), 18.02 (SiC),

18.80 ($CHMe_2$), 21.41 (Ar*Me*), 25.35 (CMe_3), 27.40 ($CHMe_2$), 61.57 (CH_2Me), 72.06 (CH_2CHMe_2), 72.71 (SiOC), 78.46 (HCOTs), 127.88, 129.40, 133.80, 144.64 (Ar) and 166.22, 168.77 (CO); *m/z* (CI, NH_3) 520 [$(M + NH_4)^+$], 503 $(M + H)^+$ and 155.

Ethyl 2-Methylpropyl (2S,3R)-*Epoxybutane*-1,4-*dioate* **22**.—Ethyl 2-methylpropyl (2*S*,3*S*)-2-*tert*-butyldimethylsilyloxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate (1 g) was subjected to the ring closure procedure described above for the corresponding diethyl ester to give ethyl 2-methylpropyl (2*S*,3*R*)-epoxybutane-1,4-dioate **22** in 81% yield after flash chromatography [light petroleum (b.p. 40–60 °C)]–ethyl acetate, 85:15 v/v) and distillation (Kugelrohr, 90 °C, 0.01 mmHg) [Found: $(M + 1)^+$, 217.1089. $C_{10}H_{17}O_5$ requires 217.1076. Found: C, 55.3; H, 7.5. $C_{10}H_{16}O_5$ requires C, 55.55; H, 7.46%]; δ_H(400 MHz; CCl_4, [$^2H_6$]-benzene/[$^2H_5$]-benzene = δ 7.20 ppm) 0.850, 0.853 (each 3 H, d, *J* 6.74, Me_2CH), 1.145 (3 H, t, *J* 7.14, CH_2Me), 1.841 (1 H, sept., *J* 6.72, Me_2CH), 3.301 (1 H, d, *J* 4.50, HCOC), 3.317 (1 H, d, *J* 4.50, COCH), 3.77 (1 H, dd, *J* 6.65, 10.65, $CH_aH_bCHMe_2$), 3.82 (1 H, dd, *J* 6.76, 10.65, $CH_aH_bCHMe_2$) and 4.04 (2 H, q, *J* 7.14, $MeCH_2$); δ_C 13.84 (CH_2Me), 18.73 ($CHMe_2$), 27.46 ($CHMe_2$), 52.42 (COC), 61.76 (CH_2Me), 71.76 (CH_2CH), 165.49 (CO) and 165.56 (CO); *m/z* 217 [$(M + 1)^+$, 8%], 161 (55), 143 (100), 115 (42), 87 (16), 71 (20) and 57 (26); $[\alpha]_D^{24}$ 3.33 (*c* 3.2, $CHCl_3$); ≥97% ee as determined by ^1H NMR spectroscopy in the presence of the chiral solvating agent.

Transesterification of Diethyl (±)-2-*Hydroxy*-3-(4-*methylphenylsulfonyloxy*)*butane*-1,4-*dioate*.—In a typical procedure, a mixture of diethyl 2-hydroxy-3-(4-methylphenylsulfonyloxy)butane-1,4-dioate (1.98 g, 5.5 mmol), 2-methylpropan-1-ol (0.8 cm^3) and toluene-*p*-sulfonic acid (70 mg) in benzene (15 cm^3) was boiled under reflux under nitrogen. The reaction was monitored by TLC [light petroleum (b.p. 40–60 °C)–ethyl acetate (3:2, v/v)]. When under UV detection the amount of mixed ester present appeared to be greater than that of either the starting ester or the di-2-methylpropyl ester, the reaction was stopped. The mixture was concentrated under reduced pressure, the residue was dissolved in diethyl ether, washed with sodium hydrogen carbonate solution (55 cm^3) and water and dried (Na_2SO_4). The solvent was evaporated under reduced pressure and the residue was purified by flash chromatography [light petroleum–ethyl acetate (8:2, v/v)] to give the mixed diester (1.24 g, 58%). This ester was readily distinguishable from the regioisomeric ester **22** made *via* α-chymotrypsin catalysis, as above, on the basis of the ^{13}C NMR spectrum of the *tert*-butyldimethylsilyl derivative. Similar transesterifications were carried out on the (2*S*,3*S*)- and (2*R*,3*R*)-diesters which were converted into the corresponding epoxy diesters **30** and **31** (Scheme 6) as above, $[\alpha]_D$ −2.57 and 2.52, respectively.

Bis-(2-*methylpropyl*) (2S,3S)-2,3-*Dihydroxybutane*-1,4-*dioate* (*Diisobutyl* DL-*Tartrate*).—A solution of (D)-(−)-tartaric acid (3.97 g), 2-methylpropanol (8 cm^3) in benzene (16 cm^3) was boiled under reflux with a catalytic amount of toluene-*p*-sulfonic acid, using a Dean and Stark trap, overnight. The solution was concentrated under reduced pressure and the oily residue was distilled (Kugelrohr, 200 °C, 0.1 mmHg), to give bis-(2-methylpropyl) (2*S*,3*S*)-2,3-dihydroxybutane-1,4-dioate **24** (1.47 g, 21%) [Found: $(M + 1)^+$, 263.1490. $C_{12}H_{23}O_6$ requires 263.1494]; $[\alpha]_D^{26}$ −12.4 (*c* 1.04, $CHCl_3$); ν_{max}/cm^{-1} 1736; δ_H(400 MHz; $CDCl_3$) 0.91 (2 × 3 H, t, *J* 7.4, 2 × CH_2Me), 1.03 (2 H, d, *J* 6.04, $CHMe$), 1.18 (2 H, d, *J* 7.41, $CHMe$), 1.61 (2 × 2 H, m, 2 × CH_2Me), 3.50 (2 H, br s 2 × OH), 4.50 (2 H, s, $CHOH$) and 4.99 (2 H, m, 2 × $CHMe$); δ_C 9.38, 9.51 (2 × CH_2CH_3), 19.18, 19.36 (2 × $CHCH_3$), 28.57, 28.63 (2 × CH_2Me), 72.02, 72.11 (2 × COH), 74.87 (2 × CO_2CH) and 171.27 (2 × CO); *m/z* 263 [$(M + 1)^+$, 11%], 207 (18), 161 (48), 132 (29), 115 (16), 89 (23), 76 (65) and 57 (100). This ester could be tosylated and subjected to the acid-catalysed transesterification against ethanol as described above for the corresponding transesterification of the diethyl ester against 2-methylpropanol. The mixed ester was converted into the corresponding epoxide as described above, $[\alpha]_D$ 2.98.

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