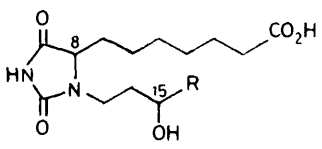
Heterocyclic Prostaglandin Analogues. Part 3.[1] The Relationship of Configuration to Biological Activity for Some Hydantoin Prostaglandin Analogues

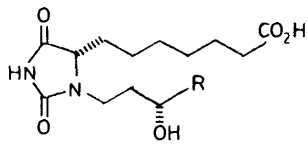
By **Michael A. Brockwell, A. Gordon Caldwell,** and **Norman Whittaker,*** The Wellcome Research Laboratories, Beckenham, Kent BR3 3BS
Michael J. Begley, Department of Chemistry, University of Nottingham, Nottingham NG7 2RD

The enantiomers of the two diastereoisomers of 5-(6-carboxyhexyl)-1-(3-cyclohexyl-3-hydroxypropyl)hydantoin have been synthesised. Potent inhibition of platelet aggregation in this series is associated with the configuration corresponding to that in the natural inhibitors, such as prostaglandins E_1 and D_2.

In Part 2 of this series [1] we described the synthesis of some hydantoin prostaglandin (PG) analogues (1; R = alkyl, branched alkyl, or cycloalkyl) having powerful PG-like biological activity. Each analogue was obtained as a mixture of two racemic diastereoisomers which were separated by h.p.l.c. For each variant of R, the diastereoisomers were distinguishable by proton n.m.r. spectroscopy and the chromatographically less polar compound was invariably the more potent inhibitor of platelet aggregation in human platelet-rich plasma. On the basis of this correlation of physical characteristics with biological potency, it is concluded that the less polar diastereoisomers in this series all have the same relative configuration at C-8 and C-15.† Their formulation as (±)-(2) follows from an *X*-ray crystal structure analysis (see later) of the less polar diastereoisomer of



(1)



(2)

(1; R = cyclohexyl). The relationship between absolute configuration and biological activity in this series has now been elucidated through total synthesis (Scheme) of the enantiomers of the two diastereoisomers of (1; R = cyclohexyl).

Attempts to resolve the amino-diester (3) as the (+)-tartrate in water at room temperature were unsuccessful. Crystals formed after a few days but these proved to be the α-amino-acid (4a), resulting from a slow, selective hydrolysis of the ester function α to the amino-group, presumably through activation by the $^{-}\overset{+}{N}H_3$ moiety. This finding did, however, suggest an alternative route to the required optically active intermediate (7). First, the α-amino-acid (4a) could be obtained in high yield simply by heating the amino-diester (3) with less than one equivalent of acetic acid in water at 100 °C for a few hours. The *N*-acetyl compound (4b) obtained from (4a) was then exposed to the action of porcine renal acylase I ‡ which liberated the (*S*)-amino-acid (5) in 86% yield but left the (*R*)-acetamido-compound (6) unchanged. Esterification of (5) with ethanol–thionyl chloride provided the (*S*)-amino-diester (7) almost quantitatively.

Following the general synthetic route already described,[1] reaction of (7) with cyclohexyl vinyl ketone and reduction of the amino-ketone (8) with sodium borohydride yielded a mixture of epimeric amino-alcohols. These were separated by h.p.l.c. on silica and the chromatographically more polar and less polar amino-alcohols were identifiable unequivocally as (9) and (10), respectively, from the following considerations. In the racemic series, reaction of the more polar amino-alcohol with cyanic acid gives rise to the hydantoin diastereoisomer now formulated as (±)-(2; R = cyclohexyl) on the basis of *X*-ray analysis (see later). Accordingly, that amino-alcohol must have the relative stereochemistry depicted in (9). Since the optically active more polar amino-alcohol was derived from the (*S*)-amino-acid (5), its absolute configuration is therefore represented by formula (9).

Treatment of the amino-alcohols (9) and (10) individually with cyanic acid gave, after hydrolysis, the (8*S*,15*R*)-§ and (8*S*,15*S*)-hydantoins (11) and (12), respectively. Finally, the hydantoins were separately exposed to an excess of 1N-aqueous sodium hydroxide at room temperature overnight and the resulting 1 : 1 mixtures of C-8 epimers were separated by h.p.l.c. In this way, the (8*R*,15*R*)- and (8*R*,15*S*)-species (13) and (14) were conveniently obtained without repetition of the whole synthesis using the amino-diester derived from the (*R*)-acetamido-acid (6).

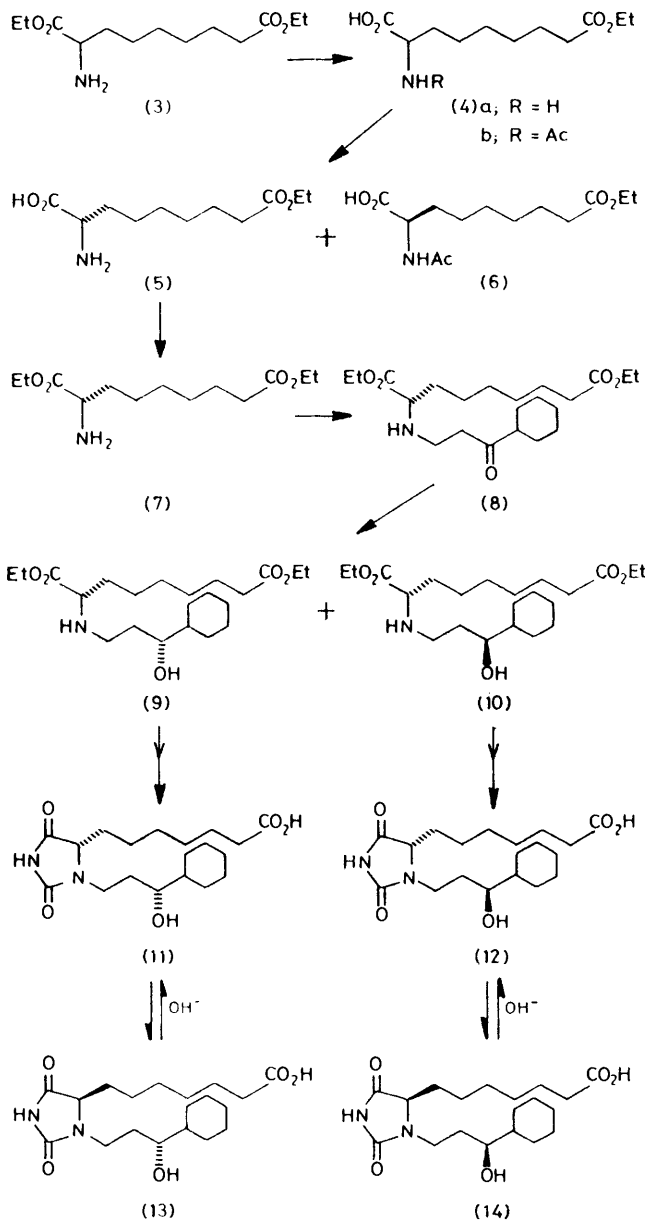
Compound (11) had *ca.* 40 times the potency of PGE_1 (0.75 × PGI_2) as an inhibitor of platelet aggregation in human platelet-rich plasma. In direct comparison, it was twice as potent as the racemic compound (±)-(11)

† Prostanoic acid numbering is used throughout the Discussion.

‡ Consideration of the rates of hydrolysis of *N*-acetylglycines substituted at the α-position by groups of varying lipophilicity [2] suggested that the ethoxycarbonylhexyl compound would be a better substrate than the corresponding carboxyhexyl compound. As expected, however, a substantial proportion of enzyme was required to achieve a reasonable reaction rate even with the former.

§ With cycloalkyl at C-15 in place of the pentyl moiety of the natural prostaglandins, the α-hydroxy-configuration is designated 15*R* according to the Sequence Rule.

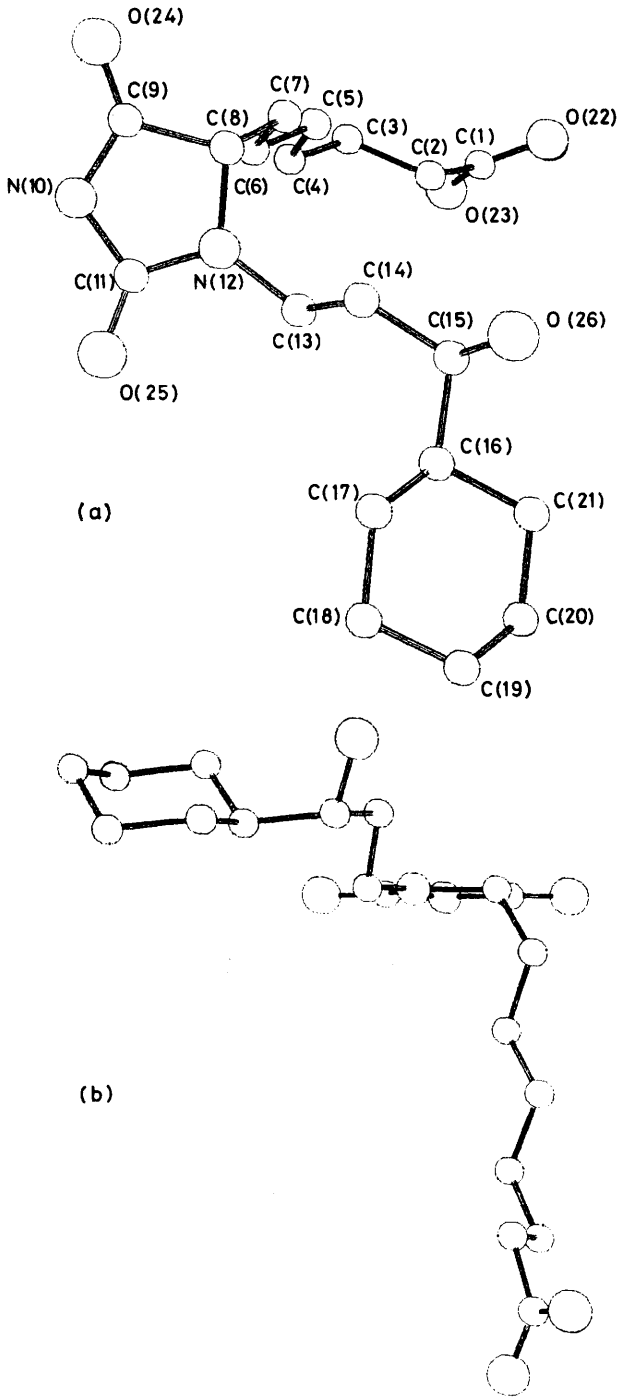
and, not surprisingly therefore, its enantiomer (14) proved to be inactive. Compound (12) and its enantiomer (13) were, like their racemic mixture, only weakly active (0.05 × PGE_1) inhibitors of platelet aggregation.



SCHEME

Since potent anti-aggregatory activity is associated with the (8*S*,15*R*)-configuration, it appears that the mode of action of compound (11) is similar to that of the natural PGs, such as PGE_1 and PGD_2, which are configurationally equivalent to (11) at C-8 and C-15. Interestingly, where the 'natural' chirality is present only at C-8 or C-15, as in (12) and (13), respectively, weak but significant biological activity is retained, implying some binding to the receptor in question. In the case of the biologically inactive (14), where both C-8 and C-15 have the unnatural configuration, receptor binding apparently does not occur.

X-*Ray Crystal Structure Analysis of the Less Polar Diastereoisomer of* (1; R = *cyclohexyl*).—The structure was solved from diffractometer data by direct methods, and refined by least-squares and difference-Fourier methods to R 8.06%, with anisotropic temperature



Two views of the molecule of the less polar diastereoisomer of (1; R = cyclohexyl); (a) shows the crystallographic atomic numbering. Hydrogen atoms are omitted

factors, and including hydrogen atoms without refinement.

The structure revealed the relative configuration at C(8) and C(15) to be that formulated as (±)-(2). Two general views of the molecule are shown in the Figure, in and perpendicular to the plane of the heterocyclic ring. Bond lengths, bond angles, and torsion angles are displayed in Table 1 and are unexceptional. Both nitrogen atoms are planar [although N(10) bonded to hydrogen is less well defined] presumably due to their amide nature, reflected in the relatively short C(9)–N(10) and C(11)–N(12) bond lengths. The heterocyclic ring is also planar, while the cyclohexyl ring adopts the expected chair conformation. In the α-chain the C(3)–C(4) bond is, surprisingly, in the *gauche* conformation. The overall conformation of the molecule, clearly revealed in the Figure, is L-shaped with the α and ω side-chains roughly perpendicular to one another. This unusual conformation is similar to that found in PGB_1 [3] and is in contrast to the more usual ' hairpin ' or approximately parallel disposition of side-chains observed for other prostaglandins.[4] These conformations may well be peculiar to the solid state as the crystal structure revealed three intermolecular hydrogen bonds [N(10)–O(25′) 2.81 Å, O(23)–O(26′) 2.63 Å, and O(26)–O(22′) 2.70 Å].

EXPERIMENTAL

Bio-sil silica (20—44 μ) was used for high performance liquid chromatography (h.p.l.c.); for the hydantoins, elution was monitored at 250 nm by means of a Cecil Instruments CE 212 u.v. detector. Porcine renal acylase I was obtained from Miles Laboratories. For measurements of specific rotation and o.r.d., the concentration (*c*) is expressed in g per 100 ml; o.r.d. data are cited for solutions in dioxan.

TABLE 1

Bond lengths, bond angles, and torsion angles in the less polar diastereoisomer of (1; R = cyclohexyl)

(*i*) Bond lengths (Å)

C(1)–C(2)	1.492(9)	C(11)–N(12)	1.330(7)
C(1)–O(22)	1.170(7)	C(11)–O(25)	1.220(6)
C(1)–O(23)	1.285(8)	N(12)–C(13)	1.455(7)
C(2)–C(3)	1.494(10)	C(13)–C(14)	1.524(8)
C(3)–C(4)	1.521(10)	C(14)–C(15)	1.508(8)
C(4)–C(5)	1.493(10)	C(15)–C(16)	1.525(8)
C(5)–C(6)	1.528(9)	C(15)–O(26)	1.445(7)
C(6)–C(7)	1.515(9)	C(16)–C(17)	1.519(8)
C(7)–C(8)	1.507(9)	C(16)–C(21)	1.524(8)
C(8)–C(9)	1.518(8)	C(17)–C(18)	1.545(9)
C(8)–N(12)	1.467(7)	C(18)–C(19)	1.507(10)
C(9)–N(10)	1.334(7)	C(19)–C(20)	1.503(10)
C(9)–O(24)	1.209(7)	C(20)–C(21)	1.527(9)
N(10)–C(11)	1.386(7)		

(*ii*) Bond angles (°)

C(2)–C(1)–O(22)	122.8(7)	N(12)–C(11)–O(25)	127.4(5)
C(2)–C(1)–O(23)	113.9(6)	C(8)–N(12)–C(11)	111.5(4)
O(22)–C(1)–O(23)	123.3(7)	C(8)–N(12)–C(13)	123.7(4)
C(1)–C(2)–C(3)	113.6(6)	C(11)N(12)–C(13)	124.7(5)
C(2)–C(3)–C(4)	112.9(6)	N(12)–C(13)–C(14)	112.0(5)
C(3)–C(4)–C(5)	114.8(6)	C(13)–C(14)–C(15)	112.4(5)
C(4)–C(5)–C(6)	113.0(6)	C(14)–C(15)–C(16)	115.3(5)
C(5)–C(6)–C(7)	111.5(5)	C(14)–C(15)–O(26)	106.8(4)
C(6)–C(7)–C(8)	114.1(5)	C(16)–C(15)–O(26)	111.5(4)
C(7)–C(8)–C(9)	114.5(5)	C(15)–C(16)–C(17)	114.0(5)
C(7)–C(8)–N(12)	113.5(5)	C(15)–C(16–C(21)	110.4(5)
C(9)–C(8)–N(12)	101.1(4)	C(17)–C(16)–C(21)	110.6(5)
C(8)–C(9)–N(10)	107.0(5)	C(16)–C(17)–C(18)	111.4(6)
C(8)–C(9)–O(24)	125.3(6)	C(17)–C(18)–C(19)	110.8(5)
N(10)–C(9)–O(24)	127.6(5)	C(18)–C(19)–C(20)	111.1(6)
C(9)–N(10)–C(11)	112.4(5)	C(19)–C(20)–C(21)	111.1(6)
N(10)–C(11)–N(12)	108.0(5)	C(16)–C(21)–C(20)	112.7(5)
N(10)–C(11)–O(25)	124.6(5)		

TABLE 1 (*continued*)

(*iii*) Torsion angles (°)

O(22)–C(1)–C(2)–C(3)	127.0
C(2)–C(3)–C(4)–C(5)	64.6
C(5)–C(6)–C(7)–C(8)	−174.8
C(7)–C(8)–C(9)–N(10)	123.0
N(12)–C(8)–C(9)–O(24)	178.4
C(9)–C(8)–N(12)–C(11)	0.2
O(24)–C(9)–N(10)–C(11)	−178.9
N(10)–C(11)–N(12)–C(8)	−0.9
O(25)–C(11)–N(12)–C(13)	4.5
N(12)–C(13)–C(14)–C(15)	−173.9
C(14)–C(15)–C(16)–C(17)	−52.5
O(26)–C(15)–C(16)–C(21)	−55.7
C(15)–C(16)–C(21)–C(20)	−179.8
C(17)–C(18)–C(19)–C(20)	−57.4
O(23)–C(1)–C(2)–C(3)	−54.1
C(3)–C(4)–C(5)–C(6)	−178.1
C(6)–C(7)–C(8)–C(9)	−57.9
C(7)–C(8)–C(9)–O(24)	−59.2
C(7)–C(8)–N(12)–C(11)	−122.9
C(9)–C(8)–N(12)–C(13)	176.2
C(9)–N(10)–C(11)–N(12)	1.3
N(10)–C(11)–N(12)–C(13)	−176.9
C(8)–N(12)–C(13)–C(14)	64.1
C(13)–C(14)–C(15)–C(16)	−60.6
C(14)–C(15)–C(16)–C(21)	−177.7
C(15)–C(16)–C(17)–C(18)	−178.6
C(17)–C(16)–C(21)–C(20)	53.1
C(18)–C(19)–C(20)–C(21)	56.5
C(1)–C(2)–C(3)–C(4)	172.8
C(4)–C(5)–C(6)–C(7)	175.3
C(6)–C(7)–C(8)–N(12)	57.5
N(12)–C(8)–C(9)–N(10)	0.6
C(7)–C(8)–N(12)–C(13)	53.2
C(8)–C(9)–N(10)–C(11)	−1.2
C(9)–N(10)–C(11)–O(25)	180.0
O(25)–C(11)–N(12)–C(8)	−179.5
C(11)–N(12)–C(13)–C(14)	−120.3
C(13)–C(14)–C(15)–O(26)	174.9
O(26)–C(15)–C(16)–C(17)	69.5
C(21)–C(16)–C(17)–C(18)	−53.5
C(16)–C(17)–C(18)–C(19)	56.2
C(19)–C(20)–C(21)–C(16)	−54.7

Diastereoisomers of Racemic Diethyl 2-(3-Cyclohexyl-3-hydroxypropylamino)nonanedioate [(±)-(9) *and* (±)-(10)].—(a) *Separation*. The mixture of isomers was prepared by the general method previously described,[1] starting from diethyl 2-aminononanedioate and 1-cyclohexylprop-2-en-1-one; on t.l.c. the diastereoisomers gave overlapping elliptical spots, R_F 0.33 and 0.43 (SiO_2; $CHCl_3$–MeOH, 50 : 1 v/v). A solution of the mixture (10 g) in ethyl acetate (50 ml) was treated with 2.6N-ethereal hydrogen chloride (12 ml), followed by ether (100 ml), and the mixture was kept at 5 °C overnight. The resulting fine solid, which was difficult to filter, was isolated by centrifugation (15 min at 4 000 r.p.m.) and recrystallised three times from ethyl acetate to give small shining plates (3.6 g), m.p. 117—118 °C, of the *hydrochloride* of the less polar diastereoisomer of *diethyl 2-(3-cyclohexyl-3-hydroxypropylamino)nonanedioate* (Found: C, 60.7; H, 9.4; N, 3.3; Cl, 8.4. $C_{22}H_{42}ClNO_5$ requires C, 60.6; H, 9.7; N, 3.2; Cl, 8.15%). Evaporation of the centrifugation-liquors left a viscous oily mixture (5.8 g) of diastereoisomers in which the more polar predominated; it slowly solidified. A portion, on repeated recrystallisation

from ethyl acetate, gave the pure *hydrochloride* as colourless needles, m.p. 64—66 °C (Found: C, 60.45; H, 9.8; N, 3.1; Cl, 8.2%).

(b) *Reactions with Cyanic Acid.* (i) Treatment of the hydrochloride of the less polar diastereoisomer with potassium cyanate followed by ester hydrolysis [1] gave the more polar diastereoisomer of 5-(6-carboxyhexyl)-1-(3-cyclohexyl-3-hydroxypropyl)hydantoin (1; R = cyclohexyl) as needles (from ethyl acetate), m.p. 124—126 °C, R_F 0.55 (SiO_2; $CHCl_3$–MeOH–HOAc, 90 : 5 : 5 v/v/v).

(ii) Similarly, the crude more polar diastereoisomer yielded predominantly (83%, by h.p.l.c.) the less polar diastereoisomer of the hydantoin (1; R = cyclohexyl). After h.p.l.c. (SiO_2; CH_2Cl_2–MeOH–HOAc, 96.5 : 2.5 : 1.0 v/v/v), this compound crystallised from ethyl acetate–light petroleum (b.p. 60—80 °C) as small needles, m.p. 96—98 °C, R_F 0.60 (SiO_2; $CHCl_3$–MeOH–HOAc, 90 : 5 : 5 v/v/v). In another such purification of the less polar hydantoin diastereoisomer, a much less soluble form was obtained; recrystallisation, by concentration of a solution in hot ethyl acetate to small bulk, with seeding, gave aggregates of prisms, m.p. 129—131 °C (Found: C, 61.95; H, 8.8; N, 7.5. Calc. for $C_{19}H_{32}N_2O_5$: C, 61.95; H, 8.75; N, 7.6%). This form was convertible by recrystallisation, with appropriate seeding, into the other form, m.p. 96—98°C.

2-*Amininononanedioic Acid* 9-*Ethyl Ester* (4a).—Diethyl 2-aminononanedioate [5] (50 g) was heated with water (100 ml) and acetic acid (6 ml, 0.54 mol. equiv.) on a steam-bath for 6 h and the resulting suspension of colourless crystals was set aside at room temperature overnight and filtered, affording the *amino-acid* (4a) (26.05 g), m.p. 214—216 °C (efferv.). Concentration of the liquors to 30 ml and heating for 2.5 h gave a further 10.12 g of the same product. A sample, recrystallised from water, had m.p. 221—222 °C (efferv.) (Found: C, 57.15; H, 9.35; N, 5.8. $C_{11}H_{21}NO_4$ requires C, 57.1; H, 9.15; N, 6.05%), *m/e* (e.i.) 186 ($M^+ - CO_2H$, base peak).

2-*Acetamidononanedioic Acid* 9-*Ethyl Ester* (4b) *and its Exposure to Porcine Renal Acylase I.*—A well stirred suspension of the amino-acid (4a) (26.05 g) in acetic acid (78 ml) was treated dropwise during 1 h with acetic anhydride (12.76 ml, 1.2 mol. equiv.). After a further 3 h, the reaction solution was stirred with water (250 ml) and ether (250 ml) for 1 h, and the mixture was then shaken with more water and ether. The ethereal phase was separated, washed with water, dried ($MgSO_4$), and evaporated; the residual oil was freed from traces of acetic acid by distillation with carbon tetrachloride *in vacuo* and finally set aside in ether (50 ml) at 0 °C to give 2-*acetamidononanedioic acid* 9-*ethyl ester* (4b) (19.12 g), m.p. 84—85.5 °C (Found: C, 57.1; H, 8.25; N, 5.0. $C_{13}H_{23}NO_5$ requires C, 57.1; H, 8.5; N, 5.15%).

A stirred suspension of compound (4b) (12.95 g) in water (390 ml) was treated dropwise with concentrated aqueous ammonia to pH 7—7.5 and the solution was filtered from sediment and diluted with water (97 ml). Porcine renal acylase I (0.162 g) and cobaltous acetate tetrahydrate (0.615 g) were added and the mixture was agitated for 1 h at 37 °C and then kept in a bath at 37 °C for 2 days; reaction was monitored * by t.l.c. (SiO_2; BuOH–HOAc–H_2O, 3 : 1 : 1 v/v/v), locating the liberated amino-acid with ninhydrin and the starting material by exposure to iodine vapour. After cooling to room temperature and filtration from sediment, the aqueous solution was treated with more acylase I (0.162 g) and maintained at 37 °C for an additional 2 days. The reaction mixture was treated with hydrogen sulphide and filtered from cobalt sulphide, and the aqueous solution was brought to pH 4 with acetic acid, washed with chloroform (7 × 900 ml), and evaporated *in vacuo*. The residual crystals were freed from traces of water by distillation with ethanol *in vacuo*, suspended in ethanol (*ca.* 10 ml), and collected, to give (S)-2-*amininononanedioic acid* 9-*ethyl ester* (5) (4.53 g), m.p. 235—237 °C (efferv.), $[\alpha]_D^{25}$ +23.5° (*c*, 1 in HOAc) (Found: C, 57.05; H, 8.85; N, 5.9%). Work-up of the alcoholic filtrate gave a further 0.18 g of (5).

The above chloroform washings were combined and evaporated and the residual oil was freed from chloroform by distillation with ether; the resulting solid was suspended in ether at 0 °C and collected, affording (R)-2-*acetamidononanedioic acid* 9-*ethyl ester* (6) (5.89 g), m.p. 105—106 °C, $[\alpha]_D^{27}$ −6.19° (*c*, 2 in EtOH). A recrystallised (EtOAc) sample gave m.p. 106.5—107.5 °C, $[\alpha]_D^{24}$ −6.48° (*c*, 2 in EtOH) (Found: C, 57.15; H, 8.45; N, 5.0%).

Diethyl (S)-2-*Amininononanedioate* (7).—Ethanol (16.3 ml) was stirred at −15 °C and treated dropwise with thionyl chloride (1.71 ml, 2.2 mol. equiv.); the (*S*)-amino-acid (5) (2.5 g) was then added in portions, keeping the temperature at −10 °C. The stirred suspension was allowed to come to room temperature and the resulting solution was set aside at room temperature for 16 h and poured on to ice–water containing concentrated aqueous ammonia (6.2 ml). The product was extracted into ether, the ethereal solution was concentrated *in vacuo* to *ca.* 50 ml, water (125 ml) was added, and the ether and water were evaporated *in vacuo*. The residual oil was freed from traces of ethyl sulphite by dispersion in water (2 × 125 ml) and evaporation of the water *in vacuo*, then taken into and recovered from ether to give *diethyl* (S)-2-*amininononanedioate* (7) † (2.6 g) as a colourless oil, R_F 0.25 (SiO_2; $CHCl_3$–MeOH, 50 : 1 v/v), $[\alpha]_D^{21}$ +16.3° (*c*, 1.02 in EtOH), $[\alpha]_D^{21}$ +13.2° (*c*, 1.02 in $CHCl_3$).

The Epimeric Amino-alcohols (9) *and* (10).—The (*S*)-amino-diester (7) (2.2 g) was treated with 1-cyclohexylprop-2-en-1-one (1.23 g, 1.05 mol. equiv.). After the mixture had been set aside at room temperature for 18 h, the resulting diethyl (2*S*)-2-(3-cyclohexyl-3-oxopropylamino)nonanedioate (8) was stirred in ethanol (35 ml) and treated with sodium borohydride (0.242 g, 0.75 mol. equiv.). After 2.5 h, the ethanol was evaporated *in vacuo* and the residue was cooled in ice–water and shaken with ether and water. The ethereal phase was separated, washed with water, dried ($MgSO_4$), and evaporated to leave a mixture of epimeric amino-alcohols. H.p.l.c. on silica, using chloroform–methanol as eluant, afforded *diethyl* (2S)-2-[(3R)-3-*cyclohexyl*-3-*hydroxypropylamino*]*nonanedioate* (9) (1.1 g), R_F 0.14 (SiO_2; $CHCl_3$), $[\alpha]_D^{22}$ +1.09° (*c*, 0.97 in EtOH), and *diethyl* (2S)-2-[(3S)-3-*cyclohexyl*-3-*hydroxypropylamino*]*nonanedioate* (10) (0.96 g), R_F 0.19 (SiO_2; $CHCl_3$), $[\alpha]_D^{22}$ −7.26° (*c*, 1.01 in EtOH), both as colourless gums.

Conversions into Hydantoins.—(a) *Amino-alcohol* (9). A stirred solution of the amino-alcohol (9) (1.1 g) in ethanol (5.5 ml) was cooled in ice–water and treated with 2N-hydrochloric acid (2.76 ml, 2 mol. equiv.) followed by a solution of potassium cyanate (0.447 g, 2 mol. equiv.) in

* In a control experiment, in which only the enzyme was omitted, no detectable hydrolysis occurred.

† An attempted resolution of the (±)-amino-diester (3) with *OO*-dibenzoyl-(+)-tartaric acid in aqueous acetic acid gave a salt which afforded an optically impure base, $[\alpha]_D^{17}$ + 3.4° (*c*, 1.02 in EtOH). Recrystallisation (EtOH) of the salt, prior to regeneration, diminished the specific rotation of the base.

water (1.35 ml). The turbid solution was set aside at room temperature overnight, the ethanol was evaporated *in vacuo*, and the residue was shaken with water and ether; the ethereal phase was separated, washed with water, dried ($MgSO_4$), and evaporated, to give an oil which was heated at 100 °C for 8.5 h. The resulting oil (1.107 g) was stirred with 0.5N-aqueous sodium hydroxide (11 ml, 2 mol. equiv.) at room temperature for 55 min and the turbid reaction solution was shaken with water (30 ml) and ether (30 ml); the aqueous phase was separated, washed with ether, and acidified with 1N-hydrochloric acid, and the liberated carboxylic acid was extracted into chloroform. The chloroform solution was washed with water, dried ($MgSO_4$), and evaporated, and the residual gum was freed from traces of chloroform by repeated addition of ether and evaporation in a current of nitrogen. The gum gave place to crystals which were suspended in a little ether at 0 °C and collected, and the crude product (0.848 g), double m.p. 121.5 and 126—128.5 °C, was purified by h.p.l.c. (SiO_2; CH_2Cl_2–MeOH–HOAc, 96.5 : 2.5 : 1.0 v/v/v). Further treatment with ether gave pure (5S)-5-(6-*carboxyhexyl*)-1-[(3R)-3-*cyclohexyl*-3-*hydroxypropyl*]*hydantoin* (11) (0.733 g), double m.p. 124 and 129—130 °C, $[\alpha]_D^{23}$ +22.56° (*c*, 1.02 in EtOH), o.r.d. $[\phi]_{400}$ + 223°, $[\phi]_{260}$ 0°, $[\phi]_{249}$ − 893°, $[\phi]_{241}$ 0°, $[\phi]_{230}$ + 3 125° (*c*, 0.247) (Found: C, 61.65; H, 8.9; N, 7.4. $C_{19}H_{32}N_2O_5$ requires C, 61.95; H, 8.75; N, 7.6%).

(b) *Amino-alcohol* (10). Reaction of the amino-alcohol (10) (0.956 g) with cyanic acid and work-up according to the procedure described in (a) above, gave (5S)-5-(6-*carboxyhexyl*)-1-[(3S)-3-*cyclohexyl*-3-*hydroxypropyl*]*hydantoin* (12) (0.548 g), m.p. 72.5—73 °C, $[\alpha]_D^{18}$ −17.0° (*c*, 1.03 in EtOH), o.r.d. $[\phi]_{400}$ − 203°, $[\phi]_{249}$ − 2 238°, $[\phi]_{235}$ 0°, $[\phi]_{230}$ + 1 627° (*c*, 0.181) (Found: C, 61.6; H, 9.05; N, 7.55%).

Epimerisations.—(a) *Hydantoin* (11). A solution of hydantoin (11) (0.4 g) in 1N-aqueous sodium hydroxide (6.52 ml, 6 mol. equiv.) was set aside at room temperature for 22 h, diluted with water (35 ml), and acidified with hydrochloric acid. The liberated carboxylic acid was extracted into chloroform and the chloroform solution was washed with water, dried ($MgSO_4$), and evaporated. The residual mixture of isomers was separated by h.p.l.c. (SiO_2; CH_2Cl_2–MeOH—HOAc, 97.5 : 2.0 : 0.5 v/v/v), yielding equal amounts of starting material and its more polar C-5 epimer. Treatment of the latter with ether at 0 °C gave (5R)-5-(6-*carboxyhexyl*)-1-[(3R)-3-*cyclohexyl*-3-*hydroxypropyl*]*hydantoin* (13) (0.159 g), m.p. 73—74.5 °C, $[\alpha]_D^{23}$ +16.93° (*c*, 1.0 in EtOH), o.r.d. $[\phi]_{400}$ + 238°, $[\phi]_{249}$ + 2 858°, $[\phi]_{237}$ 0°, $[\phi]_{230}$ − 1 905° (*c*, 0.155) (Found: C, 61.65; H, 9.05; N, 7.8%).

(b) *Hydantoin* (12). Treatment of hydantoin (12) (0.3 g) with aqueous sodium hydroxide, and work-up in the manner of (a) above, gave equal amounts of starting material and its less polar C-5 epimer. The latter, on exposure to ether, afforded (5R)-5-(6-*carboxyhexyl*)-1-[(3S)-3-*cyclohexyl*-3-*hydroxypropyl*]*hydantoin* (14) (0.118 g), double m.p. 123.5 and 129.5—130.5 °C, $[\alpha]_D^{19}$ −22.03° (*c*, 1.02 in EtOH), o.r.d. $[\phi]_{400}$ − 235°, $[\phi]_{267}$ 0°, $[\phi]_{249}$ + 784°, $[\phi]_{241}$ 0°, $[\phi]_{230}$ − 2 507° (*c*, 0.235) (Found: C, 62.15; H, 9.1; N, 7.7%).

Crystal Structure Determination.—The less polar diastereoisomer of (1; R = cyclohexyl), m.p. 129—131 °C, described above was used; a poorly shaped sample of dimensions *ca.* 0.6 × 0.5 × 0.3 mm, consisting of one major single crystal with minor satellites, was mounted and photographed. For intensity measurement the sample was transferred to a four-circle diffractometer. Unit-cell dimensions were refined by a least-squares fit on the diffractometer. With Mo-K_α radiation, intensity data were collected for $2\theta \leqslant 50°$ by use of an $\omega - 2\theta$ scan. 3 498 Reflections were measured of which 1 730 had $I \geqslant 3\sigma(I)$ and were used in the subsequent refinement. No absorption corrections were made. Data reduction and subsequent crystallographic calculations were performed using the 'CRYSTALS' system of programs.[6] Atomic scattering factors were taken from ref. 7.

TABLE 2

Atomic co-ordinates, with standard deviations indicated in parentheses

Atom	*x/a*	*y/b*	*z/c*
C(1)	0.410 6(6)	0.670(1)	0.031(1)
C(2)	0.368 7(6)	0.533(1)	0.057 7(9)
C(3)	0.339 6(7)	0.290(1)	−0.029(1)
C(4)	0.282 6(7)	0.145(1)	−0.020(1)
C(5)	0.360 7(6)	0.210(1)	0.164(1)
C(6)	0.298 6(6)	0.065(1)	0.167 2(9)
C(7)	0.377 0(5)	0.148(1)	0.356 3(8)
C(8)	0.316 7(5)	0.025 5(9)	0.368 5(8)
C(9)	0.269 1(5)	−0.230(1)	0.271 0(8)
N(10)	0.153 1(4)	−0.326 8(7)	0.154 2(6)
C(11)	0.114 6(5)	−0.168 3(9)	0.157 2(8)
N(12)	0.208 2(4)	0.037 7(7)	0.281 8(6)
C(13)	0.203 3(5)	0.249 6(9)	0.319 8(8)
C(14)	0.279 8(5)	0.401 2(9)	0.511 8(8)
C(15)	0.285 7(5)	0.631 2(9)	0.558 2(8)
C(16)	0.166 7(5)	0.627 9(9)	0.446 9(8)
C(17)	0.095 6(6)	0.493(1)	0.441 1(9)
C(18)	−0.022 2(6)	0.502(1)	0.330(1)
C(19)	0.000 7(7)	0.744(1)	0.402(1)
C(20)	0.068 1(8)	0.877(1)	0.404(1)
C(21)	0.184 7(6)	0.870(1)	0.511 0(9)
O(22)	0.499 6(5)	0.842 3(9)	0.148 7(8)
O(23)	0.339 2(6)	0.585(1)	−0.133 9(8)
O(24)	0.328 0(4)	−0.323 6(8)	0.300 3(6)
O(25)	0.012 4(3)	−0.215 3(6)	0.062 8(5)
O(26)	0.368 7(4)	0.764 2(6)	0.746 2(5)
H(2A)	0.432	0.602	0.189
H(2B)	0.295	0.536	0.007
H(3A)	0.414	0.286	0.031
H(3B)	0.283	0.226	−0.158
H(4A)	0.260	−0.017	−0.088
H(4B)	0.209	0.152	−0.078
H(5A)	0.435	0.204	0.223
H(5B)	0.384	0.375	0.233
H(6A)	0.282	−0.099	0.105
H(6B)	0.222	0.061	0.100
H(7A)	0.450	0.128	0.418
H(7B)	0.404	0.314	0.422
H(8)	0.384	0.115	0.504
H(10)	0.102	−0.493	0.086
H(13A)	0.233	0.335	0.298
H(13B)	0.118	0.216	0.233
H(14A)	0.363	0.420	0.597
H(14B)	0.244	0.324	0.529
H(15)	0.318	0.712	0.539
H(16)	0.116	0.534	0.318
H(17A)	0.144	0.567	0.569
H(17B)	0.081	0.332	0.396
H(18A)	−0.068	0.410	0.327
H(18B)	−0.072	0.421	0.201
H(19A)	0.049	0.822	0.526
H(19B)	−0.077	0.748	0.325
H(20A)	0.081	1.032	0.447
H(20B)	0.017	0.800	0.273
H(21A)	0.237	0.956	0.640
H(21B)	0.227	0.957	0.506
H(23)	0.371	0.633	−0.200
H(26)	0.390	0.920	0.752

Crystal data. $C_{19}H_{32}N_2O_5$, $M = 368$. Triclinic, $a = 18.744(3)$, $b = 7.098(2)$, $c = 13.182(3)$ Å, $\alpha = 109.06(1)$, $\beta = 136.80(1)$, $\gamma = 94.21(1)°$, $U = 994.69$ Å^3, $D_c = 1.23$ g cm^{-3}, $Z = 2$. Space group $P\bar{1}$ from intensity statistics and subsequent refinement, μ(Mo-K_α) = 0.96 cm^{-1}.

Structure solution and refinement. The structure was solved, at the third attempt, by direct methods using the MULTAN program,[8] following changes in normalisation and choice of starting-set reflections. An **E** map based on the best set of phases revealed all 26 non-hydrogen atoms as the largest peaks. Atomic parameters were refined by full-matrix least-squares, initially isotropically and subsequently with anisotropic vibrations. A difference-Fourier synthesis revealed the approximate positions of all hydrogen atoms. The positions of hydrogen atoms bonded to carbon were then calculated accurately from bond length and angle considerations and, together with the remaining hydrogen atoms taken directly from the difference map, included in the structure-factor calculations without refinement. A weighting scheme based on a Chebyshev polynomial was adopted. Refinement converged to an R value of 0.080 6 after a total of 15 cycles, when the largest parameter shifts were 0.2 σ. A final difference-map showed no features >0.3 Å^{-3}. Final atomic co-ordinates are listed in Table 2; temperature factors and observed and calculated structure factors are listed in Supplementary Publication No. SUP 22940 (18 pp.).*

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