Enzymatic Resolutions of Cyclic Amino Alcohol Precursors[1]

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Racemic butyrates of *trans*-2-azido as well as *trans*-2-nitro and *trans*-2-cyano cycloalkanols were hydrolysed to the corresponding optically active alcohols with the aid of lipases from *Candida cylindracea* and *Pseudomonas* sp. respectively. These precursors of amino alcohols could be produced in optical yields ranging from 85 to >98% e.e. Differences in the catalytic behaviour of the two enzymes depending on ring sizes were observed.

Cyclic amino alcohols constitute important entities of many biologically active natural and synthetic products.[2] For instance, 2-aminocyclopentanol and 2-aminocyclohexanol are used as starting materials for carbocyclic nucleosides[3] or for bactericidal oxo quinolinecarboxylic acid derivatives.[4] Aminocyclohexanols are essential partial structures of many antibiotics (*e.g.* aminoglycoside antibiotics[5]) as well as of glucosidase inhibitors (*e.g.* valiolamine[6]). 2-Aminocyclohexanol itself has been used for the preparation of a carbocyclic muramyldipeptide analogue,[7] a potential immunostimulant. 2-Aminocyclo-oct-5-enol was used for the synthesis of several pyridohomotropanes,[8] potent nicotinic agonists. *trans*-2-Aminomethylcyclohexanol is a starting material for the synthesis of several fused heterocycles with potential pharmacological activity.[9] Three out of the four diastereoisomeric 2-dimethylaminomethylcyclohexyl benzoates exhibit local anaesthetic effects similar to lidocaine.[10]

In almost all cases racemates of the respective amino alcohols were employed for synthetic studies. There are only a few reports on optically enriched or optically pure derivatives.[11] Since enantiomers often show remarkable differences in biological effects,[12] simple methods for obtaining (preferably both) antipodes of this class of substances seem highly desirable.

In our ongoing studies on the use of enzymes in the preparation of optically active amino alcohols we recently developed a short and efficient access to (+)- and (−)-*trans*-2-aminocyclohexanol.[13] We now report on the extension of this method to other ring sizes and to other functionalities as precursors of the amino function. In terms of simplicity of preparations as well as economical considerations azido, cyano, and nitro groups were chosen as most suitable precursors of the amino functionality.

Results and Discussion

Synthesis of Starting Materials.—The azidoalkanols, with the exception of *trans*-2-azidocyclo-octanol (**13**), were prepared *via* the respective epoxides according to the literature.[14] Medium ring epoxides (ring size 8—12 carbon atoms) are known to be highly resistant to nucleophilic reagents and/or to give rise to transannular reaction products.[15] There is a procedure for the synthesis of (**13**) in the literature,[16] but for safety reasons, because it uses hydrogen azide, it is limited to only mg amounts of product. After several unsuccessful attempts we accomplished the conversion from 1,2-epoxycyclo-octane into (**13**) similarly to Swift *et al.*,[14] changing the solvent (diethylene glycol monomethyl ether) and the temperature (100 °C) together with an extended reaction time (1 week) in low yield (20%). In the synthesis of the unsaturated azido alcohol (**17**) we also obtained a by-product (*ca.* 10%), identified as 4-azidocyclohex-2-enol, which was separated by column chromatography. We also attempted the synthesis of *trans*-2-azidocyclopent-3-enol by the same method and isolated both the expected product and 4-azidocyclopent-2-enol. These substances were, however, extremely unstable and isomerized within short time to the mixture mentioned above. Nevertheless, when we subjected the mixture of the esters of these alcohols to enzymatic resolution the lipases seemed to hydrolyse the same enantiomer of each ester with about the same optical purity. In our limited attempts to prepare the *cis*-isomer of (**5**) by nucleophilic displacement of its triflate derivative,[17] we obtained a *cis*/*trans* mixture, which on esterification and subsequent enzymatic hydrolysis was shown to proceed in essentially the same optical yield as the pure *trans*-isomer (**5**). This was shown by the analysis of the n.m.r. spectra of the respective MTPA esters[18] of racemic and resolved *cis*/*trans* mixtures.

The nitro alcohol (**25**) was prepared by reduction of 2-nitrocyclohexanone according to the procedure of Dampawan *et al.*,[19] but with chromatographic separation of the resulting isomers [*trans* (75%), *cis* (10%)].

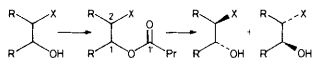
The cyano alcohol (**29**) according to a modified literature method,[20] was obtained from 1,2-epoxycyclohexane with hydrogen cyanide. In the ring opening reaction not only the desired *trans* nitrile (**29**), but *cis*-2-hydroxycyclohexanecarbonitrile and *trans*-cyclohexane-1,2-diol as by-products[21] were formed as well. The latters were separated from (**29**) by fractional crystallization.

The esterifications were carried out as described elsewhere in the literature.[22]

Enzymatic Hydrolysis.—As shown in a preliminary communication,[13] among the group of hydrolytic enzymes, lipases rather than esterases and proteases were most suitable for our purposes. We also found a significant preference for butyrates by most of the lipases tested, as compared to acetates or longer chain esters. In an initial screening, simply by t.l.c. monitoring, the following lipases were tested on our substrates (all enzymes were obtained in crude form as described in the experimental part and used without further purification).

Lipase from *Candida cylindracea* (CC), *Pseudomonas* sp. (P), *Aspergillus* sp. (A), and lipase from porcine pancreas (PPL). Whereas the first three enzymes showed good to excellent activity, the fourth showed little activity in hydrolysis of the esters under investigation. Testing for good enantioselection was carried out by recording the course of conversion *versus* time needed for hydrolysing 0.2 mmol ester in 0.02M phosphate buffer (pH 6.5 for CC, pH 7.0 for P and A).

Line P in Figure 1 showed good enantioselection by the enzyme for the tested substrate (**5**), the reaction rate slowing down significantly or even stopping when a conversion of 50% was reached. Line A showed poor enantiodifferentiation, since



(*R*, *R*)	(*S*, *S*)	R	X
(1)	(2)	(3)	(4)	$-(CH_2)_3-$	N_3
(5)	(6)	(7)	(8)	$-(CH_2)_4-$	N_3
(9)	(10)	(11)	(12)	$-(CH_2)_5-$	N_3
(13)	(14)	(15)	(16)	$-(CH_2)_6-$	N_3
(17)	(18)	(19)	(20)	$-CH{=}CH(CH_2)_2-$[a]	N_3
(21)	(22)	(23)	(24)	$-CHCH{=}CHCH-$	N_3
(25)	(26)	(27)	(28)	$-(CH_2)_4-$	NO_2
(29)	(30)	(31)	(32)	$-(CH_2)_4-$	CN

[a] 2-Azidocyclohex-3-enol

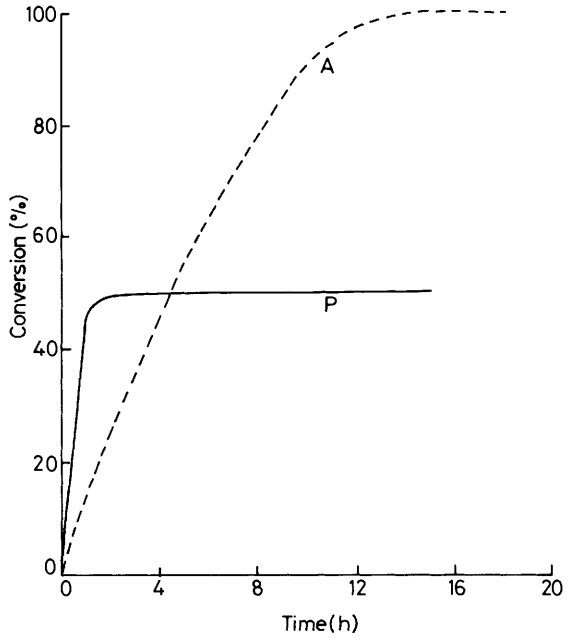


Figure 1. Activities of two different enzymes (P, A) in the hydrolysis of (**5**): —— P, -- A

there was no change in the reaction rate in passing the 50% limit. Lipase A showed no enantiodifferentiation with any of the substrates tested.

For a preparative enzymatic resolution of the substrates (**2**), (**6**), (**10**), (**14**), (**18**), (**22**), (**26**), and (**30**) using CC or P we used a procedure described earlier.[23] The course of hydrolysis was monitored by consumption of 1M NaOH and was stopped at an appropriate point to obtain an optimum in chemical and optical yield of products[24] (40% conversion for the alcohols and 60% for the corresponding *ent*-ester). The alcohol of the unconverted ester was obtained by methanolysis with catalytic amounts of sodium methoxide; compound (**26**) led to inseparable mixtures on this procedure, presumably *via aci*-forms. We accomplished the conversion of (*S*, *S*)-(**26**) into (**28**) by enzymatic hydrolysis with lipase A which showed no enantiodifferentiation. The pertinent results are summarized in the Table. While lipase from CC showed good activity and enantioselection on substrates (**6**), (**10**), (**14**), (**18**), (**22**), and (**26**), lipase Amano P proved to be more suitable for compounds (**2**) and (**30**).

The absolute configurations were determined by hydrogenation of the respective azido (**3**), (**4**), (**7**), and (**8**) alcohols to the corresponding amino alcohols or deduced from the ^{19}F n.m.r. spectra of the respective MTPA esters.[25]

Table Results of lipase-catalysed hydrolysis [a]

Product	$[\alpha]_D^{20}$ [b]	Yield (%)	E.e. [c] (%)	*E* [d]	Absolute config'n.
(3)	−12.5	40	14	1.4	(*R*, *R*)
(3) [e]	−78.1	40	92	45	(*R*, *R*)
(4)	+11.0	40	13	1.4	(*S*, *S*)
(4) [e]	+84.1	35	>98	168	(*S*, *S*)
(7)	−67.7	40	96	95	(*R*, *R*)
(8)	+68.8	35	>98	168	(*S*, *S*)
(11)	−42.6	40	89	31	(*R*, *R*)
(12)	+45.6	32	91	32	(*S*, *S*)
(15)	−62.0	40	93	52	(*R*, *R*)
(16)	+61.0	30	92	35	(*S*, *S*)
(19)	−67.0	40	>98	195	(*R*, *R*)
(20)	+64.2	29	94	47	(*S*, *S*)
(23)	−76.5	40	88	28	(*R*, *R*)
(24)	+85.0	29	>98	147	(*S*, *S*)
(27)	−49.7	40	>98	195	(*R*, *R*)
(28)	+43.3	20	85	15	(*S*, *S*)
(31)	−47.8	40	86	24	(*S*, *R*)
(31) [e]	−55.4	38	>98	183	(*S*, *R*)
(32)	+52.8	40	93	52	(*R*, *S*)
(32) [e]	+53.9	28	95	56	(*R*, *S*)

[a] If not stated otherwise done with lipase from *Candida cyclindracea*. [b] $c = 1$ in CH_2Cl_2. [c] Determined by integration of ^{19}F n.m.r. spectra of the respective (*S*)-MTPA-esters.[25] [d] Calculated from : $E = \ln[x(1 - \text{e.e.})]/\ln[x(1 + \text{e.e.})]$;[24] x = yield (%)/100. [e] *Pseudomonas* sp.

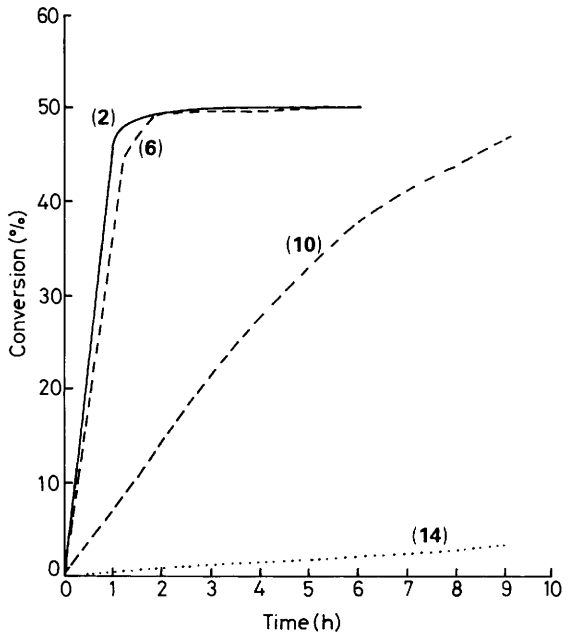


Figure 2. Activity of P depending on the ring size of the hydrolysed 2-azidocycloalkyl butyrates: (**2**), (**6**), (**10**), and (**14**)

Conclusions.—We have shown that several precursors of cyclic amino alcohols with potential biological importance can be obtained virtually in optically pure form in good yield by enzymatic resolution of the racemic butyrates.

The results given in Figure 2 indicate a significant decrease in reaction rate of hydrolysis by lipase P with increasing ring size. In contrast, the reaction rates of CC remained virtually constant with changing ring size, whereas enantioselection was very poor with (**2**) (see Table), being much better and invariable for (**6**), (**10**), and (**14**). Although we have no firm ideas on the size and structure of the active sites of P and CC, these results suggest a smaller cavity for P as compared to CC. The nature of

the functional group adjacent to the acyloxy groups being hydrolysed poses no limitations on this general method, at least in the cyclohexanol series, as can be seen by the fact that enantiodifferentiation, remains high and almost constant with a change in functionality from azido to nitro or cyano.

Experimental

M.p.s are uncorrected. Optical rotations were measured on a JASCO DIP-360 polarimeter in CH_2Cl_2 solution. N.m.r. spectra were recorded in $CDCl_3$ on a Bruker MSL 300 at 300 MHz (^1H), 75.47 MHz (^{13}C), and 282.27 MHz (^{19}F). Chemical shifts are in p.p.m. relative from TMS or $CFCl_3$ as internal standard. δ Values marked with an asterisk could be reversed. For numbering of carbon atoms in n.m.r. data see the Scheme. Reactions were monitored by t.l.c. using silica gel Merck $60F_{254}$ plates; purifications of products and separations of esters and alcohols after the enzymatic conversions were performed on silica gel Merck 60 with ethyl acetate–light petroleum as mobile phase. All esters were purified by bulb-to-bulb distillation before enzymatic hydrolysis.

All commercially obtained compounds were used as received. Crude enzyme preparations were employed without further purification. Lipases A and P are products from Amano Pharmaceutical Co., *Candida cylindracea* lipase and porcine pancreas lipase were obtained from Sigma Chem. Co.

General Preparation of the Racemic trans-2-*Azidocycloalkanols.*—A solution of 1,2-epoxycyclohexane (5.00 g, 50 mmol) in 80% aqueous ethanol (100 ml) was treated with NaN_3 (4.22 g, 65 mmol) and NH_4Cl (3.48 g, 65 mmol). This mixture was refluxed overnight and then poured into ice–water (300 ml). This solution was extracted thrice with CH_2Cl_2 (50 ml), and the combined extracts were dried (Na_2SO_4), and evaporated to afford a light brown oil. This was purified by vacuum distillation to give *trans*-2-azidocyclohexanol (**5**) (5.89 g, 82%) as a colourless oil, which slowly crystallized. B.p 98 °C/12 mmHg, m.p. 25—26 °C, n_D^{20} 1.4963 (lit.,[14] 70—75%, b.p. 62—4 °C/0.05 mmHg, m.p. 28.5—29.5 °C); δ_H 1.20—1.40 (4H, m, 4-H_2, 5-H_2), 1.70—1.80 (2H, m, 3-H_2), 2.00—2.10 (2 H, m, 6-H_2), 2.20 (1 H, br s, OH), 3.20 (1 H, m, 2-H), and 3.40 (1 H, m, 1-H); δ_C 22.9 (C-5)*, 23.3 (C-4)*, 29.9 (C-3), 33.2 (C-6), 67.1 (C-2), and 73.6 (C-1).

Other azido alcohols were prepared similarly.

trans-2-*Azidocyclopentanol* (**1**). This (4.45 g, 70%), had b.p. 94—95 °C/12 mmHg, n_D^{20} 1.4915 (lit.,[26] 64%, b.p. 72.5—74 °C/3 mmHg, n_D^{24} 1.4878); δ_H 1.35—1.75 (4 H, m, 3-H_2, 4-H_2), 1.75—2.05 (2 H, m, 5-H_2), 3.55 (1 H, m, 2-H), 3.90 (1 H, m, 1-H), and 3.95—4.00 (1 H, br s, OH); δ_C 20.2 (C-4), 28.3 (C-3), 31.8 (C-5), 66.4 (C-2), and 77.1 (C-1).

trans-2-*Azidocycloheptanol* (**9**). This (6.20 g, 80%), had b.p. 64—65 °C/0.15 mmHg, n_D^{20} 1.5019; δ_H 1.40—2.05 (10 H, m), 3.35 (1 H, m, 2-H), 3.38 (1 H, br s, OH), and 3.55 (1 H, m, 1-H); δ_C 22.7 (C-6)*, 23.4 (C-4)*, 27.2 (C-5) 29.4 (C-3), 33.1 (C-7), 70.2 (C-2), and 76.3 (C-1).

trans-2-*Azidocyclo-octanol* (**13**). Diethylene glycol monomethyl ether–water (80:20, v/v) was used as solvent, and the mixture was refluxed for a week (1.70 g, 20%), b.p. 98—99 °C/0.6 mmHg, m.p. 23—24 °C, n_D^{20} 1.5060 (lit.,[16] oil); δ_H 1.20—1.90 (12 H, m), 2.85—3.30 (1 H, br m, OH), 3.40 (1 H, m, 2-H), and 3.55 (1 H, m, 1-H); δ_C 24.1 (C-7)*, 24.5 (C-4)*, 25.9 (C-6)*, 26.0 (C-5)*, 28.5 (C-3), 32.2 (C-8), 68.9 (C-2), and 74.5 (C-1).

trans-2-*Azidocyclohex*-3-*enol* (**17**). After the reaction of 4,5-epoxycyclohexene[27] with NaN_3–NH_4Cl the product needed to be separated from 4-azidocyclohex-2-enol, which was a by-product with *ca.* 10%, by column chromatography with ethyl acetate–light petroleum (1:2) as mobile phase (2.37 g, 40%), b.p. 86 °C—87 °C/2 mmHg, n_D^{20} 1.5170 (Found: C, 51.6; H, 6.6 Calc. for $C_6H_9N_3O$: C, 51.8; H 6.5%); δ_H 1.40—2.20 (4 H, m, 5-H_2, 6-H_2), 3.55 (1 H, br s, OH), 3.65 (1 H, m, 2-H), 3.75 (1 H, m, 1-H), 5.50 (1 H, m, 4-H), and 5.90 (1 H, m, 3-H); δ_C 24.0 (C-5), 28.7 (C-6), 64.3 (C-2), 70.8 (C-1), 123.2 (C-4), and 131.5 (C-3).

trans-2-*Azidocyclohex*-4-*enol* (**21**). This (4.80 g, 81%), had b.p. 94—96 °C/6 mmHg, m.p. 17—18 °C, n_D^{20} 1.5150 (lit.,[28] 61—65%, m.p. <0 °C); δ_H 2.05—2.25 (2 H, m, 3-H_2), 2.40—2.60 (2 H, m, 6-H_2), 3.50—3.65 (m, 1 H, 2-H), 3.55—3.75 (1 H, br s, OH), 3.70—3.85 (1 H, m, 1-H), and 5.55—5.65 (2 H, m, 4-H, 5-H); δ_C 30.2 (C-3), 33.2 (C-6), 63.3 (C-2), 70.0 (C-1), 123.5 (C-5), and 124.8 (C-4).

trans-2-*Nitrocyclohexanol* (**25**). A solution of nitrocyclohexanone (7.16 g, 50 mmol) in ethanol (20 ml) was added dropwise to a suspension of $NaBH_4$ (1.89 g, 50 mmol) in ethanol (50 ml) and this was stirred overnight. The mixture was then neutralised with 10% HCl and extracted with ether (3 × 20 ml) and, after work-up, the products were isolated by column chromatography with ethyl acetate–light petroleum (1:4) as mobile phase to give 2-nitrocyclohexene (0.96 g, 15%), *cis*-2-nitrocyclohexanol (0.72 g. 10%), and (**25**) (3.04 g, 42%), b.p. 82—84 °C/1 mmHg, m.p. 48—49 °C (lit.,[29] m.p. 46—47 °C); δ_H 1.40 (2 H, m, 5-H_2), 1.85 (2 H, m, 4-H_2), 2.10 (2 H, m, 6-H_2), 2.30 (2 H, m, 3-H_2), 3.80—4.10 (1 H, br m, OH), 4.10 (1 H, m, 1-H), and 4.40 (1 H, m, 2-H); δ_C 23.6 (C-4)*, 23.8 (C-5)*, 30.6 (C-3), 33.1 (C-6), 71.3 (C-1), and 91.6 (C-2).

trans-2-*Hydroxycyclohexanecarbonitrile* (**29**). To a solution of NaCN (24.5 g, 0.5 mol) in water (300 ml) at −10 °C 20% aqueous HCl (95 ml, 0.52 mol) was added dropwise with stirring during 15 min. To this solution 1,2-epoxycyclohexane (20.2 ml, 0.2 mol) was slowly added with stirring at room temperature; the mixture was then left for a week at room temperature. After this, the deep brown mixture was treated carefully with solid CO_2 (50 g) and after 1 h the solvent was removed under reduced pressure and the residue extracted overnight with $CHCl_3$ (300 ml). The extract was evaporated and the light brown oil was distilled *in vacuo*, b.p. 108—110 °C/2 mmHg. (All operations were conducted in a fume cupboard provided with an efficient draught.) The distilled product needed to be separated from *cis*-2-hydroxycyclohexanecarbonitrile and *trans*-2-hydroxycyclohexanol, which were by-products (*ca.* 10%). After two recrystallizations from ether, pure (**29**) (15 g, 60%) was obtained, m.p. 49—51 °C (lit.,[20] m.p. 46—47 °C); δ_H 1.10—1.90 (6 H, m, 4-H_2, 5-H_2, 6-H_2), 2.00—2.25 (2 H, m, 3-H_2), 2.40—2.50 (1 H, m, 1-H), 3.50 (1 H, br s, OH), and 3.55 (1 H, m, 2-H); δ_C 23.5 (C-4), 24.0 (C-6), 28.3 (C-5), 33.9 (C-3), 37.5 (C-1), 70.4 (C-2), and 121.7 (C-7).

Preparation of Butyrates.—The esters were prepared by standard methods[22] and purified either by distillation or column chromatography.

trans-2-*Azidocyclohexyl butyrate.* A solution of (**5**) (7.00 g, 50 mmol) in absolute CH_2Cl_2 (100 ml), butyric anhydride (10.28 g, 65 mmol), pyridine (3.95 g, 65 mmol), and 4-dimethylaminopyridine (0.05 g) was mixed and left at room temperature until the conversion was complete (t.l.c., 16 h). Excess of anhydride was quenched by addition of methanol (2 ml). After 2 h the solution was washed with saturated $NaHCO_3$ (2 × 20 ml), 5% HCl (2 × 20 ml), and water (2 × 20 ml) and dried (Na_2SO_4). The mixture was then evaporated and the crude product was distilled to give (**6**) (9.50 g, 90%), b.p. 97—98 °C/0.7 mmHg, n_D^{20} 1.4673 (Found: C, 56.6; H, 8.2. Calc. for $C_{10}H_{17}N_3O_2$: C, 56.85; H, 8.1%); δ_H 0.96 (3 H, t, 4′-H_3), 1.20—1.40 (4 H, m, 4-H_2, 5-H_2), 1.60—1.70 (4 H, m, 3-H_2, 3′-H_2), 2.00—2.05 (2 H, m, 6-H_2), 2.30 (2 H, t, 2′-H_2), 3.40 (1 H, m, 2-H), and 4.65 (1 H, m, 1-H); δ_C 14.0 (C-4′), 18.4 (C-3′), 23.6 (C-4, C-5), 30.5 (C-3, C-6), 36.4 (C-2′), 63.3 (C-2), 75.2 (C-1), and 172.9 (C-1′).

The other esters were prepared similarly.

trans-2-*Azidocyclopentyl butyrate* (**2**). This (7.88 g, 80%), had b.p. 88—89 °C/2 mmHg, n_D^{20} 1.4510 (Found: C, 54.7; H, 7.8. Calc. for $C_9H_{15}N_3O_2$: C, 54.8; H, 7.7%); δ_H 0.75 (3 H, t, 4′-H_3), 1.40—2.00 (8 H, m), 2.05 (2 H, t, 2′-H_2), 3.70 (1 H, m, 2-H), and 4.80 (1 H, m, 1-H); δ_C 13.2 (C-4′), 18.1 (C-3′), 21.0 (C-4), 29.0 (C-5)*, 29.7 (C-3)*, 35.8 (C-2′), 66.1 (C-2), 78.9 (C-1), and 172.3 (C-1′).

trans-2-*Azidocycloheptyl butyrate* (**10**). This (10.70 g, 95%) had b.p. 109 °C/1 mmHg, n_D^{20} 1.4733 (Found: C, 58.2; H, 8.8. Calc. for $C_{11}H_{19}N_3O_2$: C, 58.6; H, 8.5%); δ_H 0.96 (3 H, t, 4′-H_3), 1.40—1.95 (12 H, m), 2.30 (2 H, t, 2′-H_2), 3.59 (1 H, m, 2-H), and 4.83 (1 H, m, 1-H); δ_C 13.4 (C-4′), 18.3 (C-3′), 22.4 (C-6)*, 23.3 (C-4)*, 27.7 (C-5)*, 30.0 (C-3)*, 30.8 (C-2′), 38.3 (C-7), 68.2 (C-2), 77.8 (C-1), and 172.3 (C-1′).

trans-2-*Azidocyclo-octyl butyrate* (**14**). This (9.73 g, 80%) had b.p. 122—123 °C/0.8 mmHg, n_D^{20} 1.4784 (Found: C, 59.9; H, 9.1. Calc. for $C_{12}H_{21}N_3O_2$: C, 60.2; H, 8.8%); δ_H 0.96 (3 H, t, 4′-H_3), 1.35—2.00 (14 H, m), 2.31 (2 H, t, 2′-H_2), 3.71 (1 H, m, 2-H), and 4.96 (1 H, m, 1-H); δ_C 13.5 (C-4′), 18.4 (C-3′), 23.5 (C-4)*, 25.0 (C-5)*, 25.5 (C-6)*, 25.8 (C-7)*, 28.2 (C-3), 30.8 (C-2′), 36.4 (C-8), 65.3 (C-2), 76.8 (C-1), and 172.6 (C-1′).

trans-2-*Azidocyclohex-3-enyl butyrate* (**18**). This (9.52 g, 91%) had b.p. 97—98 °C/1 mmHg, n_D^{20} 1.4795 (Found: C, 57.0; H, 7.5. Calc. for $C_{10}H_{15}N_3O_2$: C, 57.4; H, 7.2%); δ_H 0.90 (3 H, t, 4′-H_3), 1.50-1.75 (2 H, m, 3′-H_2), 1.90—2.35 (6 H, m, 2′-H_2, 5-H_2, 6-H_2), 3.90 (1 H, m, 1-H), 4.90 (1 H, m, 2-H), 5.50 (1 H, m, 4-H), and 5.90 (1 H, m, 3-H); δ_C 13.4 (C-4′), 18.3 (C-3′), 23.3 (C-5), 25.6 (C-6), 36.2 (C-2′), 60.1 (C-2), 72.2 (C-1′), 123.1 (C-4), 131.5 (C-3), and 172.7 (C-1′).

trans-2-*Azidocyclohex-4-enyl butyrate* (**22**). This (9.42 g, 90%) had b.p. 94—96 °C/1 mmHg, n_D^{20} 1.4789 (Found: C, 57.2; H, 7.6. Calc. for $C_{10}H_{15}N_3O_2$ C, 57.4; H, 7.2%); δ_H 0.95 (3 H, t, 4′-H_3), 1.65 (2 H, m, 3′-H_2), 2.10 (2 H, m, 3-H_2), 2.35 (2 H, t, 2′-H_2), 2.55 (2 H, m, 6-H_2), 3.70 (1 H, m, 2-H), 4.95 (1 H, m, 1-H), and 5.55 (2 H, m, 4-H, 5-H); δ_H 13.4 (C-4′), 16.3 (C-3′), 30.1 (C-3)*, 30.4 (C-6)*, 36.1 (C-2′), 59.4 (C-2), 71.7 (C-1), 123.5 (C-5), 124.0 (C-4), and 172.4 (C-1′).

trans-2-*Nitrocyclohexyl butyrate* (**26**). This (9.69 g, 90%) had b.p. 120—121 °C/1 mmHg, n_D^{20} 1.4580 (Found: C, 55.5; H, 8.3. Calc. for $C_{10}H_{17}NO_4$: C, 55.8; H, 8.0%); δ_H 0.88 (3 H, t, 4′-H_3), 1.20—1.50 (4 H, m, 4-H_2, 5-H_2), 1.55 (2 H, m, 3′-H_2), 1.70—1.95 (2 H, m, 6-H_2), 2.20 (2 H, t, 2′-H_2), 2.30—2.40 (2 H, m, 3-H_2), 4.50 (1 H, m, 1-H), and 5.20 (1 H, m, 2-H); δ_C 13.0 (C-4′), 18.1 (C-3′), 22.9 (C-5)*, 23.3 (C-4)*, 29.6 (C-6), 30.4 (C-3), 35.7 (C-2′), 72.1 (C-1), 87.6 (C-2), and 171.7 (C-1′).

trans-2-*Butyryloxycyclohexanecarbonitrile* (**30**). This (9.18 g, 94%) had n_D^{20} 1.4571 (Found: C, 67.3; H, 9.0. Calc. for $C_{11}H_{17}NO_2$: C, 67.7; H, 8.8%); δ_H 1.00 (3 H, t, 4′-H_3), 1.20—1.90 (8 H, m, 3-H_2, 4-H_2, 5-H_2, 3′-H_2), 2.00—2.20 (2 H, m, 6-H_2), 2.30—2.40 (2 H, m, 2′-H_2), 2.60—2.75 (1 H, m, 1-H), 4.85—4.95 (1 H, m, 2-H); δ_C 13.4 (C-4′), 16.4 (C-3′), 22.7 (C-4), 23.5 (C-6), 27.6 (C-5), 30.2 (C-3), 33.7 (C-1), 36.1 (C-2′), 71.1 (C-2), 119.7 (C-7), and 172.1 (C-1′).

Enzymatic Resolution of Racemic Esters with Lipases.—To a solution of lipase from *Candida cylindracea* (0.5 g) in phosphate buffer (0.1M; pH 6.5; 100 ml) was added (**6**) (5.00 g, 24 mmol). While vigorous stirring was maintained the pH was kept constant at pH 6.50 by addition of 1M NaOH from an autoburette. When the appropriate degree of conversion was accomplished [40% for (**7**)] the product was extracted with CH_2Cl_2. The extract was evaporated and subsequent column chromatography of the residue gave (**7**) (40%) and optically enriched (*S*, *S*)-(**6**). The latter was submitted to a further 20% conversion as described above. Subsequent work-up yielded (*S*, *S*)-(**6**) (35%). From that, (**8**) was obtained by methanolysis with catalytic amounts of NaOMe in 30% yield.

The other enzymatic resolutions were done in the same way. Results are shown in the Table.

Determination of the Optical Purity.—MTPA Esters were prepared according to the literature.[18] In accordance with literature data[25] all δ_F values of the (*S*)-MTPA-esters of (**3**), (**7**), (**11**), (**15**), (**19**), (**23**), (**27**), and (**31**) were upfield from their diastereoisomeric counterparts. In addition, comparison with the known values for the optical rotations of the amino alcohols resulting from (**3**), (**4**), (**7**), and (**8**) by hydrogenation clearly indicated the accuracy of our assignments of absolute configurations. Thus the known preference of lipase from CC for conversion of (*R*) esters[30] was substantiated.

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