Chiral Solute–Solvent Systems. Selective Interaction between *N*-Dodecanoyl-L-valine Amides and *N*-Trifluoroacetyl Esters of the Enantiomers of 2-Aminoalkan-1-ols and α-, β-, and γ-Amino-acids

By **Binyamin Feibush,* Ayala Balan, B. Altman,** and **E. Gil-Av,*** Department of Organic Chemistry, The Weizmann Institute of Science, Rehovot, Israel

Optically active 2-aminoalkan-1-ols in the form of their *O*-acyl-*N*-trifluoroacetyl derivatives have been resolved by g.l.c. with *N*-dodecanoyl-L-valine 6-undecylamide (1) as the stationary phase. A bulky *O*-acyl group substantially facilitates resolution. Resolutions of the *N*-trifluoroacetyl-esters of α-, β-, and γ-amino-acids on diamide phases are also reported. The order of emergence of the derivatives of the 2-aminoalkan-1-ols and the γ-amino-acids is D after the L-isomer, *i.e.*, the reverse of that found for the α-amino-acids.

The mechanism of resolution is discussed on the basis of association complexes involving the formation of two hydrogen bonds between the solutes and the solvent.

GAS CHROMATOGRAPHY with optically active stationary phases is known to lead to the resolution of several classes of compounds characterized by having an amido-group directly linked to an asymmetric carbon. In particular, the separation of enantiomeric *N*-trifluoroacetyl (*N*-TFA) derivatives of aliphatic and cyclic amines,[1,2] of α-amino-acid esters,[3,4] as well as of several β- and γ-amino-acid esters,[5] has been reported.

The first report on the resolution by g.l.c. of an amino-alcohol derivative, namely *O*-isobutyryl-*N*-TFA-valinol on *N*-TFA-L-valyl-L-valine isopropyl ester, was published in 1966.[3b] Even then, it had been realized that the bulkiness of the esterifying acid was critical and that the di-TFA derivative could not be separated into its enantiomers.† It was therefore, considered essential to investigate systematically the influence of the size of the groups,

TABLE 1

Resolution factors of *N*-trifluoroacetyl-*O*-acyl derivatives of aminoalkanols on *N*-dodecanoyl-L-valine 6-undecylamide [Phase (1)] as the stationary phase [a]

			Acyl group (COR')					
			Propionyl		Isobutyryl		Pivaloyl	
No.	Aminoalkanol		γ [b]	$\gamma_{D/L}$ [c]	γ [b]	$\gamma_{D/L}$ [c]	γ [b]	$\gamma_{D/L}$ [c]
(I)	2-Aminopropan-1-ol	L [d]	0.49	1.061	0.54	1.081	0.63	1.108
		D [d]	0.52		0.59		0.69	
(II)	2-Aminobutan-1-ol	L [e]	0.78	1.089	1.00	1.117	1.09	1.152
		D [e]	0.84		1.11		1.26	
(III)	2-Aminopentan-1-ol	L [e]	1.22	1.097	1.56	1.124	1.74	1.166
		D [e]	1.34		1.75		2.03	
(IV)	2-Aminohexan-1-ol	L [e]	2.02	1.094	2.56	1.123	2.83	1.160
		D [e]	2.21		2.86		3.28	
(V)	2-Aminoheptan-1-ol	L [e]	3.43	1.098	4.33	1.128	4.45	1.170
		D [e]	3.76		4.89		5.22	
(VI)	2-Amino-octan-1-ol	L [e]	5.90	1.098	7.47	1.129	7.53	1.170
		D [e]	6.48		8.44		8.81	
(VII)	2-Amino-3-methylbutan-1-ol	L [d]	0.93	1.105	1.00	1.138	1.11	1.181
		D [d]	1.02		1.14		1.32	
(VIII)	2-Amino-4-methylpentan-1-ol	L [d]	1.64	1.089	1.78	1.115	1.91	1.154
		D [d]	1.79		1.99		2.20	
(IX)	2-Amino-3,3-dimethyl-butan-1-ol [f]	L [e]					1.38	1.235
		D [e]					1.71	

[a] The column was a 150 ft × 0.02 in stainless-steel capillary connected to a F.I. detector; carrier gas He at 10 lb in^{-2}. Number of plates with respect to decyl acetate was 26 000. The γ values were not corrected for the optical purity (88.1%) of the phase.[4b] [b] Corrected relative retention time with respect to decyl acetate, which had a corrected retention time of 12 min at 140 °C, and 21 min at 130 °C. [c] $\gamma_{D/L}$ = resolution factor = ratio of the corrected retention time of the D- over that of the L-enantiomer, calculated with γ values expressed to the third decimal. [d] Peak assignment made by chromatography of enriched mixtures (L : D = 3 : 1). [e] Peak assignment made by extrapolation. [f] The *NO*-di-isobutyryl and the *NO*-dipivaloyl derivatives have at 140 °C a resolution factor of 1.174 and 1.125, respectively.

The present study is concerned with the resolution of amino-alkanols of formula $RCH(NH_2)CH_2OH$.[6] The purpose was to contribute to a better understanding of the selective solute–solvent interactions responsible for the separation on chiral stationary phases, as well as to demonstrate the validity of the method for additional classes of compounds.

† Being unaware of these facts led Parr and Howard [7] to draw incorrect conclusions from their work.

used for derivatization, on resolution. In the present study only the *O*-acyl moieties were modified, while, with two exceptions mentioned below, the amine function was throughout acylated by $(CF_3CO)_2O$.

As a stationary phase we employed *N*-dodecanoyl-L-valine 6-undecylamide [phase (1)] [4c] and for the amino-acid derivatives, in addition, *N*-dodecanoyl-L-valine t-butylamide [phase (2)].[4a]

The results are listed in Table 1. The quantity ε =

($r_{II/I} - 1$) *, which is useful for assessing the influence of structure on resolution, can be readily derived from the data in the Table. For compounds (I)—(IX), the ε values are larger by *ca.* 30% for the *O*-isobutyryl than for the *O*-propionyl derivatives, and increase by again as much for the *O*-pivaloyl-*N*-trifluoroacetyl-2-amino-alkan-1-ols. These strong effects of substituents should be kept in mind, whenever preliminary attempts of resolution are unsuccessful. A chromatogram showing the excellent peak resolution, which can be obtained, has been published previously.[8]

In the case of 2-amino-3,3-dimethylbutan-1-ol (IX) the *NO*-di-isobutyryl and the *NO*-di-isopivaloyl derivatives were also resolved. The latter, in comparison with the corresponding *O*-pivaloyl-*N*-TFA compound, had a considerably lower resolution factor ($r = 1.125$), as compared with $r = 1.235$ (Table 1, footnote *f*).

The most interesting observations made refer to the order of elution. Whereas the α-amino-acid derivatives emerge throughout in the sequence D before the L-isomer on both the L-diamides [4] and the L-dipeptide derivatives,[3] the reverse is true for the asymmetric amino-alkanols.† Since it was suspected that this difference in behaviour is linked to the distance of the amido-function from the ester group, β- and γ-amino-acid derivatives (Table 2) were also investigated. It was found, indeed, that the derivatives of the γ-amino-acids show a 'reversed' order of emergence like the 2-aminoalkan-1-ols, *i.e.* the retention of the D-isomers is larger than that of the L-isomers. On the other hand, the derivatives of the β-amino-acids have a variable behaviour depending on the particular diamide phase used (Table 2).

An incidental observation concerning the optical rotation of γ-amino-acids, though not related to the main objective of this study, is worth mentioning. It is known that the specific rotation of α- and β-amino-acids change consistently on acidification of the neutral aqueous solution for a given configuration. Thus, the L-amino-acids show a positive change,[10] and the L-β-compounds a negative one.[11] In contrast, for the γ-amino-acids studied, no consistent shift on acidification was observed. In fact, it was found that the $[\alpha]_D$ of γ-amino-δ-methylhexanoic acid (XV) and of γ-amino-ε-methylheptanoic acid (XVIII) reverse their sign on acidification of a neutral solution of each, though the compounds measured had the same configuration (see Experimental section). Acid shift of the rotation cannot, therefore, be used for the assignment of the configuration of γ-amino-acids.

Mechanism of Resolution.—Recently, we have started to consider the mechanism of the stereoselective interactions in diamide phases on the basis of known conformations and modes of intermolecular hydrogen bonding, as reported for diamides and peptides. Here we discuss briefly possible models of chiral recognition in the light of these literature data.

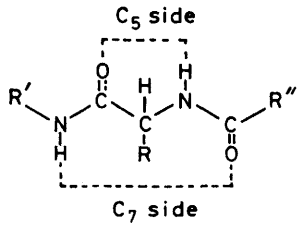
TABLE 2

Resolution factors of *N*-trifluoroacetyl isopropyl esters of some α-, β-, and γ-amino-acids on *N*-dodecanoyl-L-valine 6-undecylamide [phase (1)] and *N*-dodecanoyl-L-valine t-butylamide [phase (2)] as the stationary phase [a]

No.	Amino-acid		Phase 1 (t = 140°) r	Phase 1 (t = 140°) $r_{D/L}$	Phase 2 (t = 130°) $r_{D/L}$
(X)	$H_2N{\cdot}CH(Me){\cdot}CO_2H$	D L	0.17 0.18	0.924	0.797 [d]
(XI)	$H_2N{\cdot}CH(Me){\cdot}CH_2CO_2H$	L D	0.41 0.42	1.027	0.958 [e]
(XII)	$H_2N{\cdot}CH(Me){\cdot}[CH_2]_2{\cdot}CO_2H$	L D	1.02 1.07	1.053	1.094 [e]
(XIII)	$H_2N{\cdot}CH(CHMe_2){\cdot}CO_2H$	D L	0.29	1.00 [b]	0.816 [d]
(XIV)	$H_2N{\cdot}CH(CHMe_2){\cdot}CH_2{\cdot}CO_2H$	L D	0.84 0.86	1.023	0.928 [e]
(XV)	$H_2N{\cdot}CH(CHMe_2){\cdot}[CH_2]_2{\cdot}CO_2H$	L D	1.89 2.04	1.080	1.049 [e]
(XVI)	$H_2N{\cdot}CH(CH_2{\cdot}CHMe_2){\cdot}CO_2H$	D L	0.54 0.62	0.870	0.737 [d]
(XVII)	$H_2N{\cdot}CH(CH_2CHMe_2){\cdot}CH_2{\cdot}CO_2H$	L D	1.16 1.20	1.030	0.945 [e]
(XVIII)	$H_2N{\cdot}CH(CH_2CHMe_2){\cdot}[CH_2]_2{\cdot}CO_2H$	L D	3.37 3.62	1.077	1.047 [e]

[a] For chromatographic conditions, and the definition of $r_{D/L}$ and its calculation see footnotes in Table 1. [b] At 130 °C, $r_{D/L} = 0.966$. [c] Column 250 ft × 0.02 in; optical purity of (2), 75%. [d] Column 150 ft × 0.02 in; optical purity of (2), 75% (ref. 4*c*). [e] t = 120 °C.

The conformation and nature of the hydrogen bonding of diamides, analogous to phases (1) and (2), have been studied extensively.[12,13] The NH and CO functions of the *N*-acyl-α-amino-acid alkylamides can be grouped into two pairs, shown on opposing sides of the extended molecule and designated, respectively, 'C_5' and 'C_7'.



In the pleated sheet β-structure present in many peptides, *e.g.* in β-keratin, the interaction between different α-amino-acid units occurs through pairing of such 'C_5' and 'C_7' sides to form '$C_5 \cdots C_7$' * rings in

* ε, as defined here, is widely used in the literature dealing with isotope separation; $r_{II/I}$ = ratio of relative corrected retention time of the second peak over that of the first peak; see also ref. 5.

† It should be mentioned that on another chiral phase, namely carbonylbis-(*N*-L-valine isopropyl ester),[5] the amino-alkanol derivatives emerge in the order L- after the D-isomer.[9]

the parallel mode (Figure 1A), and ' $C_5 \cdots C_5$ ' and ' $C_7 \cdots C_7$ ' rings in the antiparallel mode of hydrogen bonding (Figure 1B). It is, further, particularly relevant to the present topic that *X*-ray analysis of a series of

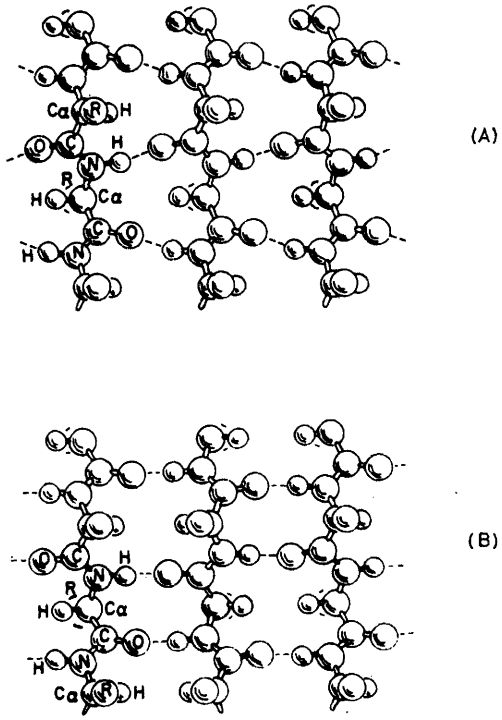
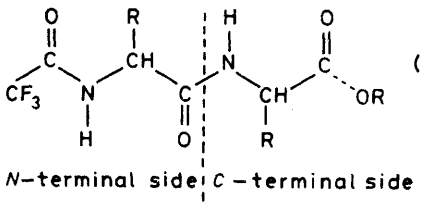


FIGURE 1 Pleated sheet structure of polypeptide chains. (A), parallel chains; (B) antiparallel chains

diamides of α-amino-acids [14] has demonstrated that the packing of molecules in the crystal shows intermolecular hydrogen-bonding motifs, as found in the pleated sheet β-structure. We propose to explain the selectivity observed in the present g.l.c. experiments by the consideration of associates involving the pairing of ' C_5 ' and ' C_7 ' sides of the chiral solute and solvent molecules.

α-Amino-acids.—The α-amino-acid derivatives [*e.g.* (X), (XIII), (XVI)] can assume the ' C_5 ' conformation only, and the interaction with the diamide can be either of the ' $C_5 \cdots C_7$ ' or the ' $C_5 \cdots C_5$ ' type. It is assumed that the ' $C_5 \cdots C_7$ ' association leads to higher stereoselectivity on the basis of the following argument.

For solvents of the dipeptide type (XIX) it has been found through variation of the configuration of the constituting moieties that stereoselectivity is essentially determined by the *N*-terminal amino-acid, whereas the *C*-terminal one makes a rather smaller contribution.



Examination of the structure reveals that the two chiral centres, around which association may occur, differ in that the one (*N*-terminal) possesses a ' C_7 ' as well as a ' C_5 ' side, whereas the other (*C*-terminal) has a ' C_5 ' side only.[15]

As can be seen in Figures 1A and 2, the amino-acid residues are parallel to each other (in translation) in hydrogen-bonded arrays of the ' C_5—C_7 ' type. As to the alkyl chains at the asymmetric centres, the orientation differs according to the relative configuration of the solute and the solvent. For the same configuration, the chains, too, are parallel. For the opposite configuration (see Figure 2) one has to interchange the positions of the hydrogen and the alkyl group at one of the asymmetric centres. The alkyl chains of the two associated molecules will, then, no more be parallel to each other (the ' non-parallel ' arrangement).

Without further experimental evidence and/or theoretical calculations, it is not possible to state whether the parallel or the ' non-parallel ' arrangement of the alkyl chains is the more stable one. However, as the experimental results show that the L-α-amino-acid derivatives are more strongly retained on the L-phase we assume that the parallel packing is more stable.

2-Aminoalkan-1-ols and γ-Amino-acids.—These solutes have only a ' C_7 ' side, (XX).

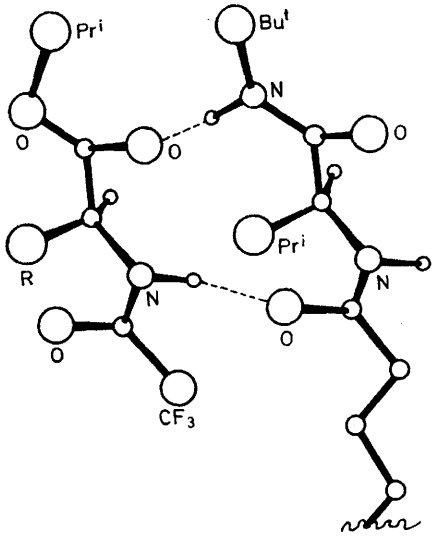


FIGURE 2 Schematic presentation of a ' $C_5 \cdots C_7$ ' hydrogen-bonded ring between an *N*-TFA-L-α-amino-acid isopropyl ester and *N*-dodecanoyl-L-valine t-butylamide (2)

The possible associations with the diamide phase are, therefore, of either the ' $C_7 \cdots C_5$ ' or the ' $C_7 \cdots C_7$ ' type. Conforming with the above, it is assumed that the ' $C_7 \cdots C_5$ ' association is the one which contributes most to stereoselectivity. There is, however, an important difference, namely: for the derivative of 2-aminoalkan-1-ols and γ-amino-acids, the ' $C_5 \cdots C_7$ ' association is of the antiparallel mode of hydrogen bonding as depicted in

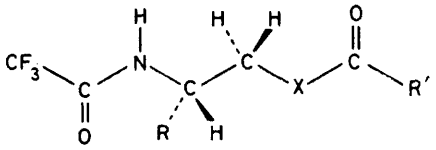
* For solute–solvent complexes the first symbol in this designation refers to the solute and the second to the solvent.

Figure 3 (and similar to that in Figure 1B). Such bonding leads to ' non-parallel ' arrangement of the alkyl groups attached to the asymmetric centres, if both molecules have the same configuration, and to a parallel orientation, if solute and solvent have opposite configuration. Correspondingly, the order of emergence of both the 2-aminoalkan-1-ol and γ-amino-acid derivatives are, indeed, the reverse of that observed for the α-amino-acids.

Recently,[16] we proposed a mechanism for the stereoselective interaction of monoamide solvents [*e.g.* (*s*)-*N*-acyl-α-(1-naphthyl)ethylamines] and solutes such as α-phenylethyl-*N*-TFA-amine and α-phenylbutyric acid *N*-t-butylamide, which have, respectively, reversed orders of emergence. The model was based on *X*-ray spectroscopic determination of the molecular packing of the solvent in the crystalline state, and the conclusions were checked by minimal energy calculations of the postulated selective solute–solvent associations. As in the cases discussed in this work, the reversal of the order of emergence can be readily explained through the requirement of a parallel, respectively, antiparallel hydrogen-bonded association of the two types of solutes with the chiral solvent.

β-*Amino-acids*. The *N*-TFA β-amino-acid esters show changes of the order of emergence depending on the particular diamide phase used for resolution. This behaviour is exceptional,[1-5] and requires further investigation. It should be pointed out that the β-amino-acid derivatives cannot form with the phase any of the hydrogen-bonded rings discussed previously, since they possess only a ' C_6 ' side. The possible '$C_6 \cdots C_5$' and '$C_6 \cdots C_7$' combinations may still, as above, determine, respectively, the order of emergence. One might, however, speculate that, contrary to the case of the α- and γ-amino-acids, there is no clear predominance of one of the types of hydrogen bonding over the other, and that the stability of the stereoselectively determined association depends on the individual solvent considered.

When comparing the different chiral phases known thus far, it is seen that there is a striking difference in selectivity for α-amino-acid derivatives, between the *N*-acyl-amino-acid esters,[3a] on the one hand, and the dipeptide [3c] and diamide derivatives on the other. It is



For 2-aminoalkan-1-ol derivatives: X = O, R′ = alkyl.
For γ-amino-acid derivatives: X = CH_2, R′ = O-alkyl.

felt that the higher efficiency of the two latter types of solvents is to be ascribed not only to the presence of a ' C_7 ' side (see above), but also to their capability of forming intermolecular hydrogen-bonded arrays as in the pleated sheet β-structure (Figure 1). Such structures should survive in part in the melt, in which the resolution is carried out. Indeed, it has recently [17] been demonstrated in our laboratory that chiral *N*-acyl-α-(1-naphthyl)ethylamine, which packs in translational stacks in the crystalline state, shows by *X*-ray photography the characteristic translational 5 Å distance also in the melt. The short-range order imposed through

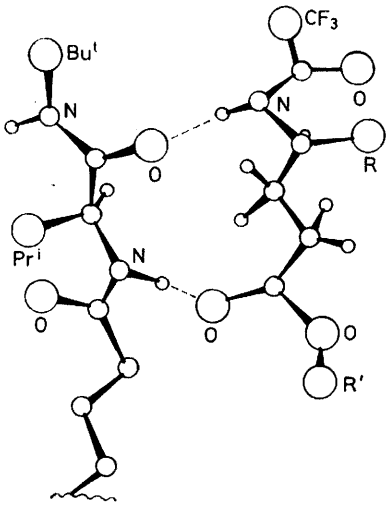


FIGURE 3 Schematic presentation of a '$C_7 \cdots C_5$' hydrogen bonded ring between *N*-TFA-L-γ-amino-acid ester and *N*-dodecanoyl-L-valine t-butylamide (2). To represent the interaction of the derivatives of 2-aminoalkan-1-ols, the α-methylene group of the solute has to be exchanged for an oxygen atom and the alkoxy-group for an alkyl radical

intermolecular solvent–solvent hydrogen bonding could, thus, stabilize those conformations, found in the crystalline state, which are taken to be responsible for chiral recognition.

The proposed mechanism of resolution no doubt oversimplifies a complex situation, and represents, at best, the most important of the possible stereoselective interactions. Its validity will be tested by studying the effect of appropriate structural modifications of both solutes and solvents, including, in particular, variation of the R substituent at the asymmetric carbon of the solvent, spectroscopic measurements, and theoretical computations. Whatever the merit of the ideas proposed, we feel that they constitute a useful working hypothesis for the further investigation of intermolecular interactions and the discovery of new selective solute–solvent systems.

EXPERIMENTAL

Materials.—*2-Aminoalkanols*. The following compounds were prepared according to Karrer *et al.*[18] by reduction with $LiAlH_4$ of the hydrochlorides of the methyl esters of appropriate α-amino-acids: (±)-alaninol [(±)-(I)], b.p. 45—48 °C/1.0 mmHg and L-alaninol [(+)-(I)], $[\alpha]_D^{23}$ +18.3° (*c*, 5 in EtOH) [lit.,[18] $[\alpha]_D^{17}$ +20.1° (in EtOH)]; (±)-2-aminobutan-1-ol (II), b.p. 45 °C/0.3 mmHg; (±)-2-aminopentan-1-ol (III), b.p. 50 °C/0.5 mmHg; (±)-2-aminohexan-1-ol (IV), b.p. 63 °C/0.5 mmHg; (±)-2-aminoheptan-1-ol (V), b.p. 71 °C/0.5 mmHg; (±)-2-amino-octan-1-ol (VI), b.p. 78—80 °C/0.5 mmHg; (±)-

valinol [(±)-(VII)], b.p. 52 °C/0.3 mmHg and L-valinol [(+)-(VII)], b.p. 55 °C/0.3 mmHg, $[\alpha]_D^{23}$ +15.7° (*c*, 5 in EtOH) [lit.,[19] $[\alpha]_D^{20}$ +15.6° (in ^{4}EtOH)]; (±)-leucinol [(±)-(VII)], b.p. 73 °C/1.0 mmHg and L-leucinol [(+)-(VIII)], b.p. 64—66 °C/0.3 mmHg, $[\alpha]_D^{23}$ +3.0° (*c*, 5 in EtOH) [lit.,[18] $[\alpha]_D^{18}$ +4.2° (*c*, 9 in EtOH)], g.l.c. of the *O*-pivaloyl-*N*-trifluoroacetyl derivative showed the presence of only 1.8% D-isomer; (±)-t-leucinol (IX). The structure of all compounds was confirmed by n.m.r. and i.r. spectra.

O-Acyl-N-trifluoroacetyl derivatives of the 2-aminoalkan-1-ols. A solution of amino-alcohol (200 mg) in benzene (10 ml) was saturated with dry gaseous HCl. The appropriate acyl chloride (0.5 ml) was added, and the mixture heated at 100 °C overnight in a sealed tube. The benzene was removed under reduced pressure, the residue dissolved in methylene chloride (10 ml), cooled, and *N*-trifluoroacetylated with trifluoroacetic anhydride (0.5 ml). The products were purified by extraction and analysed by g.l.c. *O*-pivaloyl-, *O*-isobutyl-, and *O*-propionyl-*N*-trifluoroacetyl-α-amino-alcohols were thus prepared.

β- and γ-Amino-acids. Racemic acids were synthesized by methods most apropriate for the compounds in question.

Optically enriched mixtures were prepared by resolution (XII), or by homologation of appropriate active α-amino-acids using the Arndt–Eistert reaction, which is known to proceed with conservation of configuration.

β-Aminobutyric acid (XI). Racemic (XI) was prepared by reduction of the acetylhydrazone of ethyl acetoacetate with aluminium amalgam.[20] L-β-Aminobutyric acid [(+)-(XI)] was synthesized by the Arndt–Eistert reaction following the procedure of Balenović, Cerar, and Fuks.[21] The compounds isolated in the intermediate steps had physical properties, including optical rotations, which were in excellent agreement with the literature,[21] *e.g.*, methyl *N*-phthaloyl-L-β-aminobutyrate, b.p. 144—145 °C/0.2 mmHg, $[\alpha]_D^{24}$ 29.4° (*c*, 6 in benzene) [lit.,[21] $[\alpha]_D^{16}$ +26.3° (*c*, 0.26 in benzene)]. The hydrochloride of L-(XI) (55 mg) isolated at the end of the synthesis showed, except for some minor impurity signals, the same n.m.r. spectrum as (±)-(XI): δ (in D_2O) 1.39 (d, CH_3, J = 6.5 Hz), 2.82 (d, CH_2, J = 6.5 Hz), and 3.82 (m, NC*H*–). The optical rotation at ambient temperature was $[\alpha]_D$ +13.5° (*c*, 4 in 6M-HCl) {lit.,[22] $[\alpha]_D^{20}$ +29.7° (c, 9.4 in 1M-HCl); $[\alpha]_D^{19}$ +37.07° (*c*, 6.0 in H_2O)}. This material, after conversion into its *N*-TFA isopropyl ester, permitted ready enantiomeric peak assignment, when co-injected with the corresponding derivative of racemic (XI).

γ-Aminovaleric acid (XII). Racemic (XII) was obtained in *ca.* 14% yield by the reductive amination of levulinic acid.[23] The active *N*-TFA isopropyl ester was prepared from L-(XI), synthesized according to Balenović *et al.*[21] In the intermediate steps the following compounds were isolated: *N*-phthaloyl-L-β-aminobutyric acid, $[\alpha]_D^{23}$ +7.7° (*c*, 6 in EtOH); *N*-phthaloyl-L-β-aminobutyryl chloride, m.p. 82—87 °C; and methyl L-*N*-phthaloyl-γ-aminovalerate, b.p. 129—131 °C/0.1 mmHg, $[\alpha]_D^{22}$ +31.8° (*c*, 5 in benzene). The latter compound (0.5 g) was hydrolysed with 6M-HCl (40 ml) for 5 h. The resulting crude hydrochloride L-(XII) was converted directly into the *N*-TFA isopropyl ester, which was purified by g.l.c., and had its structure confirmed by n.m.r.

In parallel a sample of active *N*-TFA isopropyl ester of D-(XII) was prepared from benzoyl-D-γ-aminovaleric acid obtained according to Fischer and Groh,[24] by adding quinine (1.02 g) to the *N*-benzoyl derivative of (±)-(XII) (0.64 g) dissolved in a mixture of hot ethanol (5.3 ml) and water (12 ml). The hot solution was filtered, and left for three days in the refrigerator. The precipitate of quinine salt [crop (i), m.p. 114—119 °C)] was recrystallized from a hot mixture of ethanol (2 ml) and water (5 ml). The crystals obtained (0.3 g) were dissolved in hot ethanol–water (1 : 7), cooled quickly, and decomposed with 1M-NaOH. The quinine liberated was filtered off, and the filtrate acidified with 5M-HCl in an ice-bath. The precipitate (13 mg) of *N*-benzoyl-L-aminovaleric acid had $[\alpha]_D^{18}$ −23.8° (*c*, 1 in EtOH) [lit.,[24] −21.9° (*c*, 10 in EtOH)].

The mother liquor was evaporated to give crop (ii), which was treated in the same manner as crop (i) above, yielding *N*-benzoyl-D-γ-aminovaleric acid (6 mg), $[\alpha]_D^{18}$ +5.70 (*c*, 5 in EtOH). The material was hydrolysed with 6M-HCl, and then converted into the *N*-TFA-isopropyl ester of D-(XII). The laevorotatory (in EtOH) *N*-benzoyl compound of (XII) has been correlated by Fischer *et al.*[24] with the dextrarotatory (in water) free amino-acid, and, subsequently, the latter has been shown to belong to the L-series.[25] The *N*-TFA-isopropyl ester, prepared from crop (ii), thus belongs to the D-series. By co-injection of the *N*-TFA-isopropyl ester of racemic (XII) with the above active esters, it was established that on both phases (1) and (2) the L- precedes the D-enantiomer.

β-Amino-γ-methylvaleric acid (XIV). Racemic (XIV) was prepared by addition of ammonia to 4-methylpent-2-enoic acid.[26] The active compound was synthesized from L-valine according to Balenović and Dvornik [27] (who started with the D-enantiomer). Physical data for the intermediate compounds are as follows: *N*-phthaloyl-L-valine, m.p. 115 °C, $[\alpha]_D^{24}$ −87° (*c*, 1 in EtOH) [lit.,[27] (for the D-enantiomer $[\alpha]_D$ +68° (in EtOH)]; *N*-phthaloyl-L-valyl chloride, m.p. 123 °C, $[\alpha]_D^{20}$ −103° (*c*, 1 in benzene) [lit.,[27] (D-enantiomer) $[\alpha]_D^{20}$ −104° * (*c*, in benzene)]; L-1-diazo-3-phthalimido-4-methylpentan-2-one (crude), $[\alpha]_D^{24}$ −81° (*c*, 1 in benzene); methyl β-phthalimido-γ-methylvalerate (crude), $[\alpha]_D^{24}$ +19° (*c*, 1 in benzene) [lit.[27] (D-enantiomer), $[\alpha]_D^{20}$ −6.27 (*c*, 2.24 in MeOH)]. In the final step the crude hydrobromide of (XIV) (8.8 g) was dissolved in water (1 200 ml) and passed through a column of Amberlite IR-4B. The free amino-acid obtained was recrystallized from EtOH–acetone (3.4 g), m.p. 205—206 °C, $[\alpha]_D^{24}$ +52.5° (*c*, 1 in H_2O) {lit.[27] (D-enantiomer), $[\alpha]_D^{24}$ −39.2° (*c*, 0.5 in H_2O)}.† The n.m.r. spectra of the hydrochlorides of both racemic and active (XIV) had δ (in D_2O): 0.99 [d, $CH(CH_3)_2$, J = 6.5 Hz], a multiplet centred at 1.99 [$CH(CH_3)_2$], a multiplet at 3.3 (NC*H*–); CH–CH_2CO_2H is the AB part of an ABX system: 2.53 (A, q, J_{AB} = 16 Hz, J_{AX} = 4.5

* The sign and absolute magnitude for the L-isomer have been confirmed by two workers in our laboratory on two different batches of L-(XIV). Also in all analogous cases, studied by us, the L-chloride had a more negative rotation than the corresponding *N*-phthaloyl-L-amino-acid. We, therefore, believe that the negative sign given by Balenović *et al.*[27] for the D-chloride is in error.

† The lower optical rotation reported in the literature for L-(XIV) could have been caused by racemization during the synthesis. Retention of configuration has been confirmed (by g.l.c.) in the homologation reactions reported here, except for one batch of L-(XI), which was racemic for unknown reasons. To avoid racemization, care has to be taken to adhere to the prescribed reaction conditions, and, where the degree of optical purity of the product is important, the enantiomeric composition must be checked. For deviations from conservation of configuration in the Arndt–Eistert reaction, see ref. 28.

Hz), and 2.45 (B, q, J_{BX} = 9.5 Hz) (Found: C, 54.75; H, 10.1. Calc. for $C_6H_{13}NO_2$: C, 54.9; H, 9.9%).

Chromatography of the *N*-TFA isopropyl esters of (XIV) enriched in the L-isomer showed that the L-isomer emerged first on phase (1), but last on phase (2).

γ-Amino-δ-methylhexanoic acid (XV). The racemic and the active compounds were synthesized by the procedure described above from (±)-(XIV) and L-(XIV) respectively. The successive reaction steps can be followed readily by characteristic signals in the n.m.r. spectra (δ, in $CDCl_3$) of the intermediate compounds: β-phthalimido-γ-methylvaleric acid (XIVa), 9.65, acidic proton CO_2H; β-phthalimido-γ-methylvaleroyl chloride (XIVb), b.p. 146/0.1 mmHg, absence of acidic proton; 1-diazo-5-methyl-4-phthalimidohexan-2-one (XIVc), 5.4 (s, C*H*-N_2); compounds (XIVa—c) possess an ABX system (N–CHR–CH_2–CO) showing two double doublets for CH_2 at 2.6—3.6; methyl γ-phthalimido-δ-methylhexanoate (XVa), 3.55 (CO_2CH_3) and a multiplet centred at 2.25 (CH_2–CH_2–CO_2H). In the synthesis of the active compound, the quantities formed and the optical rotations, $[\alpha]_D$,[24] measured were: L-(XIVa) (5.4 g) crude, +6° (*c*, 1.5 in EtOH); L-(XIVb) (4.6 g) crude, +1.1° (*c*, 1.2 in benzene); L-(XIVc) (5 g) crude, +112° (*c*, 1.05 in benzene); L-(XVa) (2.6 g) crude, +12° (*c*, 1 in benzene). Hydrolysis of L-(XVa) (1.5 g) with 6M-HCl (30 ml) yielded γ-amino-δ-methylhexanoic acid hydrochloride (0.18 g), m.p. 172—175 °C, +11.6° (*c*, 1.1 in H_2O), +6.5° (*c*, 1 in 6M-HCl), +11° (*c*, 1 in 10% NaOH). N.m.r. (D_2O) spectroscopy shows characteristic signals at 1.05 [d, $CH(CH_3)_2$, J = 5 Hz] and 2.6 (t, CH_2–CH_2–CO_2H) (Found: C, 46.5; H, 9.0; N, 7.6; Cl, 18.7. Calc. for $C_7H_{15}NO_2$·HCl: C, 46.28; H, 8.88; N, 7.7; Cl, 19.5%).

Chromatography of optically-enriched *N*-TFA isopropyl ester of (XVI) showed that the L- precedes the D-isomer on both phases (1) and (2) (Table 2).

β-Amino-δ-methylhexanoic acid (XVII). The racemic compound was prepared by addition of ammonia to 5-methylhex-2-enoic acid.[26] Active (XVII) was synthesized from L-leucine according to Balenović and Brovet-Keglević.[29] Physical data for the intermediate compounds are as follows: *N*-phthaloyl-L-leucine (40 g), m.p. 118—120 °C, $[\alpha]_D^{24}$ −29° (*c*, 1 in EtOH) {lit.,[29] $[\alpha]_D^{20}$ −22° (*c*, 5 in EtOH)}; *N*-phthaloyl-L-leucyl chloride, b.p. 132—133 °C/0.2 mmHg, $[\alpha]_D^{24}$ −41° (*c*, 2 in benzene); L-1-diazo-3-phthalimido-5-methylhexan-2-one (28 g); methyl L-β-phthalimido-δ-methylhexanoate (17.4 g), $[\alpha]_D^{24}$ +12.5° (*c*, 1 in benzene); hydrobromide of L-β-amino-δ-methylhexanoic acid (10 g, crude). The hydrobromide was dissolved in water (1 500 ml) and passed through a column of Amberlite IR-4B. The amino-acid was recrystallized from ethanol–acetone (2.6 g), m.p. 227 °C; $[\alpha]_D^{24}$ +27° (*c*, 1 in H_2O) [lit.,[29] +28° (*c*, 3 in H_2O)] (Found: C, 57.9; H, 10.4. Calc. for $C_7H_{15}NO_2$: C, 57.92; H, 10.3%). N.m.r. δ (in D_2O): 0.92 [d, $CH(CH_3)_2$, J = 6 Hz], a multiplet at 1.38—1.78 [$(CH_3)_2CH$–CH_2]; CH–CH_2–CO_2H is the AB part of an ABX system: 2.52 (A, q, J_{AB} = 16 Hz, J_{AX} = 3 Hz) and 2.43 (B, q, J_{BX} = 6 Hz); and a multiplet centred at 3.5 (NH_2CH).

Chromatography of the enriched mixture of *N*-TFA isopropyl ester of (XVII) showed that the L-enantiomer emerged first on phase 1 and last on phase 2 (see Table 2).

γ-Amino-ε-methylheptanoic acid.[30] (XVIII). Analogously to (XV), the racemic and active compounds were synthesized from (±)-(XVII) and L-(XVII), respectively.

Characteristic n.m.r. signals (δ, in $CDCl_3$) found for the intermediate compounds were: β-phthalimide-γ-methylhexanoic acid (XVIIa), +0.93 [d, $(CH_3)_2CH$, J = 5 Hz], signals at 2.55—3.45 (AB part of an ABX system, CH_2–CO_2H), and a signal for the acidic proton in the offset; β-phthalimido-γ-methylhexanoic acid chloride (XVIIb), signals at 3.2—3.96 (AB part of an ABX system, CH_2–COCl), and absence of a signal in the offset; 1-diazo-4-phthalimido-6-methylheptan-2-one (XVIIc), 5.35 (s, CO–C*H*–N_2); methyl γ-phthalimido-ε-methylheptanoate (XVIIIa), 3.58 (s, CO_2CH_3).

In the synthesis of the active compound, the quantities formed and the optical rotations, $[\alpha]_D^{24}$, measured were: L-(XVIIa) (2.8 g), m.p. 82—87 °C, +6.2° (*c*, 1 in EtOH); L-(XVIIb) (2 g), m.p. 63 °C, −1.2° (c, 1 in benzene); L-(XVIIc) (2 g, crude), +90° (*c*, 1.4 in benzene); L-(XVIIIa) (1.4 g, crude), +16° (*c*, 1.9 in benzene); hydrolysis of the latter compound (0.5 g) with 6M-HCl (10 ml) yielded the hydrochloride of L-(XVIII) (0.1 g), −8.7° (*c*, 1 in H_2O), −7.7° (*c*, 1.2 in 5M-HCl), −14.5° (*c*, 1 in 10% NaOH), {lit.,[30] free amino-acid $[\alpha]_{5800}^{20}$ −17.2° (*c*, 2 in H_2O)}; n.m.r. δ (in D_2O): 1.0 [d, $(CH_3)_2CH$, J = 6 Hz], 1.7 (m, CH–CH_2–CHN–), 2.07 (CH–CH_2–CH_2), 2.6 (t, CH_2–CH_2–CO_2H, J = 6 Hz), and 3.43 (m, NH_2–C*H*); the multiplet due to $CH(CH_3)_2$ is hidden by the assigned signals at 1.7 and 2.07 (Found: C, 49.25; H, 9.4; N, 7.05; Cl, 18.15. Calc. for $C_8H_{17}NO_2$·HCl: C, 49.1; H, 9.27; N, 7.16; Cl, 18.16%.

The order of emergence of the optical isomers of (XVIII) on phases (1) and (2) was the same as for (XV).

Physical Measurements.—The g.l.c. experiments were carried out on a Perkin-Elmer model 801 chromatograph adjusted to operate with capillary columns (stainless-steel, 150 ft × 0.02 in) and provided with a F.I.D. The optically active phases were *N*-dodecanoyl-L-valine 6-undecylamide (1), and *N*-dodecanoyl-L-valine t-butylamide (2) (Yeda Ltd., Rehovot, Israel). The coating was performed by the plug method with a 5% methylene chloride solution of the phase.

Optical rotations were measured with a Perkin-Elmer 1400 Instrument and the n.m.r. spectra taken on a Varian A 60 spectrophotometer or a Brucker HFX 10.

The authors are indebted to Drs. A. Hagler and L. Leiserowitz, The Weizmann Institute of Science, and Dr. G. Jung, University of Tuebingen, for useful discussions and to Mr. M. Lachmi, for assistance in the synthetic work.

The authors thank the Stiftung Volkswagenwerk for partial support of this research.

[8/1759 *Received*, 9th *October*, 1978]

REFERENCES

[1] B. Feibush and E. Gil-Av, *J. Gas Chromatography*, 1967, **5**, 257.

[2] J. A. Corbin and L. B. Rogers, *Analyt. Chem.*, 1970, **42**, 974.

[3] (*a*) E. Gil-Av, B. Feibush, and R. Charles-Sigler, in 'Gas Chromatography 1966,' ed. A. B. Littlewood, The Institute of Petroleum, 1967, p. 227; (*b*) *ibid.*, p. 235; (*c*) B. Feibush and E. Gil-Av, *Tetrahedron*, 1970, **26**, 1361; (*d*) S. Nakaparksin, P. Birrell, E. Gil-Av, and J. Oró, *J. Chromatog. Science*, 1970, **8**, 177; (*e*) W. Parr, C. Yang, E. Bayer, and E. Gil-Av, *ibid.*, p. 591; (*f*) W. Parr and P. Howard, *Chromatographia*, 1971, **4**, 162

[4] (*a*) B. Feibush, *Chem. Comm.*, 1971, 544; (*b*) R. Charles, U. Beitler, B. Feibush, and E. Gil-Av, *J. Chromatography*, 1975, **112**, 121; (*c*) U. Beitler and B. Feibush, *ibid.*,1976, **123**, 149.

[5] B. Feibush, E. Gil-Av, and T. Tamari, *J.C.S. Perkin II*, 1972, 1197.

[6] For preliminary communication see, B. Feibush, B. Altman, and E. Gil-Av, *J. Israel Chem. Soc.*, Proc. 41st Meeting Israel Chem. Soc., October 1971, p. 220.

[7] W. Parr and P. Howard, *J. Chromatography*, 1971, **71**, 193.
[8] H. Rubinstein, B. Feibush, and E. Gil-Av, *J.C.S. Perkin II*, 1973, 2094.
[9] T. Tamari, B. Feibush, and E. Gil-Av, unpublished data.
[10] Clough-Lutz-Jirgenson rule: see J. P. Greenstein and M. Winit, 'Chemistry of the Amino Acids,' Wiley and Sons, New York, 1961, vol. 1, p. 83.
[11] Y. Seto, T. Yamaka, K. Niwa, S. Miwa, F. Tanaka, S. Kuwata, and H. Watanabe, *Chem. Letters*, 1973, 151.
[12] *E.g.* S. Mizushima, T. Shimanouchi, M. Tsuboi, Y. Sugita, E. Kato, and E. Kondo, *J. Amer. Chem. Soc.*, 1951, **73**, 1330.
[13] Y. Koyama and T. Shimanouchi, *Biopolymers*, 1971, **10**, 1059; M. T. Cung, M. Marraud, and I. Néel, in 'The Jerusalem Symposia on Quantum Chemistry and Biochemistry,' eds. E. D. Bergmann and B. Pullman, Israel Academy of Science and Humanity, 1973, vol. 5, p. 69.
[14] T. Ichikawa and Y. Itaka, *Acta Cryst.*, 1969, **B25**, 1824; Y. Harada and Y. Itaka, *ibid.*, 1974, **B30**, 752, 1452.
[15] S. Winstein, Ph.D. Thesis, submitted to the Feinberg Graduate School, The Weizmann Institute of Science, Rehovot, May 1977.
[16] S. Winstein, B. Feibush, and E. Gil-Av, *J. Chromatography*, 1976, **126**, 97.
[17] S. Weinstein and L. Leiserowitz, to be published.
[18] P. Karrer, P. Portmann, and M. Suter, *Helv. Chem. Acta*, 1948, **31**, 1617.
[19] P. Karrer, P. Portmann, and M. Suter, *Helv. Chem. Acta*, 1949, **32**, 1156.
[20] J. Decombe, *Ann. Chim.* (*France*), 1932, **18**, 133.
[21] K. Balenović, D. Cerar, and Z. Fuks, *J. Chem. Soc.*, 1952, 3316.
[22] E. Fischer and H. Scheibler, *Annalen*, 1911, **383**, 348.
[23] F. Knoop and H. Oesterlin, *Z. physiol Chem.*, 1925, **148**, 309.
[24] E. Fischer and A. Groh, *Annalen*, 1911, **283**, 370.
[25] K. Balenović and D. Cerar, *J. Chem. Soc.*, 1955, 1631.
[26] H. D. Dakin, *J. Biol. Chem.*, 1933, **99**, 531.
[27] K. Balenović and D. Dvornik, *J. Chem. Soc.*, 1954, 2976.
[28] K. Wiberg and T. W. Hutton, *J. Amer. Chem. Soc.*, 1956, **78**, 1640.
[29] K. Balenović and D. Brovet-Keglević, *Arkiv Kemi*, 1951, **23**. 1 (*Chem. Abs.*, 1952, **46**, 10104).
[30] A. Chimiak, *Roczniki Chem.*, 1969, **43**, 299 (*Chem. Abs.*, 1969, **71**, 3631h).