754. *Interaction between Carbonyl Groups and Biologically Essential Substituents. Part VI.*[1] *A Study of Intramolecular Carbonyl–Amide Interaction in a Series of Optically Active 3-Oxoacylamides.*

By F. Bergel and Margaret A. Peutherer.

Rotational changes observed with lævulinoyl-tyrosine and -phenylalanine esters, and lævulinoylamphetamine, but not with lævulinoyl-leucine and -valine esters, or valerylamphetamine and lævulinoyl-*N*-methylamphetamine in the presence of small amounts of alkali suggested that a proportion of the molecules had cyclised to hydroxypyrrolidinone derivatives. The synthesis of a more rigid molecule, 2-acetylbenzoylamphetamine, was carried out by two routes, one theoretically leading to the open chain structure, and the other, *via* a Grignard reaction on an *N*-substituted phthalimide, to the cyclic tautomer. The products from both routes were identical, and represented benzopyrrolidinone derivatives. The possible formation of an anhydro-compound having an exocyclic methylene group is discussed. An intramolecular amide-carbonyl interaction has been established for the 2-acetylbenzoyl derivative and very probably occurs in the lævulinoyl series.

An interaction between amido- or imido-groups and the carbonyl of ketones and carboxy groupings can occur under certain *in vitro* or biosynthetic conditions. Such interactions may take place between two molecules or happen intramolecularly in the course of more complex events. An example of the first type was reported by Johnson and McCaldin [2] when they showed that amino-acid amides form a condensed ring system with trioxoindane hydrate (ninhydrin). This has been confirmed by us, and as the products from optically active alaninamide and phenylalaninamide were optically inactive in ethanol at the sodium D line, our findings support the tricyclic formula: [2] nevertheless our alanine amide derivative differed slightly in its ultraviolet absorption spectrum.

Examples of the second type have been described particularly in cases with sterically favourable situations, *e.g.*, proximity of groups. The transannular interactions between an amido and a carbonyl group of a ten-membered oxolactam, studied by Cohen and Witkop,[3] is an illustration of such spatial facilitation. Of a similar nature are the observations by Leonard *et al.*[4] with a series of nine-membered ring aminoacyloins, having a tertiary nitrogen instead of the amido-group. The investigations of Stoll *et al.*[5] have shown that the ergot alkaloids contain an unusual pyruvoyl peptide, the structure of which was confirmed by the synthesis of ergotamine by Hofmann, Frey, and Ott.[6] This structure contains, theoretically, two amido-carbonyl links, one between the amido-group of lysergic amide and the pyruvoyl keto group, and another between the carbonyl of the proline carboxyl and the imido-group of the pyruvoylphenylalanine moiety. Such cyclic systems have been investigated in detail by Shemyakin and his co-workers [7] and by Sheppard.[8] Schlientz *et al.*[9] found that a reversible isomerisation of the peptide part of the ergotamine molecule takes place in acid. A small but distinct shift in optical rotation was shown to accompany this formation of "aci"-ergotamine, but the ultraviolet absorption spectrum remained unchanged. Bicyclic intermediates formed by base-catalysed intramolecular carbonyl-imide interactions

[1] Part V, Bergel and Peutherer, preceding Paper.
[2] Johnson and McCaldin, *J.*, 1960, 3412.
[3] Cohen and Witkop, *J. Amer. Chem. Soc.*, 1955, **77**, 6595.
[4] Leonard, Fox, and Oki, *J. Amer. Chem. Soc.*, 1954, **76**, 5708.
[5] Cf. Stoll, *Fortschr. Chem. org. Naturstoffe*, 1952, **9**, 114.
[6] Hofmann, Frey, and Ott, *Experientia*, 1961, **17**, 206.
[7] Shemyakin, Antonov, Shkrob, Sheinker, and Senyavina, *Tetrahedron Letters*, 1962, 701; Antonov, Shkrob, Shchelokov, and Shemyakin, *ibid.*, 1963, 1353.
[8] Sheppard, *Experientia*, 1963, **19**, 125.
[9] Schlientz, Brunner, Thudium, and Hofmann, *Experientia*, 1961, **17**, 108.

have been postulated by Brenner and his co-workers[10] for the "amino-acid insertion" reaction of compounds such as *O*-glycylsalicoylamide and *O*-glycyl-*N*-benzoylserine amide. The latter compound is of particular interest because of its relation to intermediates suggested by Bernhard *et al.*[11] to explain the mechanism of the active site in a number of proteolytic and esteratic enzymes. Model experiments involving the base-catalysed hydrolysis of compounds such as β-benzyl-*N*-benzyloxycarbonyl-L-aspartyl-L-serine amide indicated that intermediates containing an >N–COR linkage might exist. The alkaline hydrolysis of this model compound resulted in polarimetric changes, *viz.*, the specific rotation at first became rapidly more negative and then fell gradually almost to zero. No such optical changes were observed by Fosker *et al.*[12] during the base-catalysed hydrolysis of γ-benzyl-*N*-benzyloxycarbonyl-L-glutamyl-L-serine amide. In this case, however, the cyclic intermediate would involve a six- rather than a five-membered ring system. Hassall *et al.*[13] have suggested the production of a bicyclic system during the hydrolysis of the amide link of *N*-glycyl-β-acetylalanine, involving amino, carbonyl, and amido groups.

At this point mention should be made of a hypothesis put forward on several occasions by Wrinch[14] concerning ring formations, called by her "cyclols," between two >CO·NH– groupings in peptides. The only synthetic proof of such structures was offered recently by Jones, Kenner, and Sheppard[15] during the preparation of the bulky cyclic tripeptide containing three dimethylglycine residues. Apart from this last example, all the cyclic systems described above involve the nitrogen of only one amide grouping usually linked with a carboxy or ketone carbonyl. We propose to call such structures "hemicyclols" or "cyclol-like," to distinguish them from those designated by Wrinch[14] as cyclols, consisting of *two* CO·NH partners.

The possibility of related intramolecular interaction between a secondary amide and the ketone-carbonyl group of a series of optically active 3-oxoacylamides was investigated by us, as an extension of the work[1,16,17] on the mutarotation of optically active amino-derivatives in ketonic solvents under mild conditions.

These oxoacylamides were prepared from lævulinic acid and optically active amino-acids or amines, and their optical behaviour in ethanolic solution on the addition of small amounts of aqueous alkali and acid was studied (Table, nos. 1—5). In addition, acyl derivatives not containing the ketone carbonyl group (Table, nos. 6, 7, and 8), one

Maximal $[\alpha]_D$ at 19—24° (*c* 1·3—4·0).

	Compound	EtOH	EtOH + 0·1N-NaOH	EtOH + 0·1N-HCl
1	Læv.-L-val-Et ester	−18·8°	−19·0°	−18·2°
2	Læv.-D-leu-Et ester	+17·3	+17·3	+17·2
3	Læv.-amph.	−10·0	+28·0	−7·5
4	Læv.-L-tyr-Et ester	+14·5	−5·1	+13·0
5	Læv.-*p*-NO_2-L-phe-Et ester	+9·95	−15·0	+7·0
6	*n*-Valeryl-amph.	−4·5	−5·0	—
7	Isovaleryl-amph.	+2·0	+2·0	—
8	*N*-Acetyl-L-tyr-Et ester	+22·5	+23·8	+21·6
9	Læv.-*N*-methyl-amph.	+32·3	+33·4	—
10	Amph.	+32·0	+34·2	+32·6

Læv. = lævulinoyl. Amph. = (+)-amphetamine.

lævulinoyl tertiary amide (no. 9), and amphetamine (no. 10) served as controls and were investigated under the same conditions. The infrared absorption spectra of compounds

[10] Brenner, Zimmermann, Wehrmüller, Quitt, Hartman, Schneider, and Beglinger, *Helv. Chim. Acta*, 1957, **40**, 1497; Brenner, in "Ciba Foundation Symposium on Amino Acids and Peptides with Antimetabolic Activity," Churchill, London, 1958, p. 261.

[11] Bernhard, Berger, Carter, Katchalski, Sela, and Shalitin, *J. Amer. Chem. Soc.*, 1962, **84**, 2421.

[12] Fosker, Hanson, and Law, *Chem. and Ind.*, 1963, 569.

[13] Hassall, John, Martin, and Schofield, *J.*, 1963, 3100.

[14] Wrinch, *Nature*, 1936, **137**, 411; **138**, 241; Wrinch, "Chemical Aspects of the Structure of Small Peptides," Munksgaard, Copenhagen, 1960.

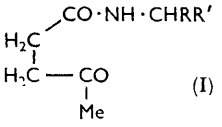
[15] Jones, Kenner, and Sheppard, *Experientia*, 1963, **19**, 126.

[16] Bergel, Lewis, Orr, and Butler, *J.*, 1959, 1431.

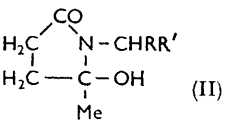
[17] Bergel and Butler, *J.*, 1961, 4047.

1—8, before addition of alkali or acid, showed both amide I and amide II bands, indicating that these oxoacyl- and acyl-amides existed substantially in the open-chain form (I). This is reasonable, as unlike most of the examples quoted above, they have no steric rigidity to facilitate intramolecular amide-carbonyl interaction.

On addition of 0·1N-hydrochloric acid, no significant alteration to the specific rotations of any of the compounds occurred (Table). On the other hand, on addition of alkali, nos. 3, 4, and 5 underwent a change of optical rotation. The amount of alkali necessary to produce the shift was small, being less than 0·1 equiv. of 0·1N-aqueous sodium hydroxide. Further addition of alkali beyond that required to cause the maximum rotational change made the specific rotation fall gradually towards zero. No changes in optical rotation were observed with those compounds not containing the following: a lævulinoyl residue with its oxo-group, a secondary amide hydrogen, or an aromatic ring in the side chain (I, R′ = CH_2·Ph, CH_2·C_6H_4·OH, CH_2·C_6H_4·NO_2). These facts led us to suggest that the alteration of the specific rotation of compounds nos. 3, 4, and 5 having both a ketone carbonyl and a secondary amide group might be associated with an intramolecular base-catalysed ring closure (I) ⟶ (II) between these two groups.



(I)



(II)

Lukeš and Linhartová [18] prepared lævulinoylmethylamine and after measuring its infrared absorption spectrum in solution, concluded that it exists in a cyclic form. They also suggested that the structure of lævulinoylaniline is that of an open secondary amide, since it showed both amide (I) and (II) absorption bands.[19] The conclusion reached by Cromwell and Cook [20] concerning the structures of the methylamido-, anilido-, benzylamido-, and cyclohexylamido-derivatives of β-benzoylpropionic acid was that they involve a tautomeric equilibrium between the corresponding β-benzoylpropionamides and the cyclic 5-hydroxy-5-phenylpyrrolidin-2-ones.

The absence of optical change on addition of alkali to compounds 1 and 2 which do not contain an aromatic ring can be explained in terms of a possible mechanism for the intramolecular interaction. The presence of the electron-withdrawing aromatic ring which facilitates loss of a proton at the amide group and hence formation of a negatively-charged nitrogen to react with the positively-charged carbon of the ketone carbonyl, may be necessary for the ring closure. This mechanism is similar to that suggested by Bernhard *et al.*[11] for imide hydrolysis. Of the two compounds 4 and 5, the larger shift was observed for lævulinoyl-*p*-nitro-L-phenylalanine ethyl ester (no. 5). This could indicate an additional influence of the electron-withdrawing nitro-group compared with the aromatic ring of tyrosine (no. 4) carrying the electron-donating hydroxyl group.

Further studies with lævulinoylamphetamine and lævulinoyl-L-tyrosine ethyl ester demonstrated that addition of 0·01N-sodium hydroxide to ethanolic solutions of these compounds produced similar rotational changes. The shifts could also be observed in aqueous solution by addition of sodium hydroxide; in ethanol by addition of ethanolic sodium hydroxide; in 50% v/v water–pyridine, and in ethanol by addition of pyridine or triethylamine. In the last three cases the changes produced were smaller than those given by aqueous or alcoholic sodium hydroxide. No alteration of specific rotation was observed when pyridine was added to a solution of lævulinoylamphetamine in carbon tetrachloride or when aqueous sodium hydroxide was added to a solution of the compound (I; R = Me, R′ = CH_2·Ph) in water containing sodium hydrogen sulphite or sulphite. Burton,

[18] Lukeš and Linhartová, *Coll. Czech. Chem. Comm.*, 1960, **25**, 502.
[19] Bellamy, " The Infra-red Spectra of Complex Molecules," Methuen and Co., London, 1958, p. 205.
[20] Cromwell and Cook, *J. Amer. Chem. Soc.*, 1958, **80**, 4573.

McWeeny, and Biltcliffe [21] found that the last two compounds inhibit the non-enzymatic browning following an amino-carbonyl interaction between sugars and amino-compounds. No evidence for the splitting of lævulinoyl-L-tyrosine ethyl ester or lævulinoylamphetamine on addition of amounts of alkali required to produce the maximal optical changes could be detected by chromatography: no ninhydrin spots corresponding to either L-tyrosine ethyl ester or (+)-amphetamine were found when chromatograms of the compounds in solutions containing aqueous alkali were developed with butanol–ethanol–water (10 : 5 : 5). If such spots had been noted, they would have indicated a decomposition by hydrolysis of the lævulinoyl derivatives to the amino-moiety together with, perhaps, the lactone of the enol form of lævulinic acid. However, once the maximal rotation change had been produced it was found to be stable to both time and change of temperature, again supporting the chromatographic evidence for the absence of breakdown products (see Experimental section). Addition of 0·01N-ethanolic hydrochloric acid to an ethanolic solution of lævulinoylamphetamine brought to the maximal rotation by addition of ethanolic sodium hydroxide, caused the change to be reversed, although the original value of the specific rotation of lævulinoylamphetamine in ethanol was not reached, even when amounts of acid far beyond those required to neutralise the alkali present had been added. This may indicate racemisation.

Other evidence for intramolecular interaction was difficult to obtain. No change in the ultraviolet absorption spectrum of lævulinoylamphetamine in ethanol was observed on addition of alkali. This was, however, not unexpected from Schlientz's work [9] with ergotamine and its "aci"-isomer. Reduction of lævulinoylamphetamine in ethanol, containing aqueous sodium hydroxide, with a Raney-nickel catalyst, conditions which might stabilise the hydroxypyrrolidinone structure, produced a racemic compound, shown to be the alcohol (III) by the loss of the ketone carbonyl absorption at 1700 cm.$^{-1}$ and the retention of the amide (I) and (II) absorptions.

$$CH_3{\cdot}CHOH{\cdot}CH_2{\cdot}CH_2{\cdot}CONH{\cdot}CH(CH_3){\cdot}CH_2{\cdot}Ph$$
(III)

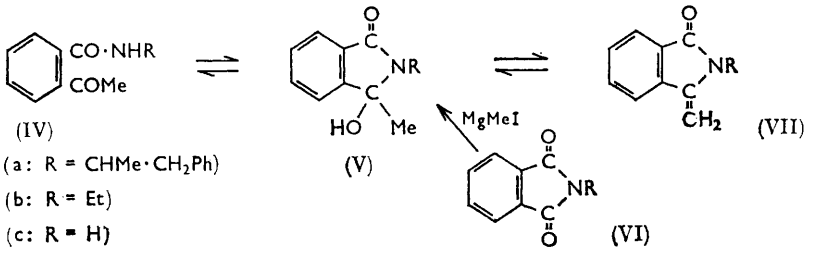
The solid obtained on removal of alcohol under reduced pressure from a solution of lævulinoylamphetamine in ethanol containing aqueous sodium hydroxide, while still having the maximal rotation value on being redissolved in ethanol, had the same infrared absorption spectrum as the starting mixture. These results suggest that the optical rotational changes noted on addition of base to alcoholic or aqueous solutions of compounds **3**, **4**, and **5** are due to the formation of small amounts of the hemicyclol (II). This is not surprising in view of the mobility of the aliphatic lævulinoyl residue.

In order to clarify this so far ambiguous situation, the 2-acetylbenzoic acid derivative of (+)-amphetamine was prepared and studied. This compound (IVa) is closely related to the lævulinic acid derivative no. **3**, except that it contains the rigid benzene ring structure which should facilitate intramolecular interaction and preserve the cyclic structure (Va). Infrared absorption had demonstrated that 2-acetylbenzoic acid itself exists as a five-membered ring lactol [22] rather than as the open-chain compound.

Two approaches were made to the problem of the structure of 2-acetylbenzoylamphetamine: (*a*) synthesis by routes which theoretically produce the open-chain compound (IVa); and (*b*) synthesis by a route leading to the cyclic structure (Va). In the first approach, 2-acetylbenzoylaniline was synthesised by the condensation of 2-acetylbenzoyl chloride on cyanomethyl 2-acetylbenzoate [23] with aniline, and by the coupling of 2-acetylbenzoic acid with aniline in the presence of dicyclohexylcarbodi-imide. In each case, the infrared spectrum of the product did not show absorption bands characteristic of the secondary amide group. This negative result together with the presence of an absorption at 1700 cm.$^{-1}$

[21] Burton, McWeeny, and Biltcliffe, *Nature*, 1962, **196**, 40.
[22] Bellamy, "The Infra-red Spectra of Complex Molecules," Methuen and Co., London, 1958, p. 169.
[23] Schwyzer, Iselin, and Feurer, *Helv. Chim. Acta*, 1955, **38**, 69.

which is normally ascribed to a CO group would indicate a γ-lactam structure.[19] Some difficulty was encountered in the synthesis of the (+)-amphetamine derivative: the mixed anhydride procedure gave a product contaminated with urethane; the dicyclohexylcarbodiimide coupling produced 2-acetylbenzoyl-*NN'*-dicyclohexylurea as the main product, and phenylthio 2-acetylbenzoate did not undergo condensation with (+)-amphetamine. Of the methods used, the coupling of cyanomethyl 2-acetylbenzoate with (+)-amphetamine gave the best yield of product. Smaller yields were obtained from the acid chloride, and from the reaction of 2-acetylbenzoic acid and (+)-amphetamine in pyridine containing benzenesulphonyl chloride. The products from all successful methods showed the absence



of secondary amide absorption and the presence of γ-lactam carbonyl absorption in the 1700 cm.$^{-1}$ region, allowing the conclusion that in the solid state structure (Va) was the stable form. The specific rotation of 2-acetylbenzoylamphetamine was +206° (amphetamine +32·0°), a value which unlike that of the lævulinoyl-derivative changed only slightly on addition of aqueous sodium hydroxide.

The alternative method of synthesising 2-acetylbenzoylamphetamine or its cyclic isomer (Va) was by the treatment of phthaloylamphetamine (VIa) with methylmagnesium iodide. In a model experiment, *N*-ethylphthalimide (VIb) was treated with the Grignard reagent following the method of Sachs and Ludwig.[24] The infrared spectrum of the compound obtained was devoid of secondary amide absorption, but showed the γ-lactam carbonyl absorption at 1690 cm.$^{-1}$ (Vb). Similarly, treatment of phthaloylamphetamine (VIa) with methylmagnesium iodide gave a product with the same infrared characteristics and hence structure (Va). The same product was obtained from the acid chloride, cyanomethyl ester, and benzenesulphonyl chloride routes.

The possibility that the anhydro-products (VII) might have been formed was brought to light when the nuclear magnetic resonance spectrum of (Va) was recorded. This indicated the presence of an exocyclic methylene group and the absence of tertiary hydroxyl. The simpler compounds (VIIc) and (Vb) were then submitted to comparative tests by ultraviolet and nuclear magnetic resonance measurements. The ultraviolet spectrum of methylenephthalimidine (VIIc) showed absorption in ethanol at 220, 255, and 305 mμ. The compounds (Va and b) while possessing absorption at 220 and 251, and 220 and 253 mμ, respectively, did not absorb in the 300 mμ region. Overnight treatment of (Va and b) with acetic anhydride,[15] which should produce the anhydro-compounds (VIIa and b) gave products with absorption at 305 mμ. This indicates formation of the conjugated system present in the structure (VII). The nuclear magnetic resonance spectrum of (VIIc) showed a pair of doublets due to the exocyclic methylene group at about τ 4·7. In the spectrum of compound (Vb) the presence of an unsplit methyl resonance, together with a quartet at τ 6·1 attributed to >NEt, supports the methylcarbinol structure. The fact that two doublets were shown in the vinyl region indicated that under the conditions of measurement the compound (Vb) and hence (Va) existed as an equilibrium mixture of hydrated (Va and b) and anhydro- (VIIa and b) compounds. Regardless of the existence of such equilibria between structures (V) and (VII) and thus for the compounds derived from the open structure (IV), an intramolecular amide-carbonyl interaction has been established with

[24] Sachs and Ludwig, *Ber.*, 1904, **37**, 385.

certainty and is also likely for the lævulinoyl series. It is believed that such interactions involving carbonyl and amide groups under mild conditions may be important for other compounds either of synthetic or, more interesting still, of natural origin.

Experimental

Optical rotations were measured as described in Part I.[16] In those experiments involving addition of aqueous alkali or acid to ethanolic solutions of lævulinoyl and related compounds, the following general procedure was applied: a known weight of compound was dissolved in ethanol (2 ml.), and to it was added portions (0·01 ml.) of alkali or acid until a total volume of 0·1 ml. alkali or acid had been added. The maximal rotational values observed and the amounts of alkali to produce these values are reported below (see also Table).

Infrared absorption measurements were made with a Perkin-Elmer Infracord spectrophotometer. The samples were measured as mulls in Nujol or as capillary films. Ultraviolet absorption measurements were made with a Perkin-Elmer model 137 UV spectrophotometer.

Condensation of Ninhydrin and D-*Alanine Amide.*—Following the method of Johnson and McCaldin,[2] D-alanine amide (66 mg.) and ninhydrin hydrate (120 mg.) were condensed to give yellow prisms (52 mg., 33%) of the condensation product, m. p. 320—321° (lit.,[2] m. p. 324—325°), $[\alpha]_D^{22}$ 0° (*c* 2 in EtOH), $\lambda_{max.}$ (in MeOH) 235, 267, 297, 308, and 353 mμ (in MeOH + 0·01N-NaOH) 238, 285, 298, 308, and 357 mμ (Found: C, 67·7; H, 3·8; N, 13·1. Calc. for $C_{12}H_8N_2O_2$: C, 67·9; H, 3·8; N, 13·2%).

L-*Phenylalanine Amide.*—L-Phenylalanine ethyl ester (4·0 g.) in methanol (40 ml.) saturated with ammonia was set aside at room temperature for 50 hr. On removal of excess of reagent, a solid formed which was recrystallised from chloroform–ether to give prisms of L-phenylalanine amide (2·6 g., 77%), m. p. 94—95°, $[\alpha]_D^{20}$ −5° (*c* 2 in EtOH) (Found: C, 65·7; H, 7·8; N, 17·2. $C_9H_{12}N_2O$ requires C, 65·8; H, 7·4; N, 17·1%). Blau and Waley [25] give m. p. 93—94° and $[\alpha]_D$ +20·7° (*c* 1·75 in H_2O) but no analytical figures.

Condensation of L-*Phenylalanine Amide and Ninhydrin.*—L-Phenylalaninamide (124 mg.) and ninhydrin (120 mg.) were condensed as above to give a yellow solid, which was recrystallised from methanol to give the condensation *product* (70 mg., 32%), m. p. 285°, $[\alpha]_D^{22}$ 0° (*c* 2 in EtOH); $\lambda_{max.}$ (in MeOH) 237, 265, 298, 309, and 357 mμ (in MeOH + 0·01N-NaOH) 239, 285, 299, 309, and 361 mμ (Found: C, 74·9; H, 4·4; N, 9·7. $C_{18}H_{12}N_2O_2$ requires C, 75·0; H, 4·2; N, 9·7%).

*Lævulinoyl-*L-*valine Ethyl Ester.*—Isobutyl chloroformate (3·82 ml.) was added to a cooled (5°), stirred solution of lævulinic acid (5·3 g.) in tetrahydrofuran (10 ml.) containing triethylamine (4·05 ml.), and after 20 min. L-valine ethyl ester hydrochloride (5·3 g.) in chloroform (10 ml.) containing triethylamine (4·05 ml.) was added. After 16 hr. at 20°, the mixture was evaporated to dryness, extracted with ethyl acetate, and the extracts were washed with 2N-hydrochloric acid, saturated aqueous sodium hydrogen carbonate and water, and dried (Na_2SO_4). The mixture was filtered, the filtrate evaporated to dryness, and the residue was distilled *in vacuo* to give *lævulinoyl-*L-*valine ethyl ester* (5·5 g., 79%), b. p. 122—123°/0·05 mm., n_D^{19} 1·4570, $[\alpha]_D^{19}$ −18·8° (*c* 3·5 in EtOH), $[\alpha]_D^{19}$ −19·0° (*c* 3·3 in 2 ml. of EtOH + 0·08 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0038N); $\nu_{max.}$ 1710, 1650, and 1520 cm.$^{-1}$ (Found: C, 59·2; H, 8·9; N, 5·9. $C_{12}H_{21}NO_4$ requires C, 59·2; H, 8·7; N, 5·8%).

*Lævulinoyl-*D-*leucine Ethyl Ester.*—Following the above procedure with lævulinic acid (4·15 g.) and D-leucine ethyl ester hydrochloride (7·0 g.), *lævulinoyl-*D-*leucine ethyl ester* (7 g., 77%) was obtained as an oil, b. p. 179—180°/1 mm., n_D^{24} 1·4580, $[\alpha]_D^{24}$ +17·3° (*c* 3·3 in EtOH), $[\alpha]_D^{24}$ +17·3° (*c* 3·2 in 2 ml. of EtOH +0·06 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0028N); $\nu_{max.}$ 1720, 1650, and 1540 cm.$^{-1}$ (Found: C, 60·5; H, 8·8; N, 5·4. $C_{13}H_{23}NO_4$ requires C, 60·7; H, 9·0; N, 5·4%).

Lævulinoyl-(+)-amphetamine.—Lævulinic acid (2·32 g.) and (+)-amphetamine sulphate (7·37 g.) were condensed as described above. The solid obtained was crystallised from benzene–light petroleum (b. p. 60—80°) to give prisms of *lævulinoyl-(+)-amphetamine* (3·9 g., 83%), m. p. 78—80°, $[\alpha]_D^{19}$ −10·0° (*c* 2 in EtOH), $[\alpha]_D^{19}$ +28·0° (*c* 2 in 2 ml. of EtOH + 0·04 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0019N), $[\alpha]_D^{19}$ +19·0° (*c* 1 in 50% v/v pyridine–water), $[\alpha]_D^{20}$ +28·0° (*c* 1·6 in EtOH + pyridine), $[\alpha]_D^{20}$ +25·0° (*c* 1·6 in EtOH + triethylamine), $[\alpha]_D$ +25° (*c* 2 in 2 ml. of EtOH + 0·04 ml. of 0·1N-NaOH after heating for 1 hr. at 50°); $\nu_{max.}$ 1700,

[25] Blau and Waley, *Biochem. J.*, 1954, **57**, 538.

1640, and 1540 cm.$^{-1}$ (Found: C, 72·0; H, 8·2; N, 6·1. $C_{14}H_{19}NO_2$ requires C, 72·1; H, 8·2; N, 6·0%).

Lævulinoyl-L-tyrosine Ethyl Ester.—Lævulinic acid (1·66 g.) and L-tyrosine ethyl ester (3·0 g.) were condensed in a similar fashion, without addition of triethylamine to the chloroform solution of L-tyrosine ethyl ester. The product was crystallised from benzene–light petroleum (b. p. 60—80°) to give prisms of *lævulinoyl-L-tyrosine ethyl ester* (3·8 g., 86%), m. p. 92—94°, $[\alpha]_D^{19}$ +14·5° (*c* 2 in EtOH), $[\alpha]_D^{19}$ −5·1° (*c* 2 in 2 ml. of EtOH + 0·05 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0024N), $[\alpha]_D^{19}$ −5·1° (same solution after 17 hr. at room temperature); $\nu_{max.}$ 1715, 1640, and 1520 cm.$^{-1}$ (Found: C, 62·1; H, 6·9; N, 4·6. $C_{16}H_{21}NO_5$ requires C, 62·5; H, 6·9; N, 4·6%).

Lævulinoyl-p-nitro-L-phenylalanine Ethyl Ester.—*p*-Nitro-L-phenylalanine ethyl ester hydrochloride was prepared from *p*-nitro-L-phenylalanine as described by Bergel and Stock,[26] and was coupled (0·9 g.) with lævulinic acid (0·38 g.) to give a solid which on crystallisation from benzene–light petroleum (b. p. 60—80°) gave prisms of *lævulinoyl-p-nitro-L-phenylalanine ethyl ester* (0·8 g., 69%), m. p. 102—104°, $[\alpha]_D^{20}$ +9·95° (*c* 1·9 in EtOH), $[\alpha]_D^{20}$ −15·0° (*c* 2 in 2 ml. of EtOH + 0·06 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0028N); $\nu_{max.}$ 1720, 1640, and 1540 cm.$^{-1}$ (Found: C, 56·2; H, 6·1; N, 8·2. $C_{16}H_{20}N_2O_6,0{\cdot}5H_2O$ requires C, 55·9; H, 6·1; N, 8·2%).

n-Valeryl-(+)-amphetamine.—(+)-Amphetamine (2·3 g.) in benzene (10 ml.) was added with stirring to *n*-valeryl chloride (0·90 ml.) in benzene. After 1 hr. the mixture was washed with 2N-hydrochloric acid, saturated aqueous sodium hydrogen carbonate and water, and dried (Na_2SO_4). On recovery, a solid was obtained which after crystallisation from benzene gave prisms of *n-valeryl-(+)-amphetamine* (1·1 g., 70%), m. p. 35°, $[\alpha]_D^{20}$ −4·5° (*c* 2 in EtOH), $[\alpha]_D^{20}$ −5·0° (*c* 1·9 in 2 ml. of EtOH + 0·08 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0038N); $\nu_{max.}$ 1630 and 1540 cm.$^{-1}$ (Found: C, 76·7; H, 9·5; N, 6·3. $C_{14}H_{21}NO$ requires C, 76·7; H, 9·7; N, 6·4%).

Isovaleryl-(+)-amphetamine.—This compound was prepared as above from isovaleryl chloride (3·6 ml.) and (+)-amphetamine (8·0 g.). The *product* (5·0 g., 76%) was obtained from ethanol–water as prisms, m. p. 110°, $[\alpha]_D^{20}$ +2·0° (*c* 2 in EtOH), $[\alpha]_D^{20}$ +2·0° (*c* 2 in 2 ml. of EtOH + 0·04 ml. of 0·1N-NaOH, *i.e.*, mixture 0·0019N); $\nu_{max.}$ 1630 and 1540 cm.$^{-1}$ (Found: C, 76·2; H, 9·4; N, 6·5%).

Lævulinoyl-N-methyl-(+)-amphetamine.—α-Angelicalactone (4·8 g.) prepared by the method of Helberger *et al.*[27] and *N*-methyl-(+)-amphetamine (7·3 g.) were heated on a steam-bath for 2 hr. The mixture was then extracted with ethyl acetate, washed with 2N-hydrochloric acid, 2N-sodium hydroxide, water, and dried (Na_2SO_4). Recovery furnished a viscous oil which could not be distilled *in vacuo* and was passed down an alumina column in benzene. After removal of solvent, the *oil* was dried in a desiccator for 2 days over phosphorus pentoxide and sulphuric acid; it had $\nu_{max.}$ 1720 and 1650 cm.$^{-1}$, $[\alpha]_D^{20}$ + 32·3° (*c* 1·3 in EtOH) (Found: C, 71·4; H, 8·7; N, 5·7. $C_{15}H_{21}NO_2$ requires C, 72·8; H, 8·6; N, 5·7%).

Cyanomethyl 2-Acetylbenzoate.—2-Acetylbenzoic acid (6·5 g.), prepared by the method of Yale,[28] triethylamine (17·9 ml.), and chloroacetonitrile (11·1 ml.) were stirred at 0° until solution was complete. The mixture was then warmed at 40° for 2 hr., extracted with ethyl acetate, washed with 2N-hydrochloric acid, citric acid solution, saturated aqueous sodium hydrogen carbonate, saturated aqueous sodium chloride, and dried (Na_2SO_4). The mixture was filtered, concentrated to small volume (2 ml.) under reduced pressure, and passed through alumina with benzene as eluant. On evaporation of the eluate, a brown oil was obtained which slowly solidified at room temperature. The solid was recrystallised from benzene to give prisms of *cyanomethyl 2-acetylbenzoate* (4·5 g., 55%), m. p. 95°; $\nu_{max.}$ 2250 (weak) and 1760 cm.$^{-1}$ (Found: C, 65·1; H, 4·2; N, 6·9. $C_{11}H_9NO_3$ requires C, 65·0; H, 4·5; N, 6·9%).

2-Acetylbenzoylaniline.—(*a*) 2-Acetylbenzoyl chloride (2·0 g.), prepared by the method of Halford and Weissmann,[29] and aniline (2·04 g.) were stirred at room temperature for 1 hr. The mixture was washed with 2N-hydrochloric acid, saturated aqueous sodium hydrogen carbonate, water, and dried (Na_2SO_4). Recovery and crystallisation, from ethanol–water gave 2-acetylbenzoylaniline (2·0 g., 76%), m. p. 150°; $\nu_{max.}$ 3300, 1700, and 1150 cm.$^{-1}$.[30] (*b*) Cyanomethyl

[26] Bergel and Stock, *J.*, 1954, 2409.
[27] Helberger, Ulubay, and Civelekoglu, *Annalen*, 1949, **561**, 215.
[28] Yale, *J. Amer. Chem. Soc.*, 1947, **69**, 1547.
[29] Halford and Weissmann, *J. Org. Chem.*, 1952, **17**, 1646.
[30] Karslake and Huston, *J. Amer. Chem. Soc.*, 1909, **31**, 479.

2-acetylbenzoate (3·2 g.), acetic acid (4 drops), and aniline (5·15 g.) were warmed at 60° for 1 hr. The mixture was poured into water (200 ml.) and the organic layer was dried (Na_2SO_4) and evaporated to dryness. The product, recrystallised as above, had m. p. 149°; $\nu_{max.}$ 3250, 1680, and 1150 cm.$^{-1}$. (*c*) 2-Acetylbenzoic acid (5·0 g.) in tetrahydrofuran (5 ml.) and aniline (2·85 g.) in tetrahydrofuran (5 ml.) were mixed, and dicyclohexylcarbodi-imide (6·2 g.) was added. The mixture was stirred at room temperature for 3 hr. and left overnight. Acetic acid (3 ml.) was added, and the mixture was filtered and evaporated to dryness. The ethyl acetate extract of the residue was washed with 2N-hydrochloric acid, saturated aqueous sodium hydrogen carbonate, and water, dried (Na_2SO_4) and evaporated to dryness to give 2-acetylbenzoylaniline, m. p. 150°; $\nu_{max.}$ 3300, 1680, and 1150 cm.$^{-1}$.

D(+)-2-α-*Methylphenethyl*-3-*hydroxy*-3-*methylisoindolinone* (Va).—(a) The condensation of 2-acetylbenzoic acid (5·0 g.) and (+)-amphetamine sulphate (11·2 g.) with dicyclohexylcarbodi-imide as described above gave as the main product (3·0 g.), 2-*acetylbenzoyl*-NN′-*dicyclohexylurea*, m. p. 95°; $\nu_{max.}$ 3250, 1700, 1680, 1640, and 1530 cm.$^{-1}$ (Found: C, 71·4; H, 8·1; N, 7·9. $C_{22}H_{30}N_2O_3$ requires C, 71·3; H, 8·1; N, 7·6%). (*b*) *Phenylthio*-2-*acetylbenzoate* was prepared from 2-acetylbenzoic acid (5·0 g.) and thiophenol (3·12 ml.) following the usual mixed anhydride method as described for the lævulinic acid derivatives. The product was recrystallised from ethanol to give prisms of *phenylthio*-2-*acetylbenzoate* (6·5 g., 83%), m. p. 105° (Found: C, 70·3; H, 5·1; N, 12·3. $C_{15}H_{12}O_2S$ requires C, 70·3; H, 4·7; N, 12·5%). The ester (8·0 g.) and (+)-amphetamine (4·7 g.) were boiled in ethanol overnight. No condensation resulted. (*c*) Cyanomethyl 2-acetylbenzoate (0·68 g.), acetic acid (4 drops), and (+)-amphetamine (0·9 g.) were warmed at 60° in tetrahydrofuran (10 ml.) overnight. The reaction was worked up as in the corresponding preparation of 2-acetylbenzoylaniline, to give an oil which solidified when kept over phosphorus pentoxide. Crystallisation from benzene–light petroleum (b. p. 60—80°) gave prisms of D(+)-2-α-*methylphenethyl*-3-*hydroxy*-3-*methylisoindolinone* (Va) (200 mg., 21%), m. p. 125°, $[\alpha]_D^{22}$ +205·8° (*c* 0·3 in EtOH); $\nu_{max.}$ 3300, 1680, and 1140 cm.$^{-1}$ (Found: C, 76·9; H, 7·0; N, 5·2. $C_{18}H_{19}NO_2$ requires C, 76·8; H, 6·8; N, 5·0%). (*d*) The product from 2-acetylbenzoyl chloride (5·6 g.) and (+)-amphetamine, prepared as described in the corresponding preparation of 2-acetylbenzoylaniline, was a yellow oil (7·0 g., 80%) which was purified by treatment in ethanol with animal charcoal, and by elution with benzene from an alumina column. A small yield of solid (10 mg.), recrystallised from ethyl acetate–light petroleum (b. p. 60—80°), was obtained, m. p. 126°, $[\alpha]_D^{22}$ +200° (*c* 0·8 in EtOH); $\nu_{max.}$ 3200, 1680, and 1140 cm.$^{-1}$. (*e*) 2-Acetylbenzoic acid (3·4 g.) was dissolved in pyridine (10 ml.) and benzenesulphonyl chloride (1·3 g.) added to the cooled (5°) solution. After 15 min., (+)-amphetamine (2·8 g.) was added and the mixture left overnight at room temperature. It was then poured into ice–water (20 ml.), extracted with ethyl acetate, and worked up as in the corresponding preparation of 2-acetylbenzoylaniline. A small yield of solid with physical characteristics as described in (*c*) and (*d*) above was obtained.

(*f*) 2-*Carboxybenzoyl*-(+)-*amphetamine* was prepared by the addition of phthalic anhydride (5·5 g.) to a solution of (+)-amphetamine (5·0 g.) in ether (200 ml.). The precipitate was crystallised from ethyl acetate–light petroleum (b. p. 60—80°) to give prisms of 2-*carboxybenzoyl*-(+)-*amphetamine* (8·0 g., 76%), m. p. 125°, $[\alpha]_D^{20}$ + 20·7° (*c* 1·5 in EtOH) (Found: C, 71·6; H, 5·9; N, 5·0. $C_{17}H_{17}NO_3$ requires C, 72·1; H, 6·1; N, 4·9%). This product (6·5 g.) was refluxed in 4% ethanolic hydrochloric acid (30 ml.) for 2 hr. The solution was evaporated to dryness and shaken with water (20 ml.) and chloroform (20 ml.). The chloroform layer was washed with saturated aqueous sodium hydrogen carbonate and water, and dried (Na_2SO_4). Recovery and crystallisation from ethanol gave prisms of *phthaloyl*-(+)-*amphetamine* (3·4 g., 61%), m. p. 80°, $[\alpha]_D^{20}$ +157·1° (*c* 2·1 in EtOH) (Found: C, 76·6; H, 5·7; N, 5·5. $C_{17}H_{15}NO_2$ requires C, 76·9; H, 5·7; N, 5·3%). The Grignard procedure [24] was followed with phthaloyl-(+)-amphetamine (2·5 g.) and methylmagnesium iodide. The product obtained was an oil (1·5 g., 58%) from which a small amount of solid was obtained. This was crystallised from ethyl acetate–light petroleum (b. p. 60—80°) to give D(+)-2-α-methylphenethyl-3-hydroxy-3-methyl-isoindolinone (Va).

2-*Ethyl*-3-*hydroxy*-3-*methyl*isoindolinone (Vb).—This compound was prepared by the method of Sachs and Ludwig [24] from *N*-ethylphthalimide and methylmagnesium iodide. The product was obtained as a solid, m. p. 130° (lit.,[24] m. p. 93—94°) (Found: C, 68·8; H, 7·0; N, 7·3. Calc. for $C_{11}H_{13}NO_2$: C, 69·1; H, 6·9; N, 7·3%).

Methylenephthalimidine was prepared by the method of Dent.[31]

[31] Dent, *J.*, 1938, 1.

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