279. *Interaction between Carbonyl Groups and Biologically Essential Substituents. Part I. The Effect of Ketones on Optically Active Amino-derivatives.*

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Mutarotation has been shown to occur when a number of optically active α-amino-esters, and one amine, are dissolved in ketones. The magnitude and the rate of this time-consuming change depend on the ketone used. With the L- and D-enantiomorphs so far tested, this change has been respectively towards higher negative and positive values of α. *cyclo*Pentanone and L-tyrosine ethyl ester yield a crystalline azomethine; this and infrared absorption measurements on mixtures of *cyclo*hexanone and (+)-amphetamine have given some insight into the mechanism of this interaction under mild conditions.

The interaction between carbonyl groups and groups such as NH_2 and SH in biologically important compounds has been widely studied from chemical and biochemical angles, *e.g.*, interaction of aldehydes and ketones and of amino-groups in the biosynthesis of alkaloids,[1] and the interconversion of α-amino-acids and keto-acids [2] in transamination and decarboxylation in presence of pyridoxal and pyridoxamine derivatives. The latter series of reactions has been explored in model experiments by, *e.g.*, Snell,[3] Metzler, Ikawa, and Snell,[4] and Christensen.[5] The degradation of α-amino-acids by carbonyl compounds *in vitro* has been reviewed by Schönberg and Moubacher,[6] and investigated more recently by Spenser *et al.*[7] and others; the reaction between sugars and amino-acids has been examined by Lewin [8] and between hydroxy-ketones and amino-acids by Hurd and Buess.[9] Even more interesting biologically may be intramolecular transannular interaction in peptides which is under study by means of models [10] and natural products.[11]

The effects of aldehydes and ketones on the optical rotatory powers of amino-acids have formed the subjects of a number of publications most of which have been recently discussed by Hargreaves and Richardson.[12] Two teams claimed mutarotation under their experimental conditions: Gulland and Mead [13] found such an effect with D-phenylalanine sodium salt and 2 : 3-, 2 : 5-, and 3 : 4-dimethoxybenzaldehyde in 50% aqueous ethanol, and Loiseleur and Crovisier [14] with amino-acids and formaldehyde at pH 7.

In a preliminary communication [15] we reported similar results with amino-acid esters, and (+)-amphetamine (2-amino-1-phenylpropane) in presence of ketones under practically anhydrous conditions. These observations, which are presented in Tables 1 and 2, indicated that various optically active α-amino-esters, one α-amino-amide, and one

[1] Robinson, "The Structural Relations of Natural Products," 1955, Clarendon Press, Oxford; Manske, *J.*, 1954, 2987; James, in "The Alkaloids, Chemistry and Physiology," ed. Manske and Holmes, 1950, Acad. Press, New York, Vol. I, p. 56.
[2] Williams *et al.*, "The Biochemistry of B Vitamins," Amer. Chem. Soc. Monograph, No. 110, 1950, p. 652.
[3] Snell, *J. Amer. Chem. Soc.*, 1945, **67**, 194.
[4] Metzler, Ikawa, and Snell, *ibid.*, 1954, **76**, 648.
[5] Christensen, *ibid.*, 1958, **80**, 99.
[6] Schönberg and Moubacher, *Chem. Rev.*, 1952, **50**, 261.
[7] Spenser, Crawhall, and Smyth, *Chem. and Ind.*, 1956, 796.
[8] Lewin, *Biochem. J.*, 1956, **63**, 14.
[9] Hurd and Buess, *J. Amer. Chem. Soc.*, 1956, **78**, 5667.
[10] Cohen and Witkop, *ibid.*, 1955, **77**, 6595; Anet, Bailey, and Robinson, *Chem. and Ind.*, 1953, 944; Leonard *et al.*, *J. Amer. Chem. Soc.*, 1954, **76**, 630, 3463, 5708; 1958, **80**, 4858.
[11] Grob and Meier, *Helv. Chim. Acta*, 1956, **39**, 776; Stoll, Hofmann, and Petrzilka, *ibid.*, 1951, **34**, 1544; Wrinch, *Nature*, 1957, **180**, 502; Hausmann, Weisiger, and Craig, *J. Amer. Chem. Soc.*, 1955, **77**, 723.
[12] Hargreaves and Richardson, *J.*, 1957, 3823.
[13] Gulland and Mead, *J.*, 1935, 210.
[14] Loiseleur and Crovisier, *Bull. Soc. Chim. biol.*, 1942, **24**, 241.
[15] Bergel and Lewis, *Chem. and Ind.*, 1955, 774.

optically active amine, when dissolved in ketones, interacted there, with the rotatory power of the solution being initially of the same order as in ethanol, and changing gradually. Table 1 shows that with acetone this occurred only if a primary amino-group was present. *N*-Monoalkyl, *NN*-dialkyl, and *N*-acyl derivatives and salts of the amino-compounds (some measured in ethyl methyl ketone) gave maximal values immediately, as did proline ethyl ester, a cyclic amino-ester (cf. Johnson and McCaldin on interaction of proline with

TABLE 1.

	Amino-derivative	$[\alpha]_D$ in EtOH	Max. $[\alpha]_D^{22}$ ($\pm 3°$) in ketone
1	L-Alanine Et ester	+3° (*c* 3·94)	−130° (*c* 5·0) in $COMe_2$
2	D-Alanine Et ester	−2 (*c* 3·24)	+127 (*c* 3·48) ,,
3	L-Phenylalanine Et ester	+22 (*c* 3·1)	−125 (*c* 3·25) ,,
4	D-Phenylalanine Et ester	−22 (*c* 3·0)	+118 (*c* 3·0) ,,
5	L-Tyrosine Et ester	+18 (*c* 3·2)	−124 (*c* 4·12) ,,
6	L-Tyrosine amide	−23 (*c* 0·075)	−74 (*c* 0·086) in 1 : 1 (v/v) $COMe_2$–MeOH
7	(+)-Amphetamine	+33 (*c* 3·87)	+84 (*c* 4·66) in $COMe_2$
8	*N*-Methyl-L-tyrosine Me ester	+32 (*c* 1·44)	+24 (*c* 1·48) ,,
9	*NN*-Dimethyl-L-tyrosine Et ester	+38 (*c* 2·05)	+20 (*c* 2·14) ,,
10	*N*-Formyl-L-tyrosine Et ester	+37 (*c* 3·1)	+37 (*c* 2·21) in COMeEt
11	*N*-Phthaloyl-L-tyrosine Et ester	−196 (*c* 1·9)	−197 (*c* 2·32) ,,
12	L-Tyrosine Et ester hydrochloride	+25 (*c* 2·0)	+25 (*c* 2·19) in 3 : 22 (v/v) $COMe_2$–EtOH
13	L-Proline Et ester	−40 (*c* 2·1)	−25 (*c* 2·2) in $COMe_2$

[a] In MeOH.

ninhydrin [16]). The amino-esters containing a primary amino-group, on the other hand, required under our experimental conditions about 1—3 hours at 20—25° before they had their maximal rotation, which for all the amino-esters tested was of the same order of magnitude. L-Tyrosine amide, because of its insolubility in pure acetone, was measured in 1 : 1 acetone–methanol mixture, there requiring several days to reach its maximal rotation which was lower than those of the esters. For (+)-amphetamine also the increment was of a lower order.

The relatively high values of [α] obtained with the enantiomorphs of alanine and phenylalanine esters probably offer a means of determining the absolute configuration of α-amino-acid derivatives, although there are not yet sufficient examples for certainty. But combined with the procedure of Lutz and Jirgensons,[17] which consists of measuring polarimetrically α-amino-acids in hydrochloric acid (the rotatory power of L-α-amino-acids is displaced in a positive direction), esterification of α-amino-acids, followed by determination of their maximal rotation in a ketone, should help to decide their absolute configuration; those belonging to the L-series should be relatively highly lævorotatory and those of the D-series highly dextrorotatory. This might be particularly useful in the case of amino-acids with two asymmetric centres. To test this "rule," *p*-di-(2-chloroethyl)amino-L-phenylalanine ethyl ester (Melphalan ester) [18] was measured; the specific rotation in ethanol was +6·5° ± 1°, in acetone −68° ± 2°, and in *cyclo*pentanone −86° ± 2°. The ethyl esters of (+)- and (−)-α-amino-γ-[*p*-di-(2-chloroethyl)aminophenyl]-butyric acids [19,20] were also investigated; these compounds had been tentatively assigned, by Smith and Luck,[20] the D- and the L-configuration respectively on the basis of their anti-tumour activities and the greater activity of the L-phenylalanine homologue (Melphalan). The specific rotation value for the (+)-ester in ethanol was +23° ± 1° but in *cyclo*pentanone −70° ± 2°; that of the (−)-ester in ethanol was −26° ± 1°, but in *cyclo*pentanone +73° ± 2°; thus their absolute configurations appear to be the opposite

[16] Johnson and McCaldin, *J.*, 1958, 817.
[17] Lutz and Jirgensons, *Ber.*, 1930, **63**, 448.
[18] Bergel and Stock, *J.*, 1954, 2409.
[19] Davis, Roberts, and Ross, *ibid.*, 1955, 890.
[20] Smith and Luck, *J. Org. Chem.*, 1958, **23**, 837.

to those deduced from the biological results, a possibility already referred to by Smith and Luck [20] when taking into account their measurements in acid (Lutz–Jirgenson's rule [17]).

TABLE 2. *Maximal* $[\alpha]_D^t$.

Et ester from	*cyclo*Pentanone (*c* 3—3·25; *t* 19·5—24°)	*cyclo*Hexanone (*c* 3—4·24; *t* 22—24·5°)	Acetone (*c* 3—4·12; *t* 22—25·5°)	Et Me ketone (*c* 3—4·2; *t* 22—24°)	Et_2 ketone (*c* 3—3·25; *t* 18—24°)
L-Tyrosine	−161°	−127°	−124°	−107°	−40°
L-Phenylalanine...	−162	−142	−125	−103	−39
D-Phenylalanine...	+153	+137	+118	+106	+40

In Table 2 the effects of different ketones on the rotation of L-tyrosine and L- and D-phenylalanine esters are recorded. The values recorded are the highest obtained, *i.e.*, for dried solvents; lower values were sometimes encountered when operations were carried out under conditions of greater humidity in the laboratory. The magnitude of the maximal $[\alpha]_D$ decreases for all three esters from left to right in the Table. The times required for attainment of maximal rotation were 30—300 minutes, except for *cyclo*hexanone where it was only 4—10 minutes, although previously [15] it had been found that commercial samples of *cyclo*hexanone required up to 30 minutes.

The first question for consideration is whether the recorded maximal $[\alpha]_D$ refer to end products or to equilibrium mixtures (the sequence $COMe_2 > COMeEt > COEt_2$, for instance, would be accounted for if steric hindrance by the larger ethyl groups prevented completion of the reaction). A complete answer may require study of the concurrent changes in pH and the effect of temperature (cf. Hargreaves and Richardson [12]); a kinetic study is in progress by one of us (G. E. L.), but results already available and reported below permit preliminary conclusions.

As can be seen from Table 3, diluting acetone, *cyclo*hexanone, or *cyclo*pentanone solutions of tyrosine ethyl ester with ethanol caused a drop in the maximal rotatory power. Adding small amounts of water to acetone, either before or after dissolving the amino-ester

TABLE 3.

Ketone (% v/v)	Ethanol (% v/v)	Water (% v/v)	$[\alpha]_D^t$, *t*	Tyrosine Et ester (*c*)
Acetone				
12	88	—	−104°, 22°	4·0
96	—	4 *	− 56·9,* 20	2·09
96	—	4	−54·5, 20	2·09
100	—	—	−124, 22	4·12
*cyclo*Pentanone				
50	50	—	−137, 21°	3·25
50	49	1	−132, 23·5	3·25
50	45	5	−117, 3·25	3·25
—	98·5	1·5	−18,† 24	4·5
100	—	—	−161, 19·5	3·25
*cyclo*Hexanone				
83·4	16·6	—	−119°, 22°	3·25
50	50	—	−118, 22·5	3·25
83·5	16	0·5	−111, 23	3·25
83·0	16	1·0	−101·5, 22·5	3·25
50	49	1·0	−105·5, 24	3·25
50	45	5·0	−66·5, 24	3·25
100	—	—	−127, 22	4·24

* Water added after the ester in dry acetone had been allowed to reach maximum rotation ($[\alpha]_D^{20}$ −126°), the value falling in 20 min. to the figure given.

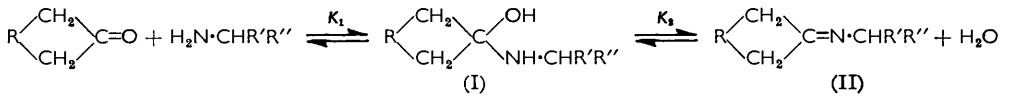
† From the solid Schiff's base, initial $[\alpha]_D^{24}$ −101°, falling to above value in 140 hr.

in it, produced even greater diminution. The values for solutions in mixtures of *cyclo*hexanone or *cyclo*pentanone with ethanol and water were also lowered. For the acetone and *cyclo*pentanone mixtures, maximal values were reached only after longer times than with the undiluted ketones, but with *cyclo*hexanone under our conditions dilution had little effect on this time.

After solutions of tyrosine ester in aliphatic ketones (average *c* 3·0 to 4·0) had been left at 20—25° for long enough to reach their maximal specific rotation, adding 50—60 times their volume of light petroleum afforded crystalline precipitates, which for diethyl

and ethyl methyl ketone represented 70—80% and for acetone *ca.* 50% of the quantity of the starting ester. This material consisted of L-tyrosine ethyl ester, characterised by m. p. and specific rotation (+16° ± 1°). When higher concentrations of the ester in diethyl ketone and a smaller volume of light petroleum were used, the mother-liquor retained a small negative rotation, although the crystalline precipitate, $[\alpha]_D^{24.5}$ +15·5° ± 0·5°, represented nearly 97% of the starting ester. When these procedures were carried out with *cyclo*hexanone solutions, the precipitates were mostly oils but in some instances these were mixed with crystals; with larger amounts of light petroleum the precipitate had low dextrorotatory values, with smaller volumes of light petroleum $[\alpha]_D$ of the oil in ethanol had a distinct negative value which slowly diminished at 25°; the rotation of the mother-liquor was also negative.

*cyclo*Pentanone solutions gave with light petroleum a crystalline compound, $[\alpha]_D^{21}$ −102° ± 2° (*c* 2·0 in ethanol), whose analyses were correct for *N-cyclo*pentylidene-L-tyrosine ethyl ester (II; R = $[CH_2]_2$, R′ = *p*-HO·C_6H_4·CH_2, R″ = CO_2Et). This compound gave on hydrogenation *N-cyclo*pentyl-L-tyrosine ester, but did apparently not react with toluene-*p*-thiol, which has been reported to form adducts with Schiff's bases.[21] When it was left at 24° in ethanol–water ($[\alpha]_D^{24}$ = −101°), its specific rotation fell in 140 hr. to −18° (see Table 3) and then precipitation with light petroleum afforded tyrosine ester. The infrared absorption spectrum of the *cyclo*pentylidene compound showed two double-bond frequencies, *viz.*, 1750 (ester C=O) and 1682 cm.$^{-1}$ (C=N, exocyclic to a five-membered ring [22]). Further infrared studies were carried out with *cyclo*hexanone and (+)-amphetamine, the latter being chosen because of the absence of the strongly absorbing ester group. *cyclo*Hexanone alone or mixed with various other solvents has a strong carbonyl absorption band near 1712 cm.$^{-1}$ (see Table 4, nos. 1—5; also Whiffen and Thompson [23]); amphetamine has two weak, primary amino-bands at 3358 and 3275 cm.$^{-1}$ (see Bellamy [24]) (these frequencies are somewhat higher for chloroform solutions; nos. 7 and 8). When the ketone and the amine were mixed in any proportion at 21° ± 3° the spectrum, initially the sum of those of the two compounds, rapidly changed: the strong band at 1712 cm.$^{-1}$ was replaced by one at 1655 cm.$^{-1}$ (cf. no. 6) of about half the intensity, and the two weak bands due to the amino-group were replaced by an intense broad band the position of which depended on the composition of the mixture (nos. 9—11).



These results, together with our synthesis of the *cyclo*pentylidenetyrosine ester, indicated a reaction between *cyclo*hexanone and amphetamine with elimination of water, leading to the azomethine (II; R = $[CH_2]_3$, R′ = $PhCH_2$, R″ = Me) in solution. The band at 1655 cm.$^{-1}$ was then due to the formation of the C=N bond.[22] The new strong broad band was caused by the production of water, the variation in frequency being due to differences in hydrogen-bonding in the different mixtures; in fact absorption at 3550 cm.$^{-1}$ was also obtained on adding water to pure *cyclo*hexanone (no. 13). In a mixture of the ketone and amine (no. 10), the water formed, being present in a relatively large quantity, absorbed approximately like pure water, namely at 3350 cm.$^{-1}$; [25] the absorption was at 3450 cm.$^{-1}$ when the amine was in excess (no. 11) because the small proportion of water was bonded to the amine. To test this interpretation the effect of drying agents was studied. When anhydrous magnesium sulphate or calcium sulphate was added to various mixtures of ketone and amine, absorption in the 3200—3500 cm.$^{-1}$ region was

[21] Stacy, Day, and Morath, *J. Amer. Chem. Soc.*, 1955, **77**, 3869.
[22] Cf. Jones and Sandorfy in " Chemical Application of Spectroscopy," Vol. IX of " Technique of Organic Chemistry," ed. West, Interscience Publ. Inc., New York, 1956, p. 532.
[23] Whiffen and Thompson, *J.*, 1946, 1005.
[24] Bellamy, " The Infra-red Spectra of Complex Molecules," Methuen and Co., London, 1954, p. 212.
[25] Gore, Barnes, and Peterson, *Analyt. Chem.*, 1949, **21**, 382.

reduced and the band near 1600 cm.$^{-1}$, where water also absorbs, disappeared. Removal of water might also be expected to change the equilibrium to give a greater concentration of azomethine (II); however, this was not proved, and for tyrosine ester and acetone (see Table 3) changes in rotation caused by addition of water could not be reversed simply by adding calcium sulphate.

Discussion and Conclusions.—Most of the azomethines described in the literature are formed from amino- and carbonyl compounds when at least one component contains an aromatic ring or the carbonyl group is that of an aldehyde. However, azomethines from ketones and non-aromatic amines are known, *e.g.*, Bergmann *et al.*[26] reported the preparation of such compounds from β-hydroxy-amines, and Witkop [27] discussed the properties of a Schiff's base from *cyclo*hexanone and *cyclo*hexylamine. Production of azomethines from amino-esters and aromatic aldehydes has been described by Velluz *et al.*,[28] Bergel and Cohen,[29] and Pfeiffer, Offermann, and Werner.[30] One could assume that analogous substances have been formed between amino-acids and carbonyl compounds in a number of reactions mentioned above. But in no case, to our knowledge, has the isolation of a simple condensation product from ketones been reported, at least not under our mild conditions.

Ingold [31] considered it plausible that H–N addition to carbonyl groups should commence with formation of amino-alcohols (I) which, in the case of primary amino-compounds, is followed by dehydration to the azomethines (II). This sequence is supported by our observations, and especially by the infrared measurements for *cyclo*hexanone–amphetamine. Here it is likely that the equilibrium constant K_2 for formation of the azomethine (II) and water is greater than K_1, as dilution of the mixture with chloroform rapidly regenerated the starting compounds, and the amino-alcohol (I), whose concentration at any time must have been very small, could not be detected. The difference in rate of reaction of tyrosine ester with *cyclo*hexanone and *cyclo*pentanone, and the stabilities of the azomethines formed, could be explained in terms of the strain theory [see Bellamy [24] (p. 128); and Brown, Brewster, and Shechter [32]] which postulates that the six-membered ring is less strained in a *cyclo*hexyl derivative than in a *cyclo*hexylidene compound while the reverse is true of the five-membered ring. Hence one could expect that *cyclo*hexanone would form an amino-alcohol, and hence an azomethine, more rapidly than would *cyclo*pentanone, but that the Schiff's base from the latter, once formed, would be more stable than that from the former. Moreover, in the case of the azomethine from *cyclo*hexanone the existence of an equilibrium between the C=N compound and an enamine tautomer (see Witkop [27]) might add further complications.

Experimental

Optical Rotations.—Measurements were for the sodium D line, with a 1 dm. polarimeter tube. A weighed amount of the amino-compound was dissolved in the ketone (2 ml.) or in the mixture of ketone with other solvents. The time of addition of the ketone was taken as zero time, and the first readings were taken usually about three minutes later.

Solvents.—Ethanol was boiled with sodium under reflux with diethyl phthalate and then distilled through a short column.[33]

Before polarimetry the ketones were warmed with small amounts of potassium permanganate until a permanent pink colour remained, dried (K_2CO_3; sometimes under reflux), and distilled through a short fractionating column. For the infrared measurements, *cyclo*hexanone was purified by formation of the bisulphite compound, which was washed with ethanol followed by

[26] Bergmann, Zimkin, and Pinchas, *Rec. Trav. chim.*, 1952, **71**, 186.
[27] Witkop, *J. Amer. Chem. Soc.*, 1956, **78**, 2873.
[28] Velluz, Amiard, and Heymès, *Bull. Soc. chim. France*, 1954, 1012.
[29] Bergel and Cohen, B.P. 556,044/1943.
[30] Pfeiffer, Offermann, and Werner, *J. prakt. Chem.*, 1942, **159**, 313.
[31] Ingold, " Structure and Mechanism in Organic Chemistry," Cornell Univ. Press, Ithaca, 1953, p. 687.
[32] Brown, Brewster, and Shechter, *J. Amer. Chem. Soc.*, 1954, **76**, 467.
[33] See Vogel, " Practical Organic Chemistry," Longmans, Green and Co., London, 1954, p. 166.

considerable amounts of ether to remove *cyclo*hexanol and then decomposed with 2N-sodium carbonate which was extracted with ether.

Amino-esters and Related Substances.—L-Tyrosine ethyl ester was either prepared in the usual manner from L-tyrosine or purchased from Kodak Ltd. and recrystallised from ethyl acetate.

Other amino-esters were obtained by refluxing the corresponding amino-acids in ethanolic hydrogen chloride. After removal of the solvent, the residual ester hydrochloride was treated with aqueous sodium carbonate or with triethylamine. In the former case the aqueous solution was extracted with ethyl acetate: after drying (Na_2SO_4), the solvent was removed *in vacuo*. In the latter case, the hydrochloride, dissolved or suspended in a small volume of ethanol-free chloroform, was treated with one equivalent of triethylamine, and the triethylamine hydrochloride precipitated by the addition of 5—10 volumes of dry ether. The solution was filtered and the solvent removed *in vacuo*. The free amino-esters were liberated from the hydrochloride only as required, to obviate dioxopiperazine formation.

NN-Dimethyl-L-tyrosine was prepared by reductive alkylation of L-tyrosine in the presence of formaldehyde.[34]

N-Methyl-L-tyrosine methyl ester was prepared by reductive alkylation of L-tyrosine methyl ester in methanol with one equivalent of aqueous formaldehyde.[34]

L-Tyrosine amide was prepared by the technique of Koenigs and Mylo.[35]

(+)-Amphetamine was prepared from the commercial sulphate by the method used for the amino-ester hydrochlorides.

Melphalan ethyl ester hydrochloride was prepared by Dr. J. A. Stock by treatment of Melphalan (CB 3025) with ethanolic hydrogen chloride at 5°; recrystallised from chloroform–ethanol, it had m. p. 165—167°.[36]

(+)- and (−)-α-Amino-γ-(*p*-di-2-chloroethylaminophenyl)butyric acid, kindly supplied by Professor J. M. Luck and Dr. H. E. Smith,[20] were esterified by Dr. J. A. Stock with saturated ethanolic hydrogen chloride at 24°. The ester hydrochlorides were precipitated with absolute ether as gums and, without further purification, submitted to paper-chromatography [Whatman No. 1 paper; butanol–ethanol–propionic acid–H_2O (10 : 5 : 2 : 5)] which showed them to be essentially homogeneous (R_F 0·88). The gums were transformed into the ester bases by treatment with aqueous sodium hydrogen carbonate and extraction with ethyl acetate. The dried oily residues from this solvent were then used for polarimetric measurements:

p-Di-2-chloroethylamino-L-phenylalanine ethyl ester, $[\alpha]_D^{24}$ +6·5° ± 1° (*c* 1·65 in ethanol), $[\alpha]_D^{24}$ −68° ± 2° (*c* 3·0 in acetone), $[\alpha]_D^{19}$ −86° ± 2° (*c* 2·4 in *cyclo*pentanone, after *ca.* 4 hr.).

Ethyl (+)-α-amino-γ-(*p*-di-2-chloroethylaminophenyl)butyrate, $[\alpha]_D^{24}$ +23° ± 1° (*c* 2·75 in ethanol), $[\alpha]_D^{24}$ −70° ± 2° (*c* 2·7 in *cyclo*pentanone, after *ca.* 2 hr.); lævorotatory isomer, $[\alpha]_D^{24}$ −26° ± 1° (*c* 2·85 in ethanol), $[\alpha]_D^{24}$ +73° ± 2° (*c* 2·85 in *cyclo*pentanone).

Precipitation of Ketonic Solutions with Light Petroleum.—To the solution of tyrosine ethyl ester in the ketone (approx. 2 ml.), remaining after polarimetric measurements had been completed, light petroleum (100—120 ml.) was added. The mixture was left at 0° overnight, and the precipitated crystals were filtered off and their melting point and rotation measured. The rotation of the mother-liquors was also measured, but was very low.

From ester (81 mg.) and acetone (2 ml.), crystals (42·9 mg., 53%) were isolated, having m. p. 98—104°, $[\alpha]_D^{24}$ +15·6° (*c* 2·37 in ethanol).

From ester (79·5 mg.) and ethyl methyl ketone (2 ml.), crystals (60 mg., 76%) were isolated, of m. p. 101—105°, $[\alpha]_D^{26}$ +16·3° (*c* 3·0 in ethanol).

From ester (80·3 mg.) and diethyl ketone (2 ml.), crystals (61·9 mg., 78%) were isolated, having m. p. 102—105°, $[\alpha]_D^{26}$ +16·5° (*c* 3·1 in ethanol).

From ester (65 mg.) and *cyclo*pentanone (2 ml.), crystals (47·4 mg., 73%) were isolated, having m. p. 115—129° (decomp.), $[\alpha]_D^{20}$ −100° (*c* 2·37 in ethanol) (see below).

From ester (80 mg.) and *cyclo*hexanone (2 ml.), oily material, $[\alpha]_D^{23}$ +5° (*c* 1·3 in ethanol) (18 mg., 23%), was produced.

The ester (120 mg.) was dissolved in diethyl ketone (1 ml.) which was mixed with light petroleum (24 ml.) and after 5 hr. at 0° the solution was decanted from the crystals {116 mg., 96·6%; $[\alpha]_D^{24.5}$ +15·5° (*c* 5·8 in ethanol)} and an aliquot part (*ca.* 1 ml.) was assayed polarimetrically (α − 0·03° ± 0·005°).

[34] Bowman and Stroud, *J.*, 1950, 1342.
[35] Koenigs and Mylo, *Ber.*, 1908, **41**, 4441.
[36] Personal communication.

To the ester (120 mg.) in *cyclo*hexanone (1 ml.) was added light petroleum (24 ml.). The turbid solution was left at 0° for *ca.* 3 hr., an oil and a few crystals being precipitated {122·7 mg.; $[\alpha]_D^{22\cdot5}$ $-48° \pm 1°$ (*c* 6·135 in ethanol), slowly diminishing}. The solution decanted from the oil had α — 0·055°. If the precipitate was a 1 : 1 mixture of *cyclo*hexylidene-L-tyrosine ethyl ester and the starting ester, the specific rotation (*c* 0·07 in light petroleum–*cyclo*hexanone) was $[\alpha]_D^{23}$ $-78·0° \pm 2°$.

N-cyclo*Pentylidene-L-tyrosine Ethyl Ester.*—Saturation of *cyclo*pentanone (5 ml.) with L-tyrosine ethyl ester (1·0 g.) at 20° gave after a few minutes a precipitate of colourless crystals (70%) which were washed with ether. The yield was increased by adding light petroleum to the mother-liquors which, like other solutions of this compound, became gradually yellow. Recrystallisation from *cyclo*pentanone–light petroleum (b. p. 100—120°) gave material of m. p. 132° (decomp.), $[\alpha]_D^{21}$ $-102°$ (*c* 2·0 in EtOH) (Found: C, 69·6; H, 7·6; N, 5·1. $C_{16}H_{21}O_3N$ requires C, 69·8; H, 7·7; N, 5·1%). This N-cyclo*pentylidene-L-tyrosine ethyl ester* was slowly degraded to tyrosine ester in ethanol–water (98·5 : 1·5).

Hydrogenation in ethanol with platinum black, addition of dilute aqueous hydrochloric acid, and removal of the solvent *in vacuo* gave a colourless solid which was precipitated from ethanol by ether as needles of N-cyclo*pentyl-L-tyrosine ethyl ester hydrochloride*, m. p. 208—209°, $[\alpha]_D^{24}$ $+33·5°$ (*c* 2·0 in ethanol) (Found: N, 4·5; Cl, 12·0. $C_{16}H_{24}O_3NCl$ requires N, 4·5; Cl, 11·3%).

The free *amino-ester* was liberated by aqueous sodium hydrogen carbonate; recrystallised from ethyl acetate–light petroleum, it had m. p. 138—139° (mixed m. p. with the *cyclo*pentylidene compound 110—115°), $[\alpha]_D^{24}$ $-1·5°$ (*c* 2·0 in ethanol) (Found: C, 68·8; H, 8·35; N, 5·0. $C_{16}H_{23}O_3N$ requires C, 69·3; H, 8·4; N, 5·05%).

TABLE 4. *Frequencies of infrared bands.*

No.	*cyclo*Hexanone (%)	Amphetamine (%)	Other solvent (% v/v)	ν (cm.$^{-1}$)
1	100	—	—	1712
2	5	—	CCl_4 95	1717
3	5	—	$CHCl_3$ 95	1705
4	5	—	C_6H_{13}·CHMe·OH 95	1708
5	5	—	NH_2Ph 95	1702
6	5	95	—	1710, 1655
7	—	100	—	3358, 3275
8	—	5	$CHCl_3$ 95	3368, 3297
9	90	10	—	*ca.* 3550
10	50	50	—	*ca.* 3350
11	10	90	—	*ca.* 3450
12	—	—	H_2O 0·15, in $CHCl_3$ 99·85	3681, 3603
13	90	—	H_2O 10	3550
14	—	—	C_6H_{13}·CHMe·OH 100	3350
15	—	—	C_6H_{13}·CHMe·OH 10 $CHCl_3$ 90%	3610, 3440
16	90	—	C_6H_{13}·CHMe·OH 10	3495

Infrared Absorption Measurements (see Table 4).—The *cyclo*hexanone–amphetamine experiments were carried out on a Perkin-Elmer Model 12C spectrometer, with a sodium chloride prism for the carbonyl band measurements and a lithium fluoride prism for those at 3000 cm.$^{-1}$. Pure *cyclo*hexanone was measured as a capillary film, and all other measurements were made in sealed cells of either 33 or 100 μ thickness. The chloroform used was washed with water, dried, and stored in a dark bottle.

Measurements of the *cyclo*pentylidene compound were made with a Perkin-Elmer Infracord spectrophotometer (sodium chloride prism), the sample being in a mull with liquid paraffin.

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