Summary

The electrophoretic mobility and heterogeneity of normal and digested human gamma globulin are increased by cysteine-hydrogen peroxide treatment. It is suggested that such mobility increases result from oxidative changes in the protein. Such treatment likewise causes a marked decrease in the antibody titers of human gamma globulin.

MADISON, WISCONSIN Received August 5, 1946

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Esters of α-Aminoisobutyric Acid

By Ralph A. Jacobson

Although the methyl and ethyl esters of the more common α-amino acids have been described, in many instances the corresponding esters of the higher alcohols are unknown. The work described in the present paper was undertaken in order to obtain information regarding the properties and, in particular, the stability toward cyclization of the higher esters of α-aminoisobutyric acid.

The preparation of α-aminoisobutyric acid in 30-33% yields by the hydrolysis of α-aminoisobutyronitrile with hydrobromic acid is described in "Organic Syntheses."² In the present study, 70-75% yields of the acid were obtained by the following two procedures: (a) the hydrolysis of α-aminoisobutyronitrile with 40% sulfuric acid according to the general procedure suggested by Cocker and Lapworth and (b) the hydrolysis of 5,5-dimethylhydantoin with 60% sulfuric acid according to the procedure of Bucherer and Steiner. Procedure (b) is to be recommended since 5,5-dimethylhydantoin is easily prepared in excellent yield.

The isobutyl, octyl and dodecyl esters of α-aminoisobutyric acid were prepared by esterification in the presence of hydrogen chloride. The hydrochlorides of the esters were first isolated for characterization and then converted to the free esters by treatment with alkali. The hydrochlorides of the esters are crystalline solids and the free esters are colorless liquids of pronounced basicity. The isobutyl ester is slightly soluble in water but the octyl and the dodecyl esters are practically insoluble. The hydrochlorides of the isobutyl and octyl esters are soluble but the dodecyl ester hydrochloride is insoluble in water.

In general, the methyl and ethyl esters of α-amino acids tend to cyclize to diketopiperazine derivatives or to form condensation products even at room temperature. The lower alcohol esters of glycine are particularly unstable and readily form glycine peptide ester chains of considerable length. The methyl ester of α-aminoisobutyric acid, (NH₂C(CH₃)₂COOCH₃), appears to be somewhat more stable possibly because of the doubly substituted alpha carbon. Thus Franchimont and Friedmann obtained tetramethyl diketopiperazine in only 34% yield even after prolonged heating of the ester at 230-240°. The present results indicate that the higher esters of α-aminoisobutyric acid possess still greater stability. At room temperature, samples of the isobutyl, octyl and dodecyl esters formed only a trace of solid material over a period of many months. At 210-220° the octyl ester yielded 3.0% of condensation product after twenty-two hours. At its boiling point (177-179°C) the isobutyl ester yielded only 0.91% of

(2) Lin and Li, J. Chinese Chem. Soc., 5, 88 (1938); C. A., 35, 6096 (1941), prepared the acid by a circuitous route.
(5) (a) Ref. 4; (b) Wagner and Buizer, "Organic Syntheses," 20, 42 (1940).
(6) Methyl α-aminoisobutyrate has been prepared by Franchimont and Friedmann, Rec. trav. chim., 27, 201 (1908), and ethyl α-aminoisobutyrate by Zeilinski and Kolikow, Z. physiol. Chem., 73, 459 (1911); Chem. Zentr., 83, 1319 (1911). The bactericidal properties of the hydrochloride of the dodecyl ester have been examined by Baker, Harrison and Miller, J. Expd. Med., 74, 611 (1941).
(7) (a) Frankel and Katchalski, THIS JOURNAL, 64, 2264 (1942); 64, 2268 (1942). (b) Pacsu, Nature, 144, 551 (1939); Pacsu and Wilson, J. Org. Chem., 7, 117 (1942); 7, 126 (1942).
Esters of \( \alpha \)-Aminoisobutyric Acid

Dec., 1946

Fig. 2.—Comparison of infra-red spectrograms of (A) reaction product of dodecyl ester of \( \alpha \)-aminoisobutyric acid and (B) tetramethyldiketopiperazine. Dry deposits on rock salt plates (about 1 mg./sq. cm.). Recorded by automatic rock salt prism spectrophotometer.

solid after twenty-two hours. A second sample, heated in a sealed tube at 240° for twenty-four hours, gave 3.1% of solid in comparison with 23.1% for the ethyl ester which was heated simultaneously. At 210–220°, the dodecyl ester yielded 0.7% of condensation product after twenty-four hours. The solubility characteristics, sublimation range, crystal structure (Fig. 1), chemical analysis and infrared spectrum (Fig. 2) corresponded to those of tetramethyldiketopiperazine. The octyl ester of \( \alpha \)-amino-\( \alpha \)-methylbutyric acid yielded only 0.50% of condensation product after twenty-four hours at 220°.

The effect upon stability of replacing one of the hydrogens on the nitrogen of an \( \alpha \)-aminoisobutyric ester is also of interest. The introduction of a phenyl group into the molecule would be expected to reduce the tendency toward cyclization or formation of condensation products. This appears to be the case, the isobutyl and octyl esters of \( N \)-phenyl-\( \alpha \)-aminoisobutyric acid exhibiting unusual stability. Both esters were recovered unchanged (except for darkening) after twenty-four hours at 210–220°.

Experimental

Preparation of 5,5-Dimethylhydantoin.—This compound was prepared by the procedure of Bucherer and Steiner.

\( \alpha \)-Aminoisobutyronitrile.—A modification of the procedure of Dubsky and Wensink was employed for the preparation of this compound.

A mixture of 2265 g. of acetone cyanhydrin and 7700 g. of liquid ammonia was allowed to stand in a closed container for several days. The ammonia was then distilled out, and solid potassium hydroxide added to the mixture. The upper layer was removed, dried with additional potassium hydroxide and distilled. The \( \alpha \)-aminoisobutyronitrile was obtained as a colorless liquid boiling at 60° under 21 mm. pressure; yield 1720 g. or 77% of the theoretical.

\( \alpha \)-Aminoisobutyronitrile was also prepared in a yield of 57% by passing ammonia gas into acetone cyanhydrin at atmospheric pressure.

Preparation of \( \alpha \)-Aminoisobutyric Acid by the Hydrolysis of 5,5-Dimethylhydantoin.—A solution of 142.5 g. of

(8) Dubsky and Wensink, Ber., 49, 1134 (1916).
5,5-dimethylhydantoin in 587 g. of 60% sulfuric acid was refluxed gently for twenty-four hours. Seven hundred sixty-three grams of barium carbonate was added slowly and a current of steam passed into the mixture on the steam-bath until ammonia was no longer evolved. The mixture was cooled with sulfuric acid bottle until acid to congo paper, filtered, and the solid on the filter washed several times by stirring with hot water. The filtrate and washings were concentrated to a volume of 1 liter. Basic lead carbonate was added until effervescence ceased and the mixture was no longer acid to congo paper but still acid to litmus. The lead sulfate was removed by filtration, washed with hot water, and the filtrate and washings concentrated until crystallization of the α-aminoisobutyric acid began. The yield from this and succeeding crops amounted to 98 g. or 76% of the theoretical amount.

### Preparation of α-Aminoisobutyric Acid by the Hydrolysis of α-Aminoisobutyronitrile

A solution of 84 g. of α-aminoisobutyronitrile in 1225 g. of 40% sulfuric acid was warmed gently for eight hours, during which the solid dissolved. The excess alcohol was recovered by vacuum distillation and the ester hydrochloride was obtained as a solid cake. Crystalization of a portion of this product from butyl acetate gave white crystals melting at 95°. The main portion was treated in the cold with a 20% solution of potassium hydroxide in the presence of ether. The ether layer was separated, dried with magnesium sulfate, and distilled. The dodecyl ester was obtained as a colorless liquid boiling at 130–132° under 2 mm.; yield 325 g. or 78.3% of the theoretical.

### Esters of α-Aminoisobutyric Acid

The method described below for the dodecyl ester of α-aminoisobutyric acid illustrates the general preparative procedure employed. The properties of the esters and their hydrochlorides are summarized in Table I.

#### Table I

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<td>Isobutyl α-aminoisobutyric Acid</td>
<td>C9H13O2N</td>
<td>C9H13O2N.HCl</td>
<td>C9H13O2N.HCl</td>
<td>120.5</td>
<td>85%</td>
<td>1.4330</td>
<td>102–103</td>
<td>1.4340</td>
<td>1.4499</td>
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<tr>
<td>Dodecyl α-aminoisobutyric Acid</td>
<td>C19H35O2N</td>
<td>C19H35O2N.HCl</td>
<td>C19H35O2N.HCl</td>
<td>210.5</td>
<td>95%</td>
<td>1.4330</td>
<td>102–103</td>
<td>1.4640</td>
<td>1.4699</td>
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### Notes

1. Isobutyl, octyl, and dodecyl esters of α-aminoisobutyric acid and the octyl ester of α-amino-α-methylbutyric acid have been prepared.
2. The higher esters of α-aminoisobutyric acid and α-amino-α-methylbutyric acid appear to be relatively resistant to cyclization.
3. The isobutyl and octyl esters of N-phenyl-α-aminoisobutyric acid have been prepared and shown to be unusually resistant to cyclization.

### Acknowledgment

The author is indebted to Dr. R. N. MacDonald for a specimen of tetramethyldiketopiperazine and to Drs. M. B. Hall and J. R. Downing for the infrared absorption spectra shown in Fig. 2.

### Summary

1. The isobutyl, octyl and dodecyl esters of α-aminoisobutyric acid and the octyl ester of α-amino-α-methylbutyric acid have been prepared.
2. The higher esters of α-aminoisobutyric acid and α-amino-α-methylbutyric acid appear to be relatively resistant to cyclization and intermolecular condensation.
3. The isobutyl and octyl esters of N-phenyl-α-aminoisobutyric acid have been prepared and shown to be unusually resistant to cyclization.