ture for 12 hours the crystalline methiodide was recovered and recrystallized from acetone. A further quantity of II was obtained from the mother liquors. The total yield of II was 7.95 g. (77%), m.p. 154–155° (dec.).

Anal. Calcd. for $C_{14}H_{27}O_5N_2I$ (430): C, 39.1; H, 6.3; N, 6.5. Found: C, 39.1; H, 6.4; N, 6.4.

γ-Carboxy-γ-aminopropyltrimethylammonium Iodide Hydroiodide (III).—II (2.7 g.) was refluxed with 20 ml. of 27% aqueous hydriodic acid for 2.5 hours and the excess hydriodic acid removed by distillation under reduced pressure and subsequent repeated distillations with water. The crystalline material thus obtained was recrystallized from aqueous hydriodic acid, sp. gr. 1.70, to give 2.0 g. (77%) of III, m.p. 211–212° (dec.).

Anal. Calcd. for $C_7H_{18}O_2N_2I_2$ (416): N, 6.7. Found: N, 7.0.

III was relatively unstable and was therefore characterized as the dipicrate, m.p. 208–209° (dec.).

Anal. Calcd. for $C_{19}H_{22}O_{16}N_8$ (618): C, 36.9; H, 3.6; N, 18.1. Found: C, 36.8; H, 3.5; N, 18.2.

1-Acetamido-1-carbethoxycyclopropane (IV).—To 2.0 g. of silver nitrate in 15 ml. of carbon dioxide-free water was added 2.0 ml. of a filtered 50% aqueous solution of sodium hydroxide, the resulting precipitate washed 6 times with carbon dioxide-free water and then added to 4.1 g. of II in 15 ml. of water. The reaction mixture was shaken for one-half hour, filtered, the clear filtrate lyophilized, and the residue transferred, with the aid of a small amount of ethanol, to a small distilling flask. The solvent was removed by distillation *in vacuo* and the quaternary base heated slowly to a bath temperature of 170–195° at a pressure of 1–2 mm. A thick yellow oil distilled at 110–115° (1–2 mm.) and crystallized upon cooling to give 0.75 g. (33%) of IV. This product was repeatedly recrystallized from a mixture of benzene and petroleum ether to give IV, m.p. 79–80°.

Anal. Calcd. for $C_8H_{13}O_3N$ (171): C, 56.1; H, 7.7; N, 8.2. Found: C, 56.2; H, 7.6; N, 8.1.

1-Acetamidocyclopropanecarboxylic Acid (V) (A).—An equimolar quantity of 0.163 *N* aqueous sodium hydroxide was added during a period of three-quarters of an hour to 0.2 g. of IV in 5 ml. of boiling water. An amount of aqueous hydrochloric acid equivalent to the amount of sodium hydroxide used was added and the reaction mixture evaporated to dryness under reduced pressure. The residue was dried *in vacuo* over phosphorus pentoxide and then extracted three times with anhydrous ether. The ethereal extract was evaporated to dryness and the residue recrystallized from ethyl acetate to give 0.1 g. (60%) of V, m.p. 158–159°.

Anal. Calcd. for $C_6H_9O_3N$ (143): C, 50.3; H, 6.3; N, 9.8. Found: C, 50.3; H, 6.3; N, 9.9.

(B).—To a solution of 0.5 g. of the hydrochloride of 1-aminocyclopropanecarboxylic acid[3] (VI) in 3 ml. of dry pyridine was added, at 0°, 1 ml. of acetic anhydride. After 60 hours the excess reagents were removed under reduced pressure, the residue extracted with ethyl acetate, and the extract concentrated to give V, m.p. 156–159° undepressed upon addition of V prepared as described above from IV.

1-Aminocyclopropanecarboxylic Acid Hydrochloride (VII).—V prepared from IV was refluxed for 5 hours with 2 *N* hydrochloric acid, the excess acid and solvent removed by distillation *in vacuo*, the residue dried over phosphorus pentoxide, and then dissolved in hot absolute ethanol. The addition of dry ether to the cold ethanol solution gave VII, m.p. 223–224° with decomposition both before and after recrystallization from acetic acid. When VII prepared as described above was mixed with an authentic sample of the hydrochloride[3] no depression of the decomposition point was observed.

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The Synthesis of D- and L-*threo*- and D- and L-*erythro*-α-Amino-β-hydroxy-*n*-caproic Acid[1]

BY ROBERT T. ADAMS[2] AND CARL NIEMANN

The four isomeric α-amino-β-hydroxy-*n*-caproic acids have been prepared from 2-hexenoic acid, and the configuration of each of these compounds has been determined.

In continuation of our studies on the 1,2,3-contiguously substituted dihydroxyamino-*n*-hexanes[3] the four isomeric α-amino-β-hydroxy-*n*-caproic acids have been prepared as starting materials for the synthesis of the isomeric 1,3-dihydroxy-2-amino-*n*-hexanes. The mixture of diastereoisomeric α-amino-β-methoxy-*n*-caproic acids, obtained as before from 2-hexenoic acid,[4,5] was benzoylated and the reaction product fractionally crystallized to give not only DL-*threo*-α-benzamido-β-methoxy-*n*-caproic acid (I) and DL-*erythro*-α-benzamido-β-hydroxy-*n*-caproic acid (II) but also (III).[6] The over-all yields of the three racemic acids, based upon 2-hexenoic acid were: I, 3.3%; II, 22.1%; and III, 1.7%. It is likely that III was formed from the corresponding α-amino-β-hydroxy acid since no special precautions were taken to exclude moisture from the methanolic solution of mercuric acetate used in the reaction with 2-hexenoic acid.[4,5]

The above three DL-acids were resolved *via* a papain-catalyzed conversion of the D-*threo* and L-*erythro* components to the corresponding *p*-toluides, the L-*threo* and D-*erythro* components being recovered unchanged.[7,8] The properties of the six resolution products, *i.e.*, D-*threo*-α-benzamido-β-methoxy-*p*-*n*-caprotoluide (IV), L-*threo*-α-benzamido-β-methoxy-*n*-caproic acid (V), L-*erythro*-α-benzamido-β-methoxy-*p*-*n*-caprotoluide (VI), D-*erythro*-α-benzamido-β-methoxy-*n*-caproic acid (VII), L-*erythro*-α-benzamido-β-hydroxy-*p*-*n*-caprotoluide (VIII), and D-*erythro*-α-benzamido-β-hydroxy-*n*-caproic acid (IX), are summarized in Table I. The isomeric α-amino-β-hydroxy-*n*-caproic acids, *i.e.*, D-*threo* (X), L-*threo* (XI), D-*erythro* (XII) and L-*erythro* (XIII), were obtained from the correspond-

(1) The prefixes *threo* and *erythro* define the relative configuration about the two asymmetric carbon atoms bearing the amino and hydroxyl groups; the letters D and L relate the configuration about the asymmetric carbon atom bearing the hydroxyl group with the configuration about the asymmetric carbon atom present in D- or L-glyceric aldehyde.

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(3) C. Niemann, A. A. Benson and J. F. Mead, *J. Org. Chem.*, **8**, 397 (1943).

(4) C. Niemann and C. T. Redemann, THIS JOURNAL, **68**, 1932 (1946).

(5) H. D. West and H. E. Carter, *Org. Syntheses*, **20**, 101 (1940).

(6) The configurations of these acids, and of other intermediates to be described, were assigned on the basis of arguments given later in this communication.

(7) E. L. Bennett and C. Niemann, THIS JOURNAL, **72**, 1798 (1950).

(8) W. H. Schuller and C. Niemann, *ibid.*, **73**, 1644 (1951).

ing resolution products by hydrolysis with 48% aqueous hydrobromic acid for the methoxy series and with 20% aqueous hydrochloric acid for the hydroxy series. The properties of these acids are given in Tables II and III.

TABLE I

ENZYMATIC RESOLUTION OF THE α-BENZAMIDO-β-METHOXY-, AND α-HYDROXY-, *n*-CAPROIC ACIDS

DL-Mixture	M.p., °C.	Resolution product	M.p., °C.	$[\alpha]_D$
I	120–121	IV	176–177	+17.3[a]
		V	151–152	−51.4[b]
II	111.5–112	VI	211–212	−14.0[c]
		VII	124–125	−14.0[d]
III	177.5–178.5	VIII	224–225	−27.2[e]
		IX	164.5–165	−34.1[f]

[a] c = 9.79% in pyridine, t = 25.0°. [b] c = 5.06% in absolute ethanol, t = 25.0°. [c] c = 5.58% in pyridine, t = 25.0°. [d] c = 3.42% in absolute ethanol, t = 25.0°. [e] c = 3.42% in pyridine, t = 27.0°. [f] c = 4.32% in absolute ethanol, t = 23.5°.

TABLE II

PROPERTIES OF THE DIASTEREOISOMERIC α-AMINO-β-HYDROXY-*n*-CAPROIC ACIDS

Compound	Configuration	M.p., °C. (dec.)	$[\alpha]_D$ in water	$[\alpha]_D$ in 6*N* HCl
X[a]	D-*threo*	184–188	+4.6[b]	+18.6[c]
XI[d]	L-*threo*	185–188	−4.6[e]	−18.5[f]
XII[g]	D-*erythro*	198–202	+2.0[h]	−27.4[i]
XIII[j]	L-*erythro*	203–205	−2.0[k]	+27.1[l]
XIII[m]	L-*erythro*	203–204	−2.1[n]	

[a] From IV. [b] c = 3.46%, t = 25.0°. [c] c = 2.42% in 6.07 *N* hydrochloric acid, t = 25.0°. [d] from V. [e] c = 3.50%, t = 25.0°. [f] c = 2.28% in 6.03 *N* hydrochloric acid, t = 25.0°. [g] from IX. [h] c = 8.81%, t = 26.0°. [i] c = 2.63% in 6.07 *N* hydrochloric acid, t = 25.0°. [j] from VIII. [k] c = 5.87%, t = 22.0°. [l] c = 2.47% in 6.07 *N* hydrochloric acid, t = 25.0°. [m] from VI. [n] c = 3.39%, t = 22.0°.

TABLE III

ELEMENTARY COMPOSITION OF THE α-AMINO-β-HYDROXY-*n*-CAPROIC ACIDS

	Analyses, %					
	Calcd.			Found		
Compound	C	H	N	C	H	N
X	49.0	8.9	9.5	48.9	8.8	9.5
XI	49.0	8.9	9.5	48.9	8.9	9.4
XII	49.0	8.9	9.5	48.9	8.8	9.5
XIII[a]	49.0	8.9	9.5	48.9	8.8	9.4
XIII[b]	49.0	8.9	9.5	48.8	8.9	9.5

[a] From VI. [b] From VIII.

The proof of the configurations of X, XI, XII and XIII rests upon three lines of experimental evidence: one, the stereochemical specificity of the enzymatic resolution; two, the optical rotatory properties of neutral and acidic solutions of the acids; and three, the oxidative degradation of XI to an optically active α-hydroxy-*n*-valeric acid.

The papain-catalyzed synthesis of aryl substituted α-acylaminoacid amides and hydrazides[9] was originally believed to be limited to the L-isomers. Although it is now known[7,8] that with certain α-acylaminoacids a substantial loss in stereochemical specificity may be observed, the fact remains, that in no case has the reaction been observed to be limited to the D-isomers. Thus if only one isomer can be isolated from the reaction mixture all evidence is consistent with the proposition that this isomer probably possesses the L-configuration. Therefore, in respect to the configuration about the α-carbon atom it is reasonable to conclude that X and XIII, which were obtained from IV, and VI (and VIII), respectively, are the L-isomers and that XI and XII are the D-isomers.

Lutz and Jirgensons[10] have shown that the specific rotation of an α-amino acid in aqueous solution is dependent, within limits, upon the *p*H of the solution, and that with increasing acid concentration the specific rotation changes in a positive sense for L-antipodes and in a negative sense for D-antipodes. When these rules are applied to the isomeric α-amino-β-hydroxy-*n*-caproic acids it is seen (*cf.* Table II) that X and XIII exhibit the behavior expected of L-antipodes and XI and XII that of D-antipodes. Thus two independent arguments lead to the same conclusion in respect to the configuration about the α-carbon atoms of the four isomeric acids.

The configuration about the β-carbon atom of each of the α-amino-β-hydroxy-*n*-caproic acids was established by oxidative degradation, following the method of Meyer and Rose.[11] Oxidation of XI with Chloramine-T gave an optically active aldehyde which in turn was oxidized with bromine water to give an optically active α-hydroxy-*n*-valeric acid which was isolated as the levorotatory barium salt. Levene and Haller[12] have shown that the barium salt of L-α-hydroxy-*n*-valeric acid is levorotatory. When an equivalent amount of hydrochloric acid was added to an aqueous solution of the barium salt prepared from XI, the optical rotation was observed to increase in a positive sense as would be expected from the rule formulated by Levene, Mori and Mikeska[13] for α-hydroxy acids of the L-series. Therefore, it can be concluded that in XI the configuration about the β-carbon atom is L, about the α-carbon atom is D and that XI is L-*threo*-α-amino-β-hydroxy-*n*-caproic acid. It follows that the enantiomorphic acid X is D-*threo*-α-amino-β-hydroxy-*n*-caproic acid and that the remaining two acids XII and XIII must possess the *erythro* configuration. Since XII was shown to have the D- and XIII the L-configuration about the α-carbon atom, these two acids are the D-*erythro*- and L-*erythro*-α-amino-β-hydroxy-*n*-caproic acids, respectively.

As for the *p*-toluides IV, VI and VIII, and the acids V, VII and IX, the nature of the resolution appears to provide sufficient basis for assigning the former compounds to the L-series and the latter compounds to the D-series in respect to configuration about the α-carbon atom. In view of the fact that the assignment of configuration about the β-carbon atom of these compounds is dependent upon retention of configuration during hydrolysis to the corresponding α-amino-β-hydroxy-*n*-caproic acids it is appropriate to point out that in the hydrolysis of O-methylthreonine and allothreonine

(9) M. Bergmann and H. Frankel-Conrat, *J. Biol. Chem.*, **119**, 707 (1937).

(10) O. Lutz and B. Jirgensons, *Ber.*, **63**, 448 (1930); **64**, 1221 (1931).

(11) C. E. Meyer and W. C. Rose, *J. Biol. Chem.*, **115**, 721 (1936).

(12) P. A. Levene and H. L. Haller, *ibid.*, **77**, 555 (1928).

(13) P. A. Levene, T. Mori and L. Mikeska, *ibid.*, **75**, 337 (1927).

with 48% aqueous hydrobromic acid no replacement of the methoxy by a bromine atom could be detected.[5,14,15]

In conclusion it should be noted that the application of Hudson's isorotation rules[16] to neutral and acidic aqueous solutions of the isomeric α-amino-β-hydroxy-*n*-caproic acids indicates that these compounds obey not only the Lutz–Jirgensons generalization,[10] but also that of Levene, Mori and Mikeska,[13] in that the partial rotation of the L-amino group becomes more positive, and that of the D-hydroxyl group more negative, on passing from neutral to acidic solutions.

Experimental[17,18]

α-Bromo-β-methoxy-*n*-caproic Acid.[4]—From 220 g. of 2-hexenoic acid, m.p. 30–32°, prepared from *n*-butyraldehyde and malonic acid in 67% yield,[4] there was obtained[4] 345–360 g. (79–83%) of crude oily α-bromo-β-methoxy-*n*-caproic acid.

α-Amino-β-methoxy-*n*-caproic Acid.—A solution of 667 g. (2.96 moles) of crude α-bromo-β-methoxy-*n*-caproic acid in 6.7 l. of concentrated ammonium hydroxide (sp. gr. 0.90) was heated at 85° in sealed bottles for 22 hours,[5] the solution cooled, concentrated under reduced pressure to a volume of 2 l., filtered and evaporated to dryness. Water (1 l.) was added and the solution again evaporated to dryness. The gummy residue was triturated with 1.5 l. of acetone and the mixture allowed to stand for two days. The solid was collected, washed with acetone, and dried to give 414 g. of crude α-amino-β-methoxy-*n*-caproic acid. The mother liquor and washings were combined, evaporated to dryness under reduced pressure, and the residue allowed to stand under acetone (1 l.) for an additional two days. Further crystallization occurred to give an additional 136 g. of the crude amino acid.

DL-*threo*-α-Benzamido-β-methoxy-*n*-caproic Acid (I), DL-*erythro*-α-Benzamido-β-methoxy-*n*-caproic Acid (II), and DL-*erythro*-α-Benzamido-β-hydroxy-*n*-caproic Acid (III).—A solution of 256 g. of crude α-amino-β-methoxy-*n*-caproic acid (the ammonolysis product from 1.38 moles of the bromo acid) and 110.4 g. (2.76 moles) of sodium hydroxide in 1 l. of water was distilled at reduced pressure until the ammonia was expelled. The solution was then maintained at 0–10° for two hours, during the first hour of which 194 g. (1.38 moles) of benzoyl chloride was added dropwise, as well as sufficient 5% aqueous sodium hydroxide to prevent the *p*H of the reaction mixture from falling below 8. The reaction mixture was then acidified to *p*H 1–2 by the addition of concentrated hydrochloric acid whereupon a light yellow oil separated which crystallized after standing at 4° for three days. This product was collected, dried, and added to 400 ml. of hot diisopropyl ether. An insoluble residue remained which was recrystallized from dioxane to give 7.2 g. of III, colorless rectangular plates, m.p. 177.5–178.5°, with slow decomposition when held at the melting point.

Anal. Calcd. for $C_{13}H_{17}O_4N$ (251): C, 62.1; H, 6.8; N, 5.6. Found: C, 62.3; H, 6.8; N, 5.7.

The isopropyl ether solution was concentrated *in vacuo* to *ca.* 250 ml. and the solution allowed to stand at 4°. After three days the solid was collected, dried, and recrystallized three times, once from chloroform, once from dioxane, and once from diisopropyl ether, to give 14.6 g. (4% based on the bromo acid) of I, square plates, m.p. 120–121°.

Anal. Calcd. for $C_{14}H_{19}O_4N$ (265): C, 63.4; H, 7.2; N, 5.3. Found: C, 63.4; H, 7.3; N, 5.3.

The mother liquors from the dioxane, chloroform and isopropyl ether recrystallizations were combined and evaporated to dryness *in vacuo*. The residue was extracted with five 600-ml. portions of hot petroleum ether (b.p. 60–70°), the petroleum ether insoluble fraction dissolved in the minimum quantity of hot isopropyl ether, and the solution allowed to stand at 4°. Crystallization occurred over a period of ten days. The solid was collected, washed with fresh solvent, and again recrystallized from diisopropyl ether to give 88.4 g. (24% based on the bromo acid) of II, soft white needles, m.p. 111.5–112.0°. Concentration of the combined mother liquors and washings gave an additional 9.1 g. (2.5%) of II, m.p. 107–109°, and 23.4 g. of a viscous red sirup which could not be crystallized.

Anal. Calcd. for $C_{14}H_{19}O_4N$ (265): C, 63.4; H, 7.2; N, 5.3. Found: C, 63.2; H, 7.3; N, 5.2.

D-*threo*-α-Benzamido-β-methoxy-*p*-*n*-caprotoluide (IV).—A solution of 9.94 g. (0.0375 mole) of I, 4.3 g. (0.04 mole) of *p*-toluidine, 2.0 g. of cysteine hydrochloride and 2.1 g. of purified papain[7] in 487 ml. of 0.5 *M* acetic acid–0.5 *M* sodium acetate buffer was prepared. The *p*H was adjusted to 4.6 at 35° by the addition of 10 ml. of 2.0 *N* sodium hydroxide and the solution incubated at 40° for approximately four days. The precipitated toluide (6.33 g.) was collected, washed with fresh buffer solution and dried. To the filtrate was added 0.5 g. of cysteine hydrochloride and 1.0 g. (0.0093 mole) of *p*-toluidine, and the *p*H adjusted to 4.60, at 28°, by the addition of glacial acetic acid, and the solution incubated at 40° for an additional four days to give a second crop (0.27 g.) of crude toluide. The first and second crops of crude toluide were recrystallized from toluene to give 5.52 g. (83%) of IV, white needles, m.p. 176–177°. The specific rotation was unchanged by further recrystallization from toluene.

Anal. Calcd. for $C_{21}H_{26}O_3N_2$ (354): C, 71.2; H, 7.4, N, 7.9. Found: C, 71.2; H, 7.3; N, 7.9.

L-*threo*-α-Benzamido-β-methoxy-*n*-caproic Acid (V).—The filtrate remaining after the removal of IV was acidified to *p*H 1–2 by the addition of concentrated hydrochloric acid, the precipitated acid collected, washed with water, and dried to give 4.25 g. of crude V, m.p. 149–150°. The filtrate was evaporated to dryness *in vacuo* and the residue extracted with 200 ml. of hot ethanol. The ethanol solution was evaporated to dryness, the residue taken up in chloroform, and the chloroform solution extracted with 5% aqueous sodium bicarbonate. The bicarbonate phase was separated and acidified to *p*H 1–2 by addition of concentrated hydrochloric acid. The oil which separated was taken up in chloroform, the solution dried over anhydrous sodium sulfate, and the solvent evaporated to give a sirupy residue which crystallized when shaken with 5 ml. of diisopropyl ether. The crystalline material was collected, washed and dried to give 0.33 g. of crude V. The crude V was recrystallized from diisopropyl ether to give 3.60 g. (72%) of V, transparent cubes, m.p. 151–152°. The specific rotation was unchanged by further recrystallization from diisopropyl ether.

Anal. Calcd. for $C_{14}H_{19}O_4N$ (265): C, 63.4; H, 7.2; N, 5.3. Found: C, 63.3; H, 7.3; N, 5.3.

L-*erythro*-α-Benzamido-β-methoxy-*p*-*n*-caprotoluide (VI).—A mixture of 5.0 g. (0.0181 mole) of II, 2.15 g. (0.02 mole) of *p*-toluidine, 1.0 g. of cysteine hydrochloride and 180 ml. of 0.5 *M* acetic acid–0.5 *M* sodium acetate buffer was warmed to 40° to effect solution and the *p*H adjusted to 4.6 by the addition of 20 ml. of 1 *N* sodium hydroxide. An enzyme solution prepared by the extraction of 7.0 g. of pulverized crude papain (Wallenstein "Hygrade") with 45 ml. of water at 4° for four hours was then added. After 4.5 days incubation at 40° there was obtained 2.11 g. of crude VI. Incubation for two more days gave an additional 0.25 g. of crude VI. The crude VI was recrystallized from acetonitrile and dried *in vacuo* at 55° to give 1.68 g. (50%) of VI, white needles, m.p. 211–212°. The specific rotation was unaltered by further recrystallization from absolute ethanol.

Anal. Calcd. for $C_{21}H_{26}O_3N_2$ (354): C, 71.2; H, 7.4; N, 7.9. Found: C, 71.3; H, 7.4; N, 7.8.

D-*erythro*-α-Benzamido-β-methoxy-*n*-caproic Acid (VII).—The filtrate remaining after the removal of VI was evaporated to dryness at reduced pressure. Water (100 ml.) was added, the mixture filtered, and the filtrate acidified to *p*H 1–2 by the addition of concentrated hydrochloric acid. The acid which separated as an oil soon crystallized and was recrystallized twice, once from diisopropyl ether, and once, from a 1:1 mixture of petroleum ether (b.p. 60–70°) and chloroform

(14) M. L. Wood, R. J. Madden and H. E. Carter, *J. Biol. Chem.*, **117**, 1 (1937).

(15) H. D. West and H. E. Carter, *ibid.*, **119**, 103, 109 (1937); **122**, 611 (1937–1938).

(16) *Cf.* F. J. Bates, *et al.*, "Polarimetry, Saccharimetry and the Sugars," National Bureau of Standards Circular C440, Washington, D. C., 1942, pp. 432–435.

(17) Microanalyses by Dr. A. Elek.

(18) All melting points are corrected; rate of heating 1°/min.

to give 1.30 g. (52%) of VII, needles, m.p. 124–125°. The specific rotation was unchanged by further recrystallization from the mixture of petroleum ether and chloroform.

Anal. Calcd. for $C_{14}H_{19}O_4N$ (265): C, 63.4; H, 7.2; N, 5.3. Found: C, 63.2; H, 7.3; N, 5.2.

L-*erythro*-α-**Benzamido-β-hydroxy-*p*-*n*-caprotoluide (VIII).**—A solution of 4.42 g. (0.0176 mole) of III, 2.14 g. (0.02 mole) of *p*-toluidine, 2.0 g. of cysteine hydrochloride and 2.3 g. of purified papain[7] in 470 ml. of 0.5 *M* acetic acid–0.5 *M* sodium acetate buffer was prepared and the *p*H adjusted to 4.6, at 40°, by the addition of 17 ml. of 2.02 *N* sodium hydroxide. Incubation at 40° for 5.5 days gave 3.15 g. of crude VIII. To the filtrate was added 0.50 g. of cysteine hydrochloride and 0.63 g. of *p*-toluidine, and the *p*H adjusted to 4.6 by the addition of glacial acetic acid. Incubation at 40° for two days gave an additional 0.09 g. of crude VIII. The crude VIII was recrystallized twice from a 1:1 mixture of acetonitrile and dioxane to give 2.05 g. (68.5%) of VIII, fine white needles, m.p. 224–225°. The specific rotation was unaltered by further recrystallization from the above mixture.

Anal. Calcd. for $C_{20}H_{24}O_3N_2$ (340): C, 70.6; H, 7.1; N, 8.2. Found: C, 70.6; H, 7.2; N, 8.3.

D-*erythro*-α-**Benzamido-β-hydroxy-*n*-caproic Acid (IX).**—The filtrate remaining after recovery of VIII was acidified to *p*H 1–2 with concentrated hydrochloric acid and allowed to stand for 24 hours at 4°. The crystalline solid which separated was collected, washed with water, and dried to give 1.28 g. of nearly pure IX, $[\alpha]^{25}D$ −33.5° (*c*, 6.80% in absolute ethanol). The filtrate was extracted continuously with chloroform for 30 hours, the chloroform solution extracted with 5% sodium bicarbonate, the bicarbonate phase acidified to *p*H 1–2 with concentrated hydrochloric acid and the crystalline precipitate collected to give, after washing and drying, 0.62 g. of crude IX. The combined crops were recrystallized from a 1:1 mixture of acetonitrile and dioxane to give 1.69 g. (76.5%) of IX, clusters of soft white needles, m.p. 164.5–165°. Further recrystallization from the same solvent caused no change in the specific rotation.

Anal. Calcd. for $C_{13}H_{17}O_4N$ (251): C, 62.1; H, 6.8; N, 5.6. Found: C, 62.0; H, 6.8; N, 5.6.

The α-Amino-β-hydroxy-*n*-caproic Acids (X–XIII).—The products from the enzymatic resolutions were hydrolyzed by refluxing either with 20% hydrochloric acid or with 48% hydrobromic acid, as indicated below. The resulting solutions were evaporated to dryness *in vacuo*, water (10–15 ml.) added, and the solutions again evaporated to dryness. The residues were extracted with water (10–20 ml.) and the extracts treated successively with freshly precipitated silver carbonate and hydrogen sulfide. The clear, colorless, halogen-free solutions were evaporated to dryness *in vacuo*, the residues recrystallized from 70% ethanol and dried *in vacuo* over phosphorus pentoxide at room temperature. The specific rotations of the acids (*cf.* Table II) were unchanged upon further recrystallization from 70% ethanol. The analytical results are summarized in Table III. A mixture of 2.10 g. of IV and 20 ml. of 48% hydrobromic acid was refluxed for 2.5 hours to give 734 mg. (84%) of X, soft white needles, m.p. 184–188°, with decomposition and sintering from 180°, when the sample was placed in the bath at 174°. Hydrolysis of V (1.46 g.) with 48% hydrobromic acid (15 ml.) for three and one-quarter hours gave 768 mg. (95%) of XI, white needles, m.p. 185–188° ((dec.), with gradual discoloration observed from 179° when the sample was introduced in the bath at 174°). Hydrolysis of 380 mg. of VI with 5 ml. of 48% hydrobromic acid for three hours gave 148 mg. (94%) of XIII, soft white needles, m.p. 203–204°, with decomposition and discoloration observed at 199° when the sample was placed in the bath at 195°. Hydrolysis of 927 mg. of VIII with 65 ml. of 20% hydrochloric acid for 11 hours gave 384 mg. (96%) of XIII, clusters of soft white needles, m.p. 203–205° ((dec.), sample introduced at 195°). IX (494 mg.) was refluxed with 15 ml. of 20% hydrochloric acid for six hours to give 230 mg. (80%) of XII, soft white needles, m.p. 198–202° (with decomposition and discoloration observed at 196.5° when the sample was placed in the bath at 191°).

Oxidative Degradation of L-*threo*-α-Amino-β-hydroxy-*n*-caproic Acid (XI).[11]—A solution of 414 mg. (0.0018 mole) of Chloramine-T in 3 ml. of water was heated to 85° and added with efficient stirring to a solution of 206.6 mg. (0.0014 mole) of XI in 4 ml. of water at 25°. Within ten seconds the formation of a copious white precipitate was observed. After 16 minutes the stirred reaction mixture was cooled in an ice-bath until precipitation was complete, the mixture filtered, and the filter cake washed thoroughly with cold water. The combined washings and filtrate were acidified to *p*H 1–2 by the addition of 0.2 ml. of 6 *N* hydrochloric acid and again filtered. A solution of bromine water containing 0.22 g. (0.0014 mole) of bromine was added to the filtrate and the mixture allowed to stand at room temperature in a stoppered vessel for 16 hours. The solution was concentrated at reduced pressure to a volume of 20 ml., extracted with seven 30-ml. portions of ether, the combined ethereal extracts dried over anhydrous sodium sulfate, the solvent removed, and the residue dissolved in 6 ml. of water. This solution was neutralized to a phenolphthalein end-point by the addition of 0.518 *N* barium hydroxide (1.75 ml. was required). Following concentration under reduced pressure to a volume of 2 ml., four volumes of ethanol were added to the hot aqueous solution. Barium α-hydroxy-*n*-valerate separated as colorless rectangular plates. The crystals were collected, washed with a small quantity of 80% ethanol, and dissolved in 1 ml. of hot water. The solution was filtered while hot and 4 ml. of absolute ethanol added. The solution was cooled and allowed to stand until crystallization was complete, the product collected, washed with 80% ethanol, and dried *in vacuo* at 110° over phosphorus pentoxide for 15 hours. The anhydrous barium salt had a specific rotation of $[\alpha]^{20}D$ −11.0° (*c*, 3.18% in water). The salt was recrystallized and dried as before to give 133 mg. (51%) of anhydrous barium α-hydroxy-*n*-valerate. The specific rotation was unchanged by this recrystallization.

Anal. Calcd. for $BaC_{10}H_{18}O_6$ (372): Ba, 37.0; C, 32.3; H, 4.9. Found: Ba, 36.9; C, 32.3; H, 5.0.

To 12.5 mg. (0.0337 millimole) of the anhydrous barium salt was added 0.134 ml. of 1.008 *N* hydrochloric acid (0.135 milliequiv.) and sufficient water to bring the total volume to 1.00 ml. The specific rotation of this solution, calculated for the free hydroxy acid in the presence of an equivalent amount of barium chloride, was $[\alpha]^{20}D$ −2° (*c*, 0.081% in 0.067 *N* hydrochloric acid).

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