Anal. Calcd. for $C_{11}H_{11}O_5N$: C, 56.60; H, 4.72; N, 18.00. Found: C, 56.44; H, 5.01; N, 17.97.

(b) **From 1-Phenyl-4,4-dicarbethoxy-2-azetidinone (V).**—To an alcoholic solution of 2 g. of V was added 40 ml. of alcohol saturated with ammonia. After one week, the solvent was removed, leaving 1.57 g. (98%) of a colorless solid, m.p. 255–256° (dec.). A sample recrystallized from alcohol, m.p. 258.5–260° (dec.), when mixed with the diamide (XIV) prepared by procedure (a), showed no depression in melting point.

1-Phenyl-4-carboxanilido-2-azetidinone (XVII).—A suspension of 0.150 g. of 1-phenyl-4,4-dicarboxy-2-azetidinone (XVI) in benzene was heated overnight under reflux with 0.190 g. of aniline and 0.05 ml. of phosphorus trichloride. Removal of benzene and digestion with a 5% solution of sodium bicarbonate gave a colorless solid (0.158 g., 93.5%), m.p. 247–251°. Recrystallization from alcohol gave colorless needles, m.p. 254–255° (80% recovery). The m.p. was constant at 254.5–255.5° on further recrystallization.

Anal. Calcd. for $C_{16}H_{14}O_2N_2$: C, 72.16; H, 5.30; N, 10.52. Found[16]: C, 71.96; H, 5.07; N, 10.90.

1-Cyclohexyl-4,4-dicarboxamido-2-azetidinone (XVIII).—To an alcoholic solution of 0.55 g. of 1-cyclohexyl-4,4-dicarbethoxy-2-azetidinone (IV) was added 20 ml. of alcohol saturated with ammonia. After one week, the solvent and excess ammonia were removed under reduced pressure. A colorless, crystalline solid (0.44 g., 100%), m.p. 165–171°, was obtained. After several recrystallizations from a mixture of acetone and cyclohexane, colorless needles, m.p. 190–192°, 77% recovery, were obtained. An analytical sample, m.p. 194–195°, was prepared by further recrystallization.

Anal. Calcd. for $C_{11}H_{17}O_3N_3$: C, 55.22; H, 7.16; N, 17.97. Found: C, 55.50; H, 7.32; N, 17.76.

Infrared Absorption Spectra.—The infrared absorption spectra were determined with a Baird Infrared Spectrophotometer, Model B. For curves in Fig. 1, the following solvents and concentrations were used: A, carbon tetrachloride, 5%; B, C, D, E and F, chloroform, 5%.

For curves in Fig. 2, the following solvents and concentrations were used: A, nujol mull; B, dioxane, 1.7%; C, dioxane, 3.8%; D, nujol mull.

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[16] A trace of ash was obtained on combustion; the percentages are corrected values.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, HOPE COLLEGE]

A Synthesis of Hydroxylysine

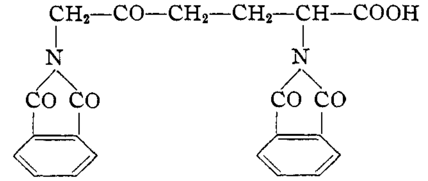
BY GERRIT VAN ZYL, EUGENE E. VAN TAMELEN AND GEORGE D. ZUIDEMA

The synthesis of hydroxylysine (I) has been accomplished by the catalytic reduction and subsequent hydrolysis of diethyl (γ-hydroxy-δ-nitro-*n*-butyl)-acetamidomalonate, which was obtained directly by condensing diethyl acetamidomalonate, acrolein and nitromethane. An explanation accounting for the racemization of hydroxylysine during its isolation from natural sources is proposed.

The amino acid hydroxylysine (I) has been isolated from isinglass by S. B. Schryver, H. W. Buston

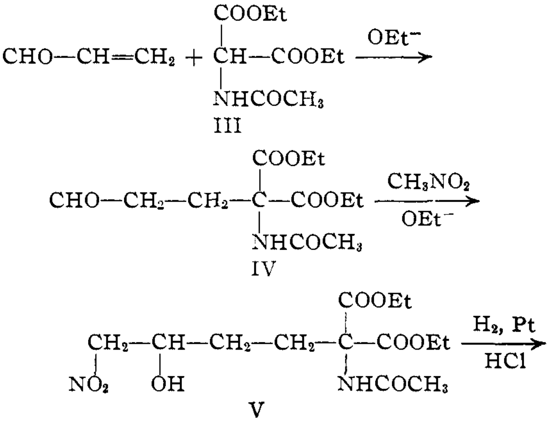
$$\underset{\text{I}}{CH_2NH_2\text{—}\overset{(2)}{CH(OH)}\text{—}CH_2\text{—}CH_2\text{—}\overset{(1)}{CH(NH_2)}\text{—}COOH}$$

and O. H. Mukherjee,[1] and from gelatin by D. D. Van Slyke, A. Hiller, R. T. Dillon and D. A. MacFadyen.[2] In spite of having on hand only inconclusive evidence, the former group was the first to propose a structure for hydroxylysine: α,ε-diamino-β-hydroxycaproic acid. Van Slyke, *et al.*,[2,3] were able to demonstrate, however, that the acid probably possessed either structure (I) or α,δ-diamino-ε-hydroxycaproic acid, since periodic acid released one mole of ammonia and one mole of formaldehyde, indicating that the hydroxyl and one of the amino groups were on adjacent carbon atoms at the end of the chain. More recently, Sheehan and Bolhofer[4] confirmed structure (I) by converting hydroxylysine to methyl α,ε-diphthalimido-δ-keto-DL-caproate (II), which was found to be identical with the material synthesized by an independent route. These coworkers were also successful in effecting the synthesis of structure (I),[5] identifying their product through the preparation of two derivatives, the monohydrochloride and the dipicrate. This paper describes an alternate synthesis of (I)—carried out in this Laboratory prior to the appearance of the publication of Sheehan and Bolhofer—as well as its conversion to derivatives identical with those obtained by these authors.



II

Hydroxylysine monohydrochloride (VII) was obtained in an over-all yield of 26% according to the following



$$CHO\text{—}CH\text{=}CH_2 + \underset{\displaystyle NHCOCH_3}{\overset{\displaystyle COOEt}{CH}}\text{—}COOEt \xrightarrow{OEt^-}$$

III

$$CHO\text{—}CH_2\text{—}CH_2\text{—}\underset{\displaystyle NHCOCH_3}{\overset{\displaystyle COOEt}{C}}\text{—}COOEt \xrightarrow[OEt^-]{CH_3NO_2}$$

IV

$$\underset{\displaystyle NO_2}{CH_2}\text{—}\underset{\displaystyle OH}{CH}\text{—}CH_2\text{—}CH_2\text{—}\underset{\displaystyle NHCOCH_3}{\overset{\displaystyle COOEt}{C}}\text{—}COOEt \xrightarrow[HCl]{H_2,\ Pt}$$

V

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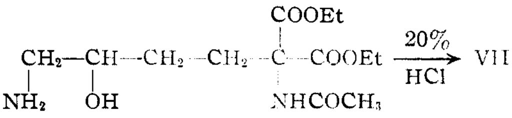
[1] S. B. Schryver, H. W. Buston and O. H. Mukherjee, *Proc. Roy. Soc.* (*London*), **98B**, 58 (1925).

[2] D. D. Van Slyke, A. Hiller, R. T. Dillon and D. A. MacFadyen, *Proc. Soc. Exp. Biol. Med.*, **38**, 548 (1938).

[3] D. D. Van Slyke, A. Hiller, D. A. MacFadyen, A. B. Hastings and F. W. Klemperer, *J. Biol. Chem.*, **133**, 287 (1940); F. W. Klemperer, A. B. Hastings and D. D. Van Slyke, *ibid.*, **143**, 433 (1942).

[4] J. C. Sheehan and W. A. Bolhofer, THIS JOURNAL, **72**, 2469 (1950).

[5] J. C. Sheehan and W. A. Bolhofer, *ibid.*, **72**, 2472 (1950).



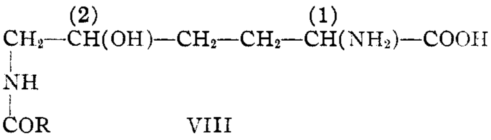
$$\underset{NH_2}{CH_2}-\underset{OH}{CH}-CH_2-CH_2-\underset{NHCOCH_3}{\overset{COOEt}{C}}-COOEt \xrightarrow[HCl]{20\%} VII$$

VI

O. A. Moe and D. T. Warner[6] have previously shown that the addition of diethyl acetamidomalonate (III) to acrolein yields γ-acetamido-γ,γ-dicarbethoxybutyraldehyde (IV), which presumably is the unisolated intermediate in the above synthesis; subsequent addition of nitromethane according to the usual mode of reaction, gave diethyl (γ-hydroxy-δ-nitro-*n*-butyl)-acetamidomalonate (V), m.p. 95.2–96.0°. The infrared spectrum of (V) was consistent with the assigned structure, since absorption bands characteristic of the hydroxyl, nitro, ester and amide groups were present. Catalytic reduction of (V), in ethyl alcohol solution, with platinum oxide in the presence of small amounts of hydrochloric acid afforded the aminoalcohol (VI), which could be obtained only as a glassy solid. (VI) was characterized, however, by means of its acetate, which was readily obtained by catalytic reduction in an ethanol–acetic acid medium. The crude (VI) was sufficiently pure for the preparation of hydroxylysine hydrochloride (VII). After the hydrolysis and decarboxylation of (VI) with 20% hydrochloric acid, the solution was treated successively with silver oxide and hydrogen sulfide; the *p*H of the resulting solution was adjusted to 6.5–7.0 with hydrochloric acid, and the monohydrochloride was isolated and purified by recrystallization (dec. 216–220°). The monohydrochloride has been reported previously by Van Slyke, *et al.*,[2] and by Sheehan and Bolhofer.[7] In agreement with the latter investigators,[5] the dipicrate was found to exist in two characteristic forms, one melting at 145–150° and the other decomposing sharply at 195°.

One interesting detail in the chemistry of hydroxylysine which requires elucidation is its partial racemization during the process of isolation. Both Schryver, *et al.*,[1] and Sheehan and Bolhofer[7] have obtained optically inactive hydroxylysine by means of hydrolysis of protein samples. From their material, Sheehan and Bolhofer were able to isolate two picrates—a monopicrate (m.p. 227°),[8] which showed optical activity, and a dipicrate, which was optically inactive.

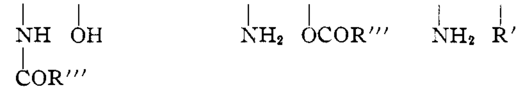
It is quite clear that *both* the asymmetric centers in hydroxylysine are destroyed; the racemization of an asymmetric carbon atom linked to a carbonyl group is of common occurrence (the mechanism being enolization or its equivalent), but a rationalization appropriate for asymmetric center (2), structure (I), is not immediately obvious. However, it may be hypothesized that hydroxylysine is bound *in the protein molecule* by an amide linkage at the ε-position



$$\underset{\underset{COR}{NH}}{CH_2}-\overset{(2)}{CH(OH)}-CH_2-CH_2-\overset{(1)}{CH(NH_2)}-COOH$$

VIII

If this be the case, a suitable path leading to inversion at asymmetric center (2) is realizable. It is well known that the N-acyl derivatives of 1,2-ethanolamines can, in the presence of mineral acid catalysts, undergo rearrangement to the corresponding O-acyl compounds, and it has been demonstrated that this process may take place with partial inversion at C*.[9]



$$-\underset{\underset{COR'''}{NH}}{C}-\underset{OH}{\overset{R'}{C^*}}-R'' \longrightarrow -\underset{NH_2}{C}-\underset{OCOR'''}{\overset{R'}{C^*}}-R'' \quad -\underset{NH_2}{C}-\underset{R'}{\overset{OCOR'''}{C^*}}-R''$$

The partial inversion of (VIII) at carbon atom (2) would then yield a mixture of O-acyl derivatives of diastereoisomeric forms of hydroxylysine; simultaneous or subsequent hydrolytic removal of the acetyl group, and accommodative epimerization at (1) would finally result in at least one of the possible DL-forms of hydroxylysine.

Experimental[10]

Diethyl (γ-Hydroxy-δ-nitro-*n*-butyl)-acetamidomalonate (V).—One hundred and seventy-five grams (0.8 mole) of diethyl acetamidomalonate and 62 cc. of acrolein were condensed in 200 cc. of absolute ethanol containing 0.1 g. of metallic sodium, according to the method of Moe and Warner.[6] The above solution was added slowly with stirring to a solution of 4 g. of sodium hydroxide in 200 cc. of nitromethane, 20 cc. of water and 20 cc. of dioxane. One additional gram of sodium hydroxide was added after stirring had continued for 16 hours. After the solution had been stirred for a total of 40 hours, it was neutralized with 9 cc. of glacial acetic acid and evaporated under reduced pressure (15–25 mm.) to about one-third of its original volume. The addition of benzene caused the separation of yellowish crystals of the desired nitroalcohol. This process of evaporation and addition of benzene was repeated several times, each time separating the precipitated compound by filtration. One hundred and eighty-five grams of the crude product were obtained in this manner. Recrystallization from 1:1 methanol–water yielded 150 g. (56%) of colorless (V), m.p. 95.2–96.0°.

Anal. Calcd. for $C_{13}H_{22}O_8N_2$: C, 46.70; H, 6.63; N, 8.38. Found: C, 46.96; H, 6.55; N, 8.26.

The infrared spectrum exhibited absorption bands at 2.85, 5.77, 5.95 and (6.40, 7.28) μ, characteristic of the hydroxyl ester, amido and nitro groups, respectively.

Diethyl (γ-Hydroxy-δ-amino-*n*-butyl)-acetamidomalonate (VI).—Twenty-two grams (0.066 mole) of (V) in 150 cc. of absolute ethanol containing 4 drops of hydrochloric acid and 0.5 g. of platinum oxide catalyst, were reduced for 16 hours at room temperature in a Parr hydrogenator at an initial pressure of 50 lb. After removal of the catalyst by filtration, the alcohol was removed under reduced pressure with a water aspirator; the last traces of solvent were removed by means of a high-vacuum pump. The resulting white, glassy solid weighed 19.2 g.

The acetate of (VI) was prepared by reducing 11 g. of (V) in 100 cc. of absolute ethanol containing 5 cc. of glacial acetic acid and 0.5 g. of platinum oxide; the conditions were

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(6) O. A. Moe and D. T. Warner, THIS JOURNAL, **70**, 2763 (1948).

(7) J. C. Sheehan and W. A. Bolhofer, *ibid.*, **72**, 2466 (1950).

(8) Reported previously by Van Slyke, *et al.* (ref. 2); by A. P. Martin and R. L. M. Synge, *Biochem. J.*, **35**, 307 (1941); and by J. G. Heathcote, *ibid.*, **42**, 305 (1948).

(9) L. Welsh, THIS JOURNAL, **69**, 128 (1947), has reported that the rearrangement of N-acetyl-(−)-ephedrine leads to a mixture of (−)-ephedrine and its diastereomer, (+)-ψ-ephedrine, and has shown that inversion takes place at the hydroxylic carbon atom. A mechanism for this transformation has been proposed (L. Welsh, *ibid.* **71**, 3500 (1949)).

(10) Melting points are uncorrected.

those described above. After the theoretical amount of hydrogen had been taken up, the solution was filtered through a Jena glass funnel. Upon evaporation of the alcohol under reduced pressure, 9.5 g. of nearly colorless crystals, melting at 121-123°, were obtained. The solid was triturated with 25 cc. of methyl ethyl ketone to yield 7.5 g. of colorless crystals, m.p. 128-129°.

Anal. Calcd. for $C_{15}H_{28}O_8N_2$: C, 49.51; H, 7.74; N, 7.69. Found: C, 49.46; H, 7.72; N, 7.85.

Hydroxylysine Monohydrochloride (VII).—The crude aminoalcohol (VI) (19.2 g.) obtained by reduction of (V) and subsequent removal of solvent, was hydrolyzed by refluxing for 23 hours with 70 cc. of 20% HCl. The resulting brown solution was decolorized with Nuchar and then evaporated to about one-third its original volume in order to remove part of the hydrochloric acid. The solution was then diluted with an equal volume of water, and freshly prepared silver oxide was added to precipitate the chloride ion. After filtration, the excess silver ion was removed with hydrogen sulfide. The *p*H of the solution was then adjusted to 6.5-7.0 with hydrochloric acid and the solution evaporated under reduced pressure at room temperature to about 10 cc. The addition of absolute ethanol effected the precipitation of hydroxylysine monohydrochloride. After recrystallization from methanol-water, 6.2 g. (47%) of VII was obtained. After two or three recrystallizations from methanol-water, the material decomposed at 216-220°. Sheehan and Bolhofer reported the decomposition point of hydroxylysine hydrochloride as 215-220°.[5]

Anal. Calcd. for $C_6H_{15}O_3N_2Cl$: C, 36.27; H, 7.61; N, 14.10; Cl, 17.85. Found: C, 35.93; H, 7.63; N, 14.16; Cl, 17.88.

The dipicrate of (VII) was prepared by the addition of an equivalent amount of picric acid to the solution of (VII) obtained after the adjustment of *p*H as described above. In agreement with the results of Sheehan and Bolhofer,[5] the dipicrate was found to exist in two characteristic forms, one melting at 145-150°, and the other decomposing sharply at 195°.

Anal. Calcd. for $C_{18}H_{20}O_{17}N_8$: C, 34.85; H, 3.25; N, 18.06. Found (m.p. 145-150°): C, 34.50; H, 3.47; N, 17.81. Found (dec. 195°): C, 34.54; H, 3.56; N, 18.08.

A monopicrate was also obtained by the addition of very small amounts of picric acid to a solution of hydroxylysine hydrochloride. The deep yellow solid melted with decomposition at 227°.

Anal. Calcd. for $C_{12}H_{17}O_{10}N_5$: C, 36.83; H, 4.38; N, 17.90. Found: C, 37.43; H, 4.49; N, 18.03.

Acknowledgment.—The authors wish to thank the Research Corporation for a grant-in-aid supporting this project.

HOLLAND, MICHIGAN RECEIVED AUGUST 21, 1950

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF "ATANOR" CÍA. NACIONAL PARA LA INDUSTRIA QUÍMICA S.A.M.]

Cactus Alkaloids. I. *Trichocereus terscheckii* (Parmentier) Britton and Rose

BY L. RETI AND J. A. CASTRILLÓN

The dried branches of *Trichocereus terscheckii* (Parmentier) Britton & Rose contain 0.25 to 1.2% alkaloids. From this source trichocereine, a new vegetable base, and the already known mezcaline have been isolated and characterized. The ratio of trichocereine to mezcaline is 5:1. The chemical structure of trichocereine has been shown to be N-dimethylmezcaline by degradation, and its synthesis has been achieved by allowing 3,4,5-trimethoxy-β-phenylethyl chloride to react with dimethylamine.

The genus *Trichocereus* of the cactus family consists of 19 recognized species, confined to South America (N. L. Britton and J. N. Rose, "The Cactaceae." The Carnegie Institution of Washington, Washington, 1919-1923). So far only seven species have been chemically examined and all contain basic substances; the genus must, therefore be, considered as alkaloidiferous. *T. Candicans*[1] and *T. lamprochlorus*[2] contain candicine and hordenine (anhaline); *T. spachianus*,[3] candicine; *T. thelegonoides*, *T. thelegonus* and *T. huascha*,[4] alkaloids of still undetermined structure. A preliminary report on the bases of *T. terscheckii* was presented at the X International Congress of Chemistry in Rome 1938.[5] An exhaustive survey on Cactus alkaloids and some related compounds has been published recently.[6]

Trichocereus terscheckii grows in the high valleys of northwestern Argentia (provinces of La Rioja, Catamarca, Tucumán, Salta, etc.) and often forms the most conspicuous plant in the landscape. It is a very large cactus, 10 to 12 meters high, resembling the American saguaro (*Carnegiea gigantea*).

Various lots of plants have been examined, collected at blossom time (October-December) near Chilecito (La Rioja) and Andalgalá (Catamarca). The content of alkaloids varied between 0.25 and 1.2%, referred to the dry plant. Preliminary tests showed that no appreciable amount of phenolic bases could be expected and that two types of alkaloids were present: (a) soluble in ether, (b) insoluble in ether but soluble in chloroform. Two basic compounds have been isolated and characterized: mezcaline (chloroform soluble) and a new vegetable alkaloid which should be named trichocereine (ether soluble). Apparently there are also other bases in small amounts. In some lots with high alkaloidal content, no mezcaline could be detected. The ratio trichlocereine to mezcaline is 5:1. This is the first case where mezcaline, the active hallucinatory principle of the "mescal-buttons" (*Anhaloniumlewinii syn. Lophophora williamsii*) has been found in a different species.

Mezcaline has been identified as the main alkaloid of the ether-insoluble fraction, by comparing several derivatives of the base found in *T. terscheckii* with the synthetic substance. Trichocereine is a new natural phenylethylamine, found so far only in this species. The structure has been determined by analytical and synthetical methods. Trichocereine, $C_{13}H_{21}O_3N$, contains three methoxyl and two N-methyl groups. Trimethylgallic acid is formed in good yields on oxida-

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[1] L. Reti, *Rev. Soc. Argentina Biol.*, **9**, 344 (1933).

[2] L. Reti and R. I. Arnolt, *Actas y Trabajos del V° Congreso Nacional de Medicina, Roasario*, **3**, 39 (1935).

[3] S. A. Haagen-Smit and M. Olivier, private communication.

[4] S. A. Haagen-Smit and M. Olivier, private communication.

[5] L. Reti, *Atti Congr. Intern Chim 10th Congr, Rome*, 1938, **5**, 396 (1939).

[6] L. Reti, "Fortschritte der Chemie organischer Naturstoffe," Vol. VI, L. Zechmeister, Ed., Springer Verlag, Wien, 1950, p. 242.