hydrothiophenecarboxylate,[2] 189 g. of hydroxylamine hydrochloride, 296 g. of anhydrous barium carbonate and 1500 ml. of absolute alcohol was refluxed on the steam-bath for twenty-two hours while a drying tube excluded moisture. The oily oxime was isolated by the same procedure as described for oxime I; yield, 175 g. (88%).

Anal. Calcd. for $C_{17}H_{23}O_4NS$: N, 4.16. Found: N, 4.38.

Ethyl 3-Amino-2-γ-benzyloxypropyl-4-thiophenecarboxylate Hydrochloride (IV).—A solution of 67.5 g. (0.20 mole) of the aforementioned oxime in 500 ml. of anhydrous ether was protected by a calcium chloride tube and saturated with dry hydrogen chloride over a period of thirty-five minutes. A brown color soon developed. After the mixture had stirred for twenty hours at room temperature the tan crystals were filtered from the dark-brown solution and washed thoroughly with dry ether; yield, 55.4 g. (78%); m. p. 121–125°. Recrystallization of a sample from methyl isobutyl ketone–ether did not improve the melting point.

Anal. Calcd. for $C_{17}H_{21}O_3NS \cdot HCl$: N, 3.93. Found: N, 4.10.

In cases where the amine hydrochloride failed to crystallize from solution, the ether and the excess hydrogen chloride were removed in a current of air. Dry ether was then stirred into the dark-brown, oily residue while cooling the mixture to induce crystallization.

Acknowledgment.—The authors wish to thank Mr. A. W. Spang and Mrs. Margaret Ledyard for the microanalyses.

Summary

Ethyl 3-keto-2-γ-phenoxypropyl-4-tetrahydrothiophenecarboxylate oxime undergoes aromatization with loss of water to yield ethyl 3-amino-2-γ-phenoxypropyl-4-thiophenecarboxylate hydrochloride when treated with hydrogen chloride in anhydrous ether. In like manner ethyl 2-γ-benzyloxypropyl - 3 - keto - 4 - tetrahydrothiophenecarboxylate oxime is converted into ethyl 3 - amino -2 - γ - benzyloxypropyl - 4 - thiophenecarboxylate hydrochloride.

Because of the favorable yields obtained, this aromatization provides a new method for the synthesis of hitherto inaccessible derivatives of 3-aminothiophene.

This transformation has furnished a structural proof and a key intermediate required for the synthesis of 2,3,4,5-tetradehydrobiotin.

DETROIT, MICHIGAN RECEIVED JANUARY 8, 1945

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF PARKE, DAVIS & COMPANY]

The Total Synthesis of 2,3,4,5-Tetradehydrobiotin[1]

BY LEE C. CHENEY AND J. ROBERT PIENING

The observation that the oximes of certain tetrahydrothiophene β-keto esters could be transformed with surprising ease into the corresponding 3-aminothiophenes[2] suggested the possibility of obtaining biotin through its aromatic analog. Such an approach to the synthesis of biotin offered certain advantages. It would not only eliminate steps involving troublesome stereoisomers, but, since the amino groups in the 3,4-diaminothiophenevaleric acid would necessarily be in a position facilitating ring closure with phosgene, it would also provide a sure means of forming the ureylene bridge. Whether or not the sulfur-containing bicyclic system could be reduced to biotin was considered an open question. The fact that the physiological activity of "aromatic biotin" would be of pharmacological interest added significance to the venture. A preliminary communication[3] has outlined the steps by which the synthesis of 2,3,4,5-tetradehydrobiotin (XVIII) was accomplished.

Either α-bromo- or α-chloropimelic acid was required as an intermediate for the preparation of the key β-keto ester (IX). A search of the literature revealed that no mono α-halopimelic acids had been described. Although α-bromosuberic acid has been prepared by the Hell–Volhard–Zelinsky method, considerable dibromo acid is formed in the reaction.[4] The direct monohalogenation of adipic acid has led to mixtures which are separated only with difficulty.[5] Consequently, v. Braun and Meyer[6] have devised a six-step process for the synthesis of pure α-bromoadipic acid.

The route to α-chloropimelic acid (VI) involved the shown sequence of reactions. Since the completion of this work, Karrer, Keller and Usteri[7] have reported the preparation of α-bromopimelic acid by the bromination of the identical malonic acid (IV) followed by decarboxylation.

β-Mercaptopropionic acid condensed readily with VI in alkaline solution to produce VII. The purification of VII was not attempted, since esterification afforded pure VIII in yields of 73–77% based on VI.

The Dieckmann cyclization of VIII, which could lead to the formation of either a cyclohexanone or a tetrahydrothiophene derivative, proceeded smoothly at room temperature in the presence of two moles of sodium ethoxide suspended in benzene to produce the β-keto ester (IX) in yields of 84–89%. It gave an intense red coloration with ferric chloride in alcoholic solution

(1) Presented in part before the Division of Organic Chemistry, 108th meeting of the American Chemical Society, New York, N. Y., September 12, 1944.

(2) Cheney and Piening, THIS JOURNAL, 67, 729 (1945).

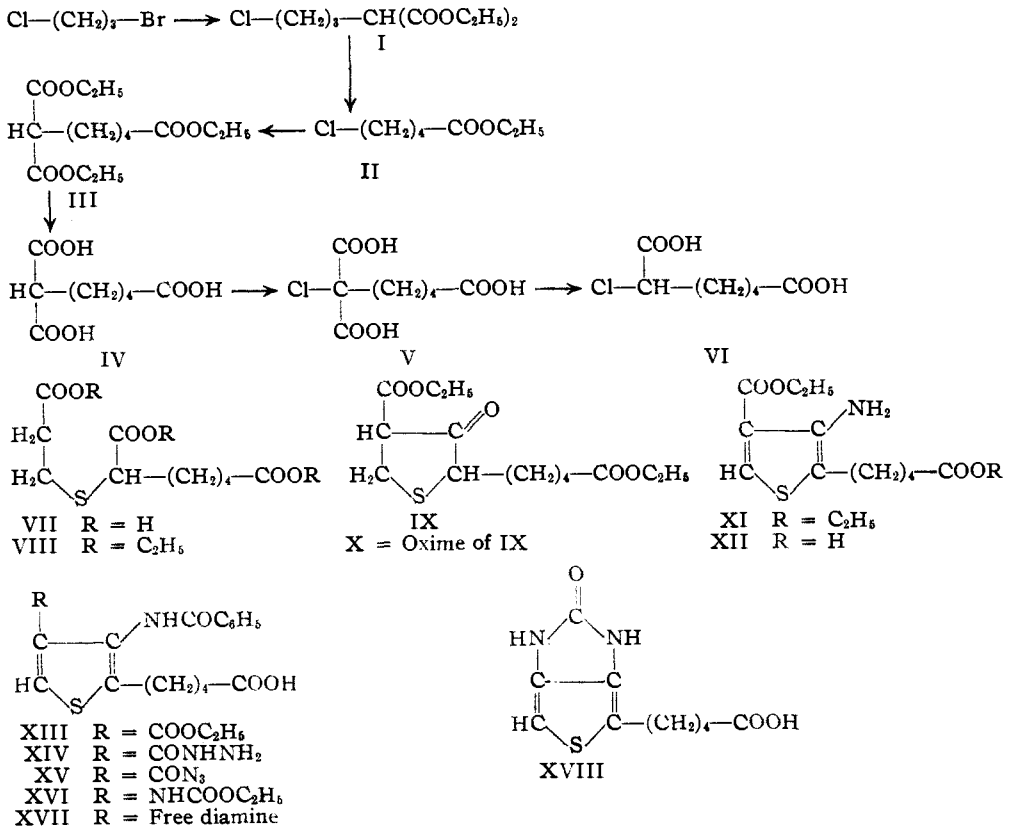
(3) Cheney and Piening, *ibid.*, 66, 1040 (1944).

(4) Gantter and Hell, *Ber.*, 15, 142 (1882); Hell and Rempel, *ibid.*, 18, 812 (1885).

(5) Gal and Gay-Lussac, *Ann.*, 155, 250 (1870); Ince, *J. Chem. Soc.*, 67, 159 (1895); Ingold, *ibid.*, 119, 961 (1921).

(6) v. Braun and Meyer, *Ber.*, 74, 19 (1941).

(7) Karrer, Keller and Usteri, *Helv. Chim. Acta*, 27, 237 (1944).



and was purified effectively through its light-green copper chelate compound. The structure of IX was established by the aromatization of its oxime (X) to the corresponding 3-aminothiophene (XI) by means of hydrogen chloride in dry ether.[2] Owing to appreciable acid hydrolysis, the product actually proved to be a mixture of the hydrochlorides of XI and XII, which was easily separated by treatment with ammonium hydroxide or sodium bicarbonate solution and extraction with ether. By virtue of the relative ease of hydrolyzing the ester of the normal side-chain in comparison with the *ortho*-substituted aromatic ester group, the diester (XI) was transformed into the acid ester (XII) by selective saponification with alcoholic sodium or potassium hydroxide solution in yields of 80–85%.

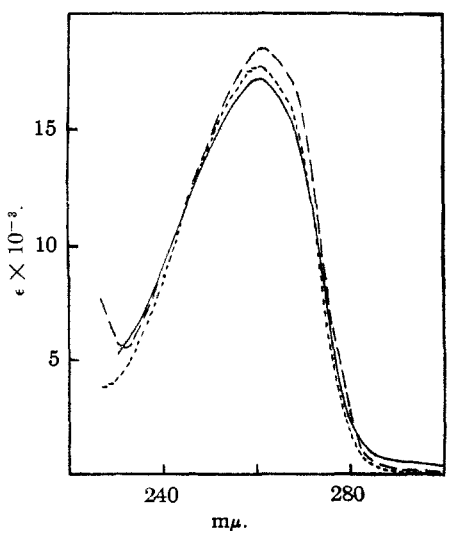


Fig. 1.—2′-Keto-3,4-imidazolido-2-thiophenevaleric acid ——; 2′-keto-3,4-imidazolido-2-γ-phenoxypropylthiophene — —; 2′-keto-3,4-imidazolido-2-γ-benzyloxypropylthiophene - - -.

The route from XII to XVIII through the Curtius degradation involved the sequential preparation of the benzoyl derivative (XIII), the hydrazide (XIV), the azide (XV) and the urethan (XVI), all obtained in yields of 83–95%. Because the susceptibility of the diamine (XVII) to atmospheric oxidation rendered its isolation impractical, the alkaline hydrolysis of XVI was conducted under nitrogen. Treatment of a protected solution of XVII with phosgene produced 2,3,4,5-tetradehydrobiotin in yields of 50–60% based on the urethan.

Confirmation of the assigned structure was afforded by the ultraviolet absorption determinations which showed that 2,3,4,5-tetradehydrobiotin has an absorption spectrum closely corresponding to the spectra of 2′-keto-3,4-imidazolido-2-γ-phenoxypropylthiophene and 2′-keto-3,4-imidazolido-2-γ-benzyloxypropylthiophene. The latter compounds have been synthesized by methods

analogous to the one described, but since in the Dieckmann cyclizations involving the ethers there is no possibility for the formation of a six-membered ring, the structures of these compounds can be assigned with certainty.

2,3,4,5-Tetradehydrobiotin has been tested for biotin and anti-biotin activity on two microorganisms. It did not replace biotin for either *L. arabinosus* or *S. cerevisiae* to a concentration of 400 γ per 10 ml. whereas biotin itself is active at a range of 0.1 to 1.0 m. γ per 10 ml. of medium. The same concentration did not inhibit the activity of a minimal amount of biotin for these two organisms. The effect of "aromatic biotin" on the biotin-deficient rat is under investigation.

Experimental[8]

Ethyl δ-Chlorovalerate (II).[9]—The following procedure is based on the method of Mellor, who omitted experimental details. A mixture of 450 g. (1.9 moles) of ethyl 3-chloropropylmalonate (I),[10] b. p. 146.5–149° at 10 mm. ($n^{20}D$ 1.4429), 1 liter of concentrated hydrochloric acid and 100 ml. of alcohol was refluxed in a wax-bath for twenty hours. The cooled mixture was diluted with two liters of water, the aqueous phase was extracted with 2.5 liters of ether in five portions, and combined oil and extracts were dried over sodium sulfate. Distillation of the solvent left 231.3 g. of crude δ-chlorovaleric acid which was dissolved in 500 ml. of absolute alcohol, cooled in an ice–salt-bath, and saturated with dry hydrogen chloride. After fifteen hours had elapsed 450 ml. of alcohol was distilled at reduced pressure, and the residual oil was poured on ice and made basic to litmus with sodium bicarbonate solution. The cold solution (*ca.* 1 liter) was extracted with 1200 ml. of ether in five portions. Distillation of the dried extracts from a modified Claisen flask (Vigreux column) gave 180.8 g. (a 57.7% yield) of colorless oil, b. p. 83.5–85° at 8 mm.; $n^{20}D$ 1.4355.

Ethyl 1,1,5-Pentanetricarboxylate (III).[7,11]—To a cooled solution of 60.5 g. (2.63 g. atoms) of sodium in 1200 ml. of absolute alcohol was added 505 g. (3.15 moles) of ethyl malonate. The stirred solution was refluxed while 433 g. (2.63 moles) of ethyl δ-chlorovalerate was added dropwise. After the suspension had been refluxed and stirred for twenty-one hours, the alcohol was distilled. The residue was cooled, diluted with 700 ml. of water, made acid to congo red, and extracted with 2 liters of ether. The extracts were washed with sodium bicarbonate solution and water and then dried over sodium sulfate. The yield of product distilling at 165–170° at 4 mm. was 551.6 g. (72.5%).

Anal. Calcd. for $C_{14}H_{24}O_6$: C, 58.32; H, 8.39. Found: C, 58.49; H, 8.54.

1,1,5-Pentanetricarboxylic Acid (IV).[7]—A solution of 560 g. of potassium hydroxide in 895 ml. of water was cooled to 10° in an ice-bath while 545 g. (1.89 moles) of ethyl 1,1,5-pentanetricarboxylate was added dropwise. The mixture was first stirred overnight at room temperature and then for twelve hours on the steam-bath. The basic solution was concentrated under reduced pressure, cooled in an ice-bath and made acid to congo red with concentrated hydrochloric acid. The acid solution was extracted seventeen times with ether. The extracts were combined, washed with saturated salt solution and dried over sodium sulfate. Removal of the ether left an oil which crystallized when dried *in vacuo;* yield, 362 g. (94%). A sample was recrystallized from ether–petroleum ether (b. p. 35–60°); m. p. 89–90°.

Anal. Calcd. for $C_8H_{12}O_6$: C, 47.06; H, 5.92. Found: C, 47.11; H, 5.88.

1-Chloro-1,1,5-pentanetricarboxylic Acid (V).—A solution of 40.6 g. (0.199 mole) of 1,1,5-pentanetricarboxylic acid in 150 ml. of dry ether was protected from moisture while 16.2 ml. (0.20 mole) of redistilled sulfuryl chloride was added over a period of one hour. The resulting solution was refluxed for two hours. Ether was removed by distillation and finally by evaporation. The oily residue was stirred until crystals appeared. Petroleum ether (b. p. 35–60°) was then stirred into the product until crystallization was completed. The yield of dry product was 47.2 g. (99%), m. p. 126° (dec.). Recrystallization of a sample from ether–petroleum ether gave white crystals melting at 127° (dec.).

Anal. Calcd. for $C_8H_{11}O_6Cl$: C, 40.26; H, 4.64. Found: C, 40.44; H, 5.00.

α-Chloropimelic Acid (VI).—Exactly 45.9 g. (0.192 mole) of 1-chloro-1,1,5-pentanetricarboxylic acid was heated in an oil-bath to 130°. Carbon dioxide was evolved until the temperature had reached 150° over a period of twenty minutes. When the evolution of gas had ceased the flask was removed from the bath and allowed to cool. The acid solidified to a hard mass. The yield was quantitative (37 g.). After being recrystallized twice from water, the white product melted at 89–90°.

Anal. Calcd. for $C_7H_{11}O_4Cl$: C, 43.20; H, 5.70; neut. equiv., 97. Found: C, 43.27; H, 5.78; neut. equiv., 98.

β-Mercaptopropionic Acid.[12]—A mixture of 383 g. (2.5 moles) of β-bromopropionic acid (Dow), 190 g. (2.5 moles) of thiourea and 1250 ml. of denatured alcohol was refluxed on the steam-bath for three hours. A solution of 250 g. (6.25 moles) of sodium hydroxide in 1500 ml. of water was added and the resulting solution was refluxed under nitrogen for one hour. After the addition of 150 g. (0.63 mole) of sodium sulfide nonahydrate, solvent was distilled under reduced pressure until crystals appeared. The mixture was cooled in an ice–salt-bath, covered with 1 liter of ether, and a cold solution of 175 ml. of concentrated sulfuric acid in 1250 ml. of water was added slowly while swirling the flask. The ether layer was separated and the aqueous phase, which was acid to congo red, was extracted with 2 liters of ether in four portions. Combined extracts were washed with a saturated solution of sodium chloride and dried over sodium sulfate. Subsequent to the removal of solvent through a Vigreux column, the residual yellow liquid was distilled under nitrogen to obtain 192.3 g. (72.3% yield) of colorless β-mercaptopropionic acid, b. p. 105–107° at 4 mm.; $n^{20}D$ 1.4910. The yellow residue was dissolved in a minimum of boiling water to obtain 31.1 g. of crude β-dithiopropionic acid.[13]

2-Carbethoxyethyl 1,5-Dicarbethoxyamyl Sulfide (VIII).[7]—A solution of 28.8 g. (0.70 mole) of sodium hydroxide in 100 ml. of water was stirred while 17.5 g. (0.165 mole) of β-mercaptopropionic acid was added. A solution of 32.2 g. (0.165 mole) of α-chloropimelic acid in 50 ml. of warm water was then added dropwise and the resulting solution was refluxed for two hours. The cooled yellow solution was acidified to congo red and the water was removed under reduced pressure. The residue was extracted with chloroform and the chloroform was distilled. The thick oily residue (VII) was dried and then dissolved in 600 ml. of absolute alcohol containing 12 ml. of concentrated sulfuric acid, and the solution was refluxed for five hours. Alcohol was distilled rapidly until about 100 ml. remained. The residue was poured on cracked ice and extracted thrice with ether. Extracts were washed with sodium bicarbonate solution and dried over sodium

(8) All melting points are corrected.

(9) Funk, *Ber.*, **26**, 2574 (1893); Mellor, *J. Chem. Soc.*, **79**, 132 (1901); Conant and Kirner, THIS JOURNAL, **46**, 244 (1924); Prelog and Heimbach-Juhasz, *Ber.*, **74**, 1702 (1941).

(10) Fischer and Bergmann, *Ann.*, **398**, 120 (1913).

(11) The compound was originally prepared in this Laboratory by Dr. Elmer J. Lawson.

(12) Other methods of preparation include that of Bülmann, *Ann.*, **348**, 125 (1906), who cites prior work; Holmberg and Schjanberg, *Arkiv Kemi, Mineral. Geol.*, **14A**, No. 7 (1940); [*C. A.*, **35**, 2114 (1941)].

(13) Bülmann, *Ann.*, **339**, 365 (1905).

sulfate. The fraction distilling at 210–213° at 3 mm. weighed 42.2 g. (73.5% yield).

Anal. Calcd. for $C_{16}H_{26}O_6S$: C, 55.15; H, 8.10. Found: C, 55.14; H, 8.15.

Because the isolation of the free β-mercaptopropionic acid was accompanied by considerable loss owing to oxidation to the disulfide, a continuous process was developed wherein the disodium salt of β-mercaptopropionic acid obtained by the hydrolysis of the S-β-carboxyethylisothiuronium bromide was condensed directly with α-chloropimelic acid.

Simplified Procedure.—A mixture of 88.0 g. (0.58 mole) of β-bromopropionic acid, 43.7 g. (0.58 mole) of thiourea, and 385 ml. of denatured alcohol was refluxed on the steam-bath for eighteen hours. After most of the alcohol was removed by distillation, the residue was added to a solution of 83 g. (2.01 moles) of sodium hydroxide in 500 ml. of water and the mixture was refluxed for 2.5 hours. A solution of 37 g. (0.89 mole) of sodium hydroxide in 200 ml. of water was then added. To the resulting stirred solution was added dropwise 97.3 g. (0.50 mole) of α-chloropimelic acid dissolved in 150 ml. of warm water. When the addition was completed, the solution was refluxed for two hours, cooled, and made acid to congo red with hydrochloric acid. The product was extracted with 3 liters of ether in five portions, the combined extracts were dried and the ether was distilled. The thick oil was refluxed for five hours with 1500 ml. of absolute alcohol and 36 ml. of concentrated sulfuric acid. After approximately 1250 ml. of the alcohol had been rapidly distilled, the residue was poured on cracked ice, diluted with cold water and extracted with 2 liters of ether in four portions. The washed and dried extracts were distilled to obtain 133.9 g. of colorless ester, b. p. 204–205° at 2 mm., which represents a yield of 77% based on the α-chloropimelic acid.

Ethyl 4-Carbethoxy-3-keto-2-tetrahydrothiophenevalerate (IX).[7]—The alcohol was removed under reduced pressure from a solution of 5.13 g. (0.223 g. atom) of sodium in 80 ml. of absolute alcohol by heating the mixture under dry nitrogen in a wax-bath at 180–190° for one hour after all visible alcohol had distilled. The dry sodium ethoxide was kept under nitrogen while lumps were broken up and then covered with 200 ml. of dry benzene. A solution of 38.8 g. (0.11 mole) of 2-carbethoxyethyl 1,5-dicarbethoxyamyl sulfide in 50 ml. of dry benzene was added in two portions. The mixture was shaken and cooled whenever it became warm. The sodium ethoxide went into solution very readily, but after several minutes the sodium salt of the keto ester separated. After standing for eighteen hours at room temperature, the cooled mixture was acidified with a solution of 20 ml. of glacial acetic acid in 160 ml. of water. The benzene layer was washed with sodium bicarbonate solution and dried over sodium sulfate. Removal of benzene left a light-brown oil which gave a deep-red color with ferric chloride in alcoholic solution. For purification the oil was added in small portions with vigorous shaking to an excess of saturated cupric acetate solution. The green chelate compound which precipitated was dissolved in ether. The ether solution was washed thoroughly with water, dried and concentrated to a small volume. Cooling and scratching started the crystallization, which was completed by the gradual addition of petroleum ether; weight, 33.3 g. Recrystallization of a sample from ether–petroleum ether yielded fine, light-green crystals, m. p. 118.5–119.5°.

Anal. Calcd. for $(C_{14}H_{22}O_6S)_2Cu$: C, 50.32; H, 6.63; Cu, 9.51. Found: C, 50.76; H, 6.56; Cu, 9.70.

The keto ester was regenerated by shaking an ether solution of the chelate compound with 10% sulfuric acid. The ether solution was washed with a solution of sodium bicarbonate and dried over sodium sulfate. Removal of ether left 30.0 g. of dark-red oil which lightened to a yellow color on long standing; yield, 89%.

Anal. Calcd. for $C_{14}H_{22}O_6S$: C, 55.61; H, 7.33. Found: C, 55.53; H, 7.36.

Ethyl 4-Carbethoxy-3-keto-2-tetrahydrothiophenevalerate Oxime (X).—A solution of 30.0 g. (0.099 mole) of IX in 300 ml. of absolute alcohol was refluxed for fifteen hours on the steam-bath with 34.8 g. (0.5 mole) of hydroxylamine hydrochloride and 59.2 g. (0.3 mole) of anhydrous barium carbonate while a calcium chloride tube excluded moisture. The mixture was filtered and the inorganic salts were washed thoroughly with hot absolute alcohol. The alcohol was removed under reduced pressure. The residue was taken up in ether, washed with water and dried over sodium sulfate. Ether and salt were removed and the residue dried *in vacuo*. The yellow oil, which gave a negative ferric chloride test, weighed 29.87 g.; yield, 96%.

Anal. Calcd. for $C_{14}H_{23}O_5NS$: C, 52.97; H, 7.30. Found: C, 52.77; H, 7.52.

Ethyl 3-Amino-4-carbethoxy-2-thiophenevalerate (XI) and 3-Amino-4-carbethoxy-2-thiophenevaleric Acid (XII).—In a 500-ml. three-necked flask fitted with a condenser, stirrer and inlet tube was placed a solution of 28.96 g. (0.091 mole) of X in 250 ml. of anhydrous ether. Dry hydrogen chloride was introduced through the inlet tube for thirty minutes while stirring the solution. The solution became darker in color and after several hours crystals formed on the sides of the flask. After the mixture had stirred for twenty-one hours, the crystals were collected, washed with dry ether and dried. The crude hydrochlorides weighed 25.15 g. and melted at 101–132°. The mixture was treated with an excess of 5% sodium bicarbonate solution. Ether extraction removed the amine diester (XI), which was obtained by evaporating the ether and then recrystallizing from alcohol after treatment with Darco. The pale-yellow needles, m. p. 43–44°, weighed 16.45 g. (yield, 60.5%).

Anal. Calcd. for $C_{14}H_{21}O_4NS$: C, 56.16; H, 7.07. Found: C, 56.19; H, 7.04.

The acid ester (XII) was obtained by acidification of the basic solution with dilute acetic acid. The product was collected and dried; yield, 3.56 g. (14.5%); m. p. 94°. Recrystallization of a sample from water (Darco) gave colorless fine needles, m. p. 97–97.5°.

Anal. Calcd. for $C_{12}H_{17}O_4NS$: C, 53.12; H, 6.32. Found: C, 53.14; H, 6.32.

Compound XII was also obtained readily by preferential saponification of the diester. A solution of 11.0 g. (0.037 mole) of XI in 35 ml. of alcohol was treated with 35.2 ml. of 1.045 molar alcoholic potassium hydroxide. After nineteen hours had elapsed the alcohol was removed in a jet of air on the steam-bath. The residue was dissolved in water and extracted with ether, and the basic solution was acidified with acetic acid to obtain 5.06 g. of the acid ester, m. p. 92–94°. Evaporation of the ether extract provided 4.2 g. of starting material. The yield of acid ester based on the unrecovered diester was, therefore, 82%.

3-Benzoylamino-4-carbethoxy-2-thiophenevaleric Acid (XIII).—To a stirred solution of 23.5 g. (0.087 mole) of XII and 11.6 g. (0.096 mole) of dimethylaniline in 300 ml. of dry chloroform (a calcium chloride tube excluded moisture) was added dropwise a solution of 13.4 g. (0.096 mole) of benzoyl chloride in 100 ml. of chloroform. After addition was completed (1.2 hours) the mixture was stirred for three hours, then washed twice with 5% hydrochloric acid solution followed by thorough extraction with 5% sodium bicarbonate solution. The combined alkaline extracts were washed with ether, cooled in an ice-bath and rendered acid to congo red with dilute hydrochloric acid. The oily product was taken up in 1900 ml. of ether. The ether solution was washed with water, dried over sodium sulfate and concentrated to a volume of 350 ml. Cooling and diluting with petroleum ether (b. p. 35–60°) yielded 27.1 g. (83%) of an ivory colored product, m. p. 126.5–127.5°. Recrystallization of a sample from ether–petroleum ether (Darco) produced colorless micro-needles, m. p. 127.5–128°.

Anal. Calcd. for $C_{19}H_{21}O_4NS$: C, 60.80; H, 5.63. Found: C, 60.70; H, 5.53.

3-Benzoylamino-2-δ-carboxybutyl-4-thiophenecarboxylic Acid Hydrazide (XIV).—To a solution of 9.28 g.

(0.025 mole) of XIII in 40 ml. of absolute alcohol and 117 ml. of dry benzene were added 76.5 ml. of 0.324 molar absolute alcoholic potassium ethoxide solution and 9.9 ml. of 100% hydrazine hydrate. The flask was then fitted with a Soxhlet extractor containing a thimble nearly filled with 35 g. of Drierite, and the reflux condenser was capped by a calcium chloride tube. After the solution had been refluxed vigorously for sixteen hours on the steam-bath, solvent and excess hydrazine were distilled below 55° under reduced pressure. The residue was dissolved in 75 ml. of warm water, cooled in an ice-bath, covered with 200 ml. of ether, treated with 4 ml. of 6 *N* hydrochloric acid and then acidified with 10% acetic acid solution until no more precipitate formed. Cooling and stirring soon caused the oil to crystallize. The white product was collected by suction, washed with ether followed by ice water and desiccated to yield 8.12 g. (91%) of the hydrazide, m. p. 140–141°. Recrystallization of a sample from water (Darco) failed to alter the m. p. of the fine needles.

Anal. Calcd. for $C_{17}H_{19}O_4N_5S$: C, 56.49; H, 5.30. Found: C, 56.40; H, 5.57.

Owing to partial hydrolysis of the alkali salt caused by the water liberated when the hydrazine hydrate reacted with the ester, the yields of the hydrazide were below 55% before the expedient of introducing the desiccant was adopted for the vapor-phase removal of the water.

3-Benzoylamino-2-δ-carboxybutyl-4-thiophenecarboxylic Acid Azide (XV).—After 10.68 g. (0.029 mole) of XIV had been dissolved in 85 ml. of glacial acetic acid by the application of heat, the well-stirred solution was cooled in an ice-bath and treated with 3.42 ml. of concentrated hydrochloric acid. To the suspension of the hydrochloride thus formed was added dropwise during 0.8 hour a cold solution of 2.23 g. (0.032 mole) of sodium nitrite in 55 ml. of water. Throughout the reaction the internal temperature was maintained at 0°. When addition was completed the mixture was stirred for a further 0.7 hour and then 250 ml. of ice water was added via dropping funnel during 0.5 hour. The cold, white solid was collected on a filter, washed thoroughly with ice water and desiccated *in vacuo*. The chalk-like azide, which decomposed at 99–100°, weighed 10.2 g. (93% yield). A small sample exploded when heated over a flame. It slowly developed a pink color on standing.

Anal. Calcd. for $C_{17}H_{15}O_4N_4S$: C, 54.83; H, 4.33. Found: C, 55.11; H, 4.53.

3-Benzoylamino-4-carbethoxyamino-2-thiophenevaleric Acid (XVI).—A solution of 10.2 g. (0.027 mole) of XV in 750 ml. of absolute alcohol was protected by a calcium chloride tube and heated gradually to boiling during 1.7 hours. This yellow solution was then refluxed on the steam-bath for seventeen hours. Alcohol was distilled until crystals had begun to form. Dilution with hot water followed by stirring and cooling caused the product to crystallize as a very pale-pink solid. After desiccation the urethan, m. p. 155.5–157°, weighed 10.2 g. (95% yield). Recrystallization of a sample from dilute alcohol (Darco) produced colorless micro-needles, m. p. 158–159°. Recrystallization did not raise the melting point.

Anal. Calcd. for $C_{19}H_{22}O_5N_2S$: C, 58.44; H, 5.68; N, 7.18. Found: C, 58.80; H, 5.92; N, 7.07.

2'-Keto-3,4-imidazolido-2-thiophenevaleric Acid (2,3,-4,5-Tetradehydrobiotin) (XVIII).—A mixture of 1.92 g. (0.0049 mole) of XVI, 24 g. of C. P. potassium hydroxide and 84 ml. of reagent methanol was refluxed on the steam-bath for twelve hours in purified nitrogen. The heating was continued for another hour while steam was passed through the jacket of the condenser for the complete removal of the methanol under reduced pressure. The flask, with the white residue, was then cooled in an ice-salt-bath and 175 ml. of freshly boiled water cooled to 5° was added through the condenser. The atmosphere of nitrogen was maintained throughout this and the subsequent operation. Phosgene was bubbled through the solution with shaking for 0.9 hour, until the brown solution had become colorless and acid to congo red. The resulting gummy precipitate was dissolved in 5% potassium hydroxide solution, boiled with Darco and reprecipitated. The light-brown solid was dried, pulverized and extracted twice with ether; yield, 0.682 g. (58%); m. p. 248–250°. Recrystallizations, with decolorization, first from dilute alcohol and then from a large volume of water, produced colorless needles, m. p. 254–255° (dec.).

Anal. Calcd. for $C_{10}H_{12}O_3N_2S$: C, 49.98; H, 5.03. Found: C, 50.33; H, 5.25.

"Aromatic biotin" is practically insoluble in water, ether, chloroform, benzene, ethyl acetate, acetone and ligroin; it is soluble in alcohol, methanol, acetic acid and sodium bicarbonate solution. Although stable toward dilute alkali, the compound is transformed into an amorphous red-brown powder by strong mineral acids.

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Summary

The total synthesis of 2,3,4,5-tetradehydrobiotin is described.

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[CONTRIBUTION FROM THE SCHOOL OF CHEMISTRY AND PHYSICS OF THE PENNSYLVANIA STATE COLLEGE]

Heterocyclic Basic Compounds. IV. 2-Aminoalkylamino-pyrimidines[1]

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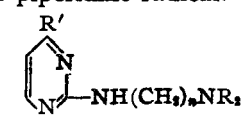
In previous publications[4] a number of basically-substituted pyridine, triazine and quinoline derivatives was described and as an extension of this work it seemed desirable to prepare some of the corresponding pyrimidine compounds. The compounds studied in this paper may be represented by the following general formula, where R is an alkyl group or is fused in an heterocyclic nucleus and R′ is an hydrogen atom or a methoxy, morpholino or piperidino radical.



These compounds were prepared by the alkylation

(1) Presented before the Division of Organic Chemistry of The American Chemical Society, Pittsburgh, Pennsylvania, September 6, 1943.

(2) This paper is taken in part from the doctoral dissertation of Robert R. Adams, The Pennsylvania State College, February, 1944.

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(4) (a) Whitmore, Mosher, Goldsmith and Rytina, THIS JOURNAL, 67, 393 (1945); (b) Mosher and Whitmore, *ibid.*, 67, 662 (1945); (c) Yanko, Mosher and Whitmore, *ibid.*, 67, 664 (1945).