186. *Experiments on the Synthesis of α-Biotin. Part I.*

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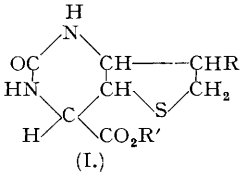
2-*Ethoxalyl*-3-*keto*-4-*ethyltetrahydrothiophen* (III; R = R′ = Et) has been synthesised as a possible intermediate to the bicyclic system (I).

The yeast growth promoting factor, biotin (bios IIb) has been shown to be identical with coenzyme R (Nilssen, Bjälfve and Burström, *Naturwiss.*, 1939, **27**, 389) and Vitamin H (du Vigneaud, Melville, György, and Rose, *Science*, 1942, **95**, 174). Later Kögl and ten Ham (*Z. physiol. Chem.*, 1943, **279**, 140) showed conclusively that the biotins isolated from egg yolk and liver were different substances; these were designated " α-biotin " and " β-biotin " respectively.

The structure of β-biotin has been elucidated by degradation and confirmed as 2′-keto-3 : 4-imidazolido-2-tetrahydrothiophen-*n*-valeric acid by total synthesis (S. A. Harris *et al.*, *J. Amer. Chem. Soc.*, 1944, **66**, 1756). Kögl, Verbeek, Erxleben, and Borg (*Z. physiol. Chem.*, 1943, **279**, 121) proposed structure (I; R = Pr^{β}, R′ = H) for α-biotin. They named the new bicyclic system " biotidin " and, numbering it as in the purines, refer to α-biotin as 2-keto-9-*iso*propyl-biotidin-6-carboxylic acid.

A short time ago we began work on a possible route to α-biotin and its homologues. Unfortunately, these experiments have been interrupted and we wish to place on record the results so far obtained.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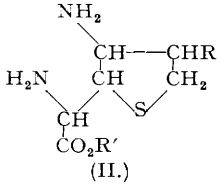
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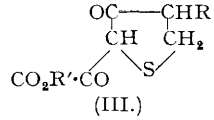
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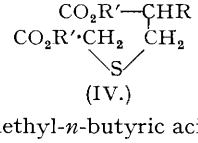
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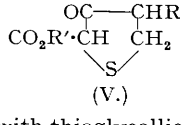
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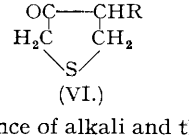
(VI.)

α-Bromomethyl-*n*-butyric acid was treated with thioglycollic acid in the presence of alkali and the resulting *dicarboxylic acid* (IV; R = Et, R′ = H) was esterified with ethanolic hydrogen chloride. Cyclisation of the *diester* (IV; R = R′ = Et) by the Dieckmann procedure gave the *keto-ester* (V; R = R′ = Et). The latter was hydrolysed to the *ketone* (VI; R = Et), and this was condensed with diethyl oxalate to give the *compound* (III; R = R′ = Et). 3-Keto-thiophans and their carboxy derivatives have been synthesised by very similar methods in connection with the synthesis of β-biotin, *e.g.*, Karrer *et al.*, *Helv. Chim. Acta*, 1944, **27**, 116, 124, 142, 237; Buchman and Cohen, *J. Amer. Chem. Soc.*, 1944, **66**, 847; Woodward and Eastman, *ibid.*, 1944, **66**, 849; Cheney and Piening, *ibid.*, 1944, **66**, 1040; 1945, **67**, 737; Avison *et al.*, *Nature*, 1944, **154**, 459; B.P. 562,313; 562,314; 567,428; 568,079.

An attempt to alkylate (VI; R = CO_2Et) (Karrer and Schmid, *Helv. Chim. Acta*, 1944, **27**, 124) with ethyl iodide in the presence of sodium in benzene yielded a negligible quantity of ethyl 3-keto-4-ethyl-tetrahydrothiophen-4-carboxylate, from which the ketone (VI; R = Et) might have been obtained by hydrolysis and decarboxylation.

α-Bromomethyl-*n*-butyric acid was prepared by the addition of hydrogen bromide to α-ethylacrylic acid (Blaise and Luttringer, *Bull. Soc. chim.*, 1905, [3], **33**, 766). The preparation of the latter has been described by Blaise and Luttringer (*loc. cit.*) and Mannich and Ganz (*Ber.*, 1922, **55**, 3492). The yields in both methods being unsatisfactory, we have devised a new method. This consists in treating ethylmalonic acid with formaldehyde in the presence of alkali to give hydroxymethyl-ethylmalonic acid. The dicarboxylic acid, when heated in acid solution, loses carbon dioxide and water to give α-ethylacrylic acid in good yield.

At the earliest opportunity we intend to attempt the conversion of (III; R = R′ = Et) into (II; R = R′ = Et) and thence to (I; R = R′ = Et) and (I; R = Et, R′ = H).

Experimental.

*α-Bromomethyl-*n*-butyric Acid.*—Diethyl ethylmalonate (140 g.) was added slowly to potassium hydroxide (120 g.) in water (96 c.c.) and the mixture refluxed for 16 hours. After cooling, formaldehyde (40%, 240 c.c.) was added and the whole kept for 10 days at room temperature. The mixture was acidified with concentrated hydrochloric acid (500 c.c.), and refluxed for 12 hours. The cold solution was extracted several times with ether. The residue from the dried ethereal extracts, after removal of solvent, was distilled and the fraction (40 g.), b. p. 176—186°, was collected. To it was added hydrogen bromide in acetic acid (120 c.c.; 50% w/v) and the mixture kept for 21 days. The product (31 g.) was collected at 124—128°/13 mm. (Found: C, 33·4; H, 5·0; equiv., 182. Calc. for $C_5H_9O_2Br$: C, 33·2; H, 5·0%; equiv., 181).

*Carboxymethyl β-Carboxy-*n*-butyl Sulphide and Carbethoxymethyl β-Carbethoxy-*n*-butyl Sulphide.* (IV; R = Et, R′ = H and R = R′ = Et).—A mixture of α-bromomethyl-*n*-butyric acid (31 g.) and water (20 c.c.) was neutralised in the cold with anhydrous potassium carbonate. The solution was added to a solution of thioglycollic acid (13·1 c.c. of 90% aqueous solution) and potassium hydroxide (18·7 g.) in water (10 c.c.) and the whole refluxed for 4 hours. After cooling, the solution was acidified to pH 4 and extracted 15 times with peroxide-free ether (50 c.c. each time). Removal of the solvent after drying left a colourless, viscous oil. A sample of this was distilled in a bulb-tube at 170°/0·01 mm. (air-bath temperature) (Found: C, 43·3; H, 6·3. $C_7H_{12}O_4S$ requires C, 43·7; H, 6·3%). The rest of the oil (22 g.) was refluxed for 4 hours with 7% ethanolic hydrogen chloride (100 c.c.). The alcohol was removed under reduced pressure and the residue treated twice again in the same way. Finally, after complete removal of alcohol, the residue was taken up in ether and washed with sodium bicarbonate solution. On distillation, the *di-ester* was obtained as a colourless oil (18·6 g.), b. p. 145—150°/11 mm. (Found: C, 52·7; H, 8·2; S, 13·4. $C_{11}H_{20}O_4S$ requires C, 53·2; H, 8·1; S, 12·9%).

Ethyl 3-Keto-4-ethyltetrahydrothiophen-2-carboxylate (V; R = R′ = Et).—Dry ethanol (2·9 c.c.) was added to powdered sodium (1·14 g.) under toluene (30 c.c.) and left to stand for some hours. To this was added the above di-ester (16 g.) and the mixture kept at 40—50° for 5 hours. The cooled solution was treated with water, acidified with acetic acid and extracted several times with ether. The ether–toluene extract was washed with sodium bicarbonate solution, dried and the solvent removed. On distillation the product was obtained as a colourless oil (8·6 g., 66%), b. p. 132—134°/15·5 mm. (Found: C, 54·3; H, 6·9; S, 15·7. $C_9H_{14}O_3S$ requires C, 53·5; H, 6·9; S, 15·8%). The *keto-ester* gives with alcoholic ferric chloride an intense bluish-violet colour which slowly fades. The *semicarbazone* was obtained in the usual way. It recrystallised from dilute alcohol as colourless needles, m. p. 159—160° (Found: C, 46·2; H, 6·6. $C_{10}H_{17}O_3N_3S$ requires C, 46·3; H, 6·6%). Substitution of ether for toluene gave a poorer yield (30%) of the keto-ester.

3-Keto-4-ethyltetrahydrothiophen (VI; R = Et).—The keto-ester (5·7 g.) was refluxed with 10% sulphuric acid (75 c.c.) for 5 hours. After cooling, the solution was extracted 8 times with ether (50 c.c. each time). The combined ethereal extracts were washed with sodium bicarbonate solution and water and dried. Removal of solvent under reduced pressure and distillation gave a colourless oil, b. p. 85°/21 mm. (bath temp.) (Found: C, 54·9; H, 7·7. $C_6H_{10}OS$ requires C, 55·4; H, 7·7%). The *dinitrophenylhydrazone* crystallised from alcohol as yellow needles, m. p. 137° (Found: N, 17·9. $C_{12}H_{14}O_4N_4S$ requires N, 18·0%).

2-Ethoxalyl-3-keto-4-ethyltetrahydrothiophen (III; R = R′ = Et).—A mixture of the ketone (2·0 g.), ethyl oxalate (3·0 g.) and dry, peroxide-free ether (10 c.c.) was added to powdered sodium (0·46 g.) under dry, peroxide-free ether. After addition of alcohol (0·5 c.c.), the mixture was gently refluxed for 3 hours and kept for 24 hours at room temperature. Cold water was added, the aqueous layer acidified with cold dilute hydrochloric acid and the product collected in ether. Removal of the solvent in vacuo left a brown solid (*ca.* 2 g.). It crystallised from alcohol in light yellow needles, m. p. 54—55° (Found: C, 52·1; H, 6·0; S, 14·4. $C_{10}H_{14}O_4S$ requires C, 52·2; H, 6·1; S, 13·9%). The *ester* in alcohol gives an intense, almost black, colour with aqueous ferric chloride.

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