

Wool Wax. Part V. The Constitution of the
α-Hydroxy-acids derived therefrom.*

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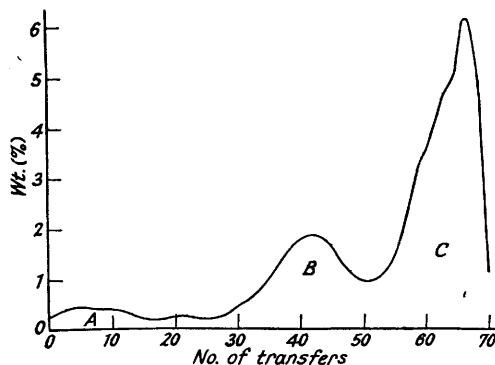
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The saponifiable fraction of wool wax from merino wool yields nearly 30% of a mixture of optically active 2-hydroxy-*n*-dodecanoic, -*n*-tetradecanoic, -*n*-hexadecanoic, -*n*-octadecanoic, and -16-methylheptadecanoic acid.

EARLIER investigations of the saponifiable fraction of wool wax (Lewkowitsch, *J. Soc. Chem. Ind.*, 1892, 11, 134; 1896, 15, 14; Darmstaedter and Lifschütz, *Ber.*, 1896, 29, 1474, 2890; Röhmann, *Biochem. Z.*, 1916, 77, 298; Abraham and Hilditch, *J. Soc. Chem. Ind.*, 1935, 54, 398T; Bertram, *J. Amer. Oil Chem. Soc.*, 1949, 26, 454) led to the view that it contains hydroxy-acids. Kuwata (*J. Amer. Chem. Soc.*, 1938, 60, 559) isolated a 2-hydroxy-*n*-hexadecanoic acid in 7% yield, and Weitkamp (*ibid.*, 1945, 67, 447) isolated also a 2-hydroxy-*n*-tetradecanoic acid in 0.2% yield.

Reduction, with lithium aluminium hydride, of a sample of the acids of wool wax extracted from merino wool and estimation of the 1 : 2-diols in the products with periodic acid indicated the presence of about 30% of α-hydroxy-acids in the original acid mixture. By solvent distribution, the mixture was separated into three broad groups (Figure) : a resinous, polar group of acids (11%) of high molecular weight, α-hydroxy-acids (27%), and unhydroxylated acids (62%).

A, Resin acids.
B, Hydroxy-acids.
C, Unhydroxylated acids.



As it was not possible to purify all the individual α-hydroxy-acids completely by solvent distribution with the 30-tube apparatus available, their methyl esters were first distilled, at 1 mm. pressure, in a micro-spinning-band fractionating column described previously (Part IV *). The pure components were isolated from the appropriate distillates by crystallisation, together, where necessary, with solvent distribution. The three pure α-hydroxy-acids obtained were identified, by oxidation with lead tetra-acetate in a current of air to the corresponding fatty acids containing one carbon atom less (cf. Mendel and Coops, *Rec. Trav. chim.*, 1939, 58, 1140), as 2-hydroxy-*n*-dodecanoic, -*n*-tetradecanoic, and -*n*-hexadecanoic acid. The C₁₈ ester fraction of the mixture is more thermolabile than the lower homologues and could not be resolved readily by fractional distillation. Also its physical properties were rather different from those of the other fractions and an infra-red study showed that it contained a large proportion of a branched-chain acid.

The wool wax acids were reduced with lithium aluminium hydride, and the 1 : 2-diols originating from the α-hydroxy-acids were separated in a yield of 22% by chromatography on active alumina. Distillation of the acetates, hydrolysis, and crystallisation of appropriate fractions yielded pure *n*-tetradecane-, *n*-hexadecane-, *n*-octadecane-, and 16-methylheptadecane-1 : 2-diol. After purification of the octadecanediol, too little

* Part IV, *J.*, 1953, 3533.

remained for further characterisation by oxidation. However, the infra-red absorption is identical with that of synthetic (\pm)-octadecane-1 : 2-diol. 16-Methylheptadecane-1 : 2-diol, on the other hand, showed the characteristic doublet in the 1360—1400-cm.⁻¹ range of the infra-red absorption spectrum (cf. Part IV). The material was further characterised by oxidation to 15-methylhexadecanoic acid, identical with a specimen prepared from 12-methyltridecanoic acid (Part III, *J.*, 1953, 98) by anodic coupling with the half ester of glutaric acid.

The two remaining α -hydroxy-acids in the mixture are thus characterised as 2-hydroxy-*n*-octadecanoic acid and -16-methylheptadecanoic acid. The properties of the compounds isolated are summarised in the Table.

R in R·CH(OH)·CO ₂ H	Me ester,		Acid,			Diol,	
	m. p.	b. p./l mm.	m. p.	[α] _D (CHCl ₃)	%	m. p.	diacetate, b. p.
C ₁₀ H ₂₁	24—25°	—	78.0—78.5°	—2.8°	2.2	—	—
C ₁₂ H ₂₅	34.8—35.0	133.5—136.5°	88.2—88.5 ^a	—3.1	13.5	77.6—77.9°	154.5°
C ₁₄ H ₂₉	45.5—45.7	153.5—154.0	93.3—93.5 ^b	—3.2	67.8	84.0—84.2	172
C ₁₆ H ₃₃	—	—	—	—	3.3	87.7—87.9	185
CHMe ₃ ·C ₁₃ H ₂₆ ...	ca. 40°	—	ca. 78°	—3.8	12.2	75.0—75.8	ca. 188

^a Weitkamp (*loc. cit.*) gives 88.2—88.5°, and Kuwata (*loc. cit.*) gives 86—87°. ^b Weitkamp (*loc. cit.*) gives 93.6°.

EXPERIMENTAL

Preparation of the Wool Wax Acids.—Saponification of wool wax extracted from merino wool and removal of the unsaponifiable material were carried out as described in Part IV (*loc. cit.*). The combined alcoholic solutions of the soluble potassium soaps were diluted with water and acidified and the fatty acids extracted with ether. After drying (Na₂SO₄), the ethereal extracts were evaporated to yield the main acid fraction. The yield (45—47 g. from 100 g. of wax) is about 90% of the acids present in the wax because the acids of low molecular weight which are very soluble in water and those with very high molecular weight which form potassium soaps insoluble in 60% ethanol were isolated separately.

Isolation of the Hydroxy-acid Fraction.—The α -hydroxy-acids were separated from the "resin acids" and the unhydroxylated acids by counter-current distribution of their methyl esters in an all-glass, 30-tube apparatus (Messrs. Otto Post & Co., New York, U.S.A.), each tube of which accommodated 100 c.c. of each phase. The methyl esters (30 g.) of the main acid fraction were introduced into the first three tubes and distributed between commercial hexane and 85% ethanol. Each fraction was separately evaporated and weighed (see Figure). The quantity of α -hydroxy-ester present in each fraction was determined by Hochstein's active hydrogen method (*J. Amer. Chem. Soc.*, 1949, **71**, 305). Those fractions which contained considerable quantities of hydroxy-esters were amalgamated (11.4 g.) and redistributed as before. In this way the esters were separated into three main groups: (a) those of high molecular weight, dark and polar methyl esters of the "resin acids" (3.3 g., 10.8%); (b) α -hydroxy-esters (8.2 g., 27.3%); and (c) unhydroxylated esters (18.6 g., 61.9%).

Separation of the α -Hydroxy-acids.—The material (4.5 g.) representing the peak of the second distribution curve was nearly pure methyl 2-hydroxy-*n*-hexadecanoate. Crystallised from ether and then from acetone, it formed a white powder (3.1 g.), m. p. 45.5—45.7° (Weitkamp, *loc. cit.*, records m. p. 45.6°), [α]_D -3.6° (c, 10 in CHCl₃) (Found: C, 71.4; H, 12.0; active H, 0.72%; sap. equiv., 288. Calc. for C₁₇H₃₄O₃: C, 71.3; H, 12.0; active H, 0.70%; equiv., 286). Saponification yielded 2-hydroxy-*n*-hexadecanoic acid, a white powder (from acetone, then hexane), [α]_D +3.6° (c, 6 in pyridine) (cf. Table) [Kuwata, *loc. cit.*, records [α]_D -1.0° (c, 5.2 in EtOH)] (Found: C, 70.4; H, 11.9; active H, 0.75%; equiv., 272. Calc. for C₁₆H₃₂O₃: C, 70.5; H, 11.8; active H, 0.74%; equiv., 272). Oxidation of the acid with lead tetra-acetate (method: Mendel and Coops, *loc. cit.*) yielded pentadecanoic acid, m. p. 51.5—52.0° (Found: C, 74.5; H, 12.3%; equiv., 241. Calc. for C₁₅H₃₀O₂: C, 74.3; H, 12.5%; equiv. 242), which did not depress the m. p. of pentadecanoic acid (m. p. 52.0—52.5°) prepared by oxidation of synthetic (\pm)-2-hydroxy-*n*-hexadecanoic acid.

The remaining fractions from the second distribution, together with the mother-liquors from the crystallisation of the methyl-2-hydroxy-*n*-hexadecanoate (altogether 5.1 g.), were redistributed in a 90-transfer experiment, being separated into three well-defined groups. Analysis of the material from the tube at the middle of each group, after crystallisation, corresponded very closely to the values expected for methyl 2-hydroxytetradecanoate, 2-hydroxy-

hexadecanoate, and 2-hydroxyoctadecanoate, respectively. A value was assumed for the partition coefficient of methyl 2-hydroxydodecanoate (which did not form a well-defined peak in the distribution curve) based on the differences found between the coefficients of the other members (C_{14} , C_{16} , C_{18}) of the homologous series. Then, allowing for the quantity of 2-hydroxy-*n*-hexadecanoic acid removed by crystallisation, it was possible to calculate fairly accurately the proportions of the individual hydroxy-acids present. These were in close agreement with those obtained later from the distillation curves of the diol acetates.

The methyl esters (20 c.c.) of the hydroxy-acids obtained by solvent distribution of total wool-wax fatty acids were separated further by distillation at 1 mm. in the micro-spinning-band fractionating column described in Part IV (*loc. cit.*) into the following main fractions: (1) b. p. 100—127° (0.8 c.c.); (2) b. p. 133.5—136.5° (1.0 c.c.); (3) b. p. 153.5—154.0° (9.8 c.c.); and (4) b. p. 165.5—172.5° (3.8 c.c.).

*Methyl 2-hydroxy-*n*-dodecanoate.* Fraction 1 was purified by counter-current distribution between hexane and 85% ethanol (40 transfers). The middle fraction, recrystallised from ether at -10° to -20°, formed a microcrystalline powder, m. p. 24—25°, $[\alpha]_D -3.0^\circ$ (*c*, 9 in $CHCl_3$) (Found: C, 68.0; H, 11.1; active H, 0.43%; sap. equiv., 234. $C_{13}H_{26}O_3$ requires C, 67.8; H, 11.4; active H, 0.44%; equiv., 230). The ester (94.3 mg.) was reduced with lithium aluminium hydride, the diol produced (48.1 mg.) was oxidised with periodic acid (Karnovsky and Rapson, *J. Soc. Chem. Ind.*, 1946, 65, 138), and the formaldehyde precipitated as the dimedone compound (74.6 mg.), m. p. 184—185° [$C_{12}H_{26}O_2$ (48.1 mg.) requires 69.2 mg. of dimedone compound, m. p. 189°].

The ester (182 mg.) gave the acid (128 mg.) which, twice recrystallised from acetone at -30°, formed a white powder (see Table) (Found: C, 66.9; H, 11.3; active H, 0.90%; equiv., 220. $C_{12}H_{24}O_3$ requires C, 66.6; H, 11.2; active H, 0.93%; equiv., 216). The acid (20 mg.) was oxidised with lead tetra-acetate, and the acid produced recrystallised from pentane at -20° as white powder, m. p. 26.5—27.0° (Found: equiv., 188. Calc. for $C_{11}H_{22}O_2$: equiv., 186), not depressing the m. p. of undecanoic acid, m. p. 27—27.5°, prepared by hydrogenation of undec-10-enic acid. The 1° discrepancy in m. p. indicates slight impurity.

*Methyl 2-hydroxy-*n*-tetradecanoate.* Fraction 2 (1 c.c.) was purified by counter-current distribution and crystallised from ether and then acetone as a white powder, $[\alpha]_D -3.6^\circ$ (*c*, 9 in $CHCl_3$) (Found: C, 70.1; H, 12.0; active H, 0.39%; sap. equiv., 261. Calc. for $C_{15}H_{30}O_3$: C, 69.7; H, 11.7; active H, 0.39%; sap. equiv., 258).

The derived free acid crystallised from acetone at -30° as a white powder (see Table) (Found: C, 68.8; H, 11.5; active H, 0.89%; equiv., 243. Calc. for $C_{14}H_{28}O_3$: C, 68.8; H, 11.6; active H, 0.82%; equiv., 244).

Oxidation of the acid with lead tetra-acetate yielded *n*-tridecanoic acid, m. p. 40.5—41.0° (Found: equiv., 215. Calc. for $C_{13}H_{26}O_2$: equiv., 214), alone or mixed with an authentic sample, m. p. 41.0—41.5°, prepared by oxidation of synthetic (\pm)-2-hydroxy-*n*-tetradecanoic acid.

Methyl 2-hydroxyoctadecanoate isomers. Fraction 4 was purified by counter-current distribution and crystallised from ether and then acetone as a white powder, m. p. 38.5—39.5°, $[\alpha]_D -4.5^\circ$ (*c*, 6 in $CHCl_3$) (Found: C, 72.3; H, 12.2%; active H, 0.32%; sap. equiv., 316. Calc. for $C_{19}H_{38}O_3$: C, 72.6; H, 12.2; active H, 0.32%; equiv., 315).

The derived mixed acids crystallised from acetone as a white powder (cf. Table) (Found: C, 71.8; H, 12.0%; active H, 0.64%; equiv., 302. Calc. for $C_{18}H_{36}O_3$: C, 72.0; H, 12.1; active H, 0.64%; equiv., 300). Oxidation with lead tetra-acetate yielded an acid, m. p. 57—58° (Found: C, 75.5; H, 12.8%; equiv., 268. Calc. for $C_{17}H_{34}O_2$: C, 75.5; H, 12.7%; equiv., 270). The mixed m. p. with synthetic 15-methylhexadecanoic acid, m. p. 60.3—60.5°, was about 58—59°.

Synthesis of 15-Methylhexadecanoic Acid.—12-Methyltridecanoic acid (4.56 g.) (Part III, *J.*, 1953, 101) and ethyl hydrogen glutarate (9.4 g.; Hesse and Bücking, *Annalen*, 1949, 563, 31) were added to a solution of sodium (0.003 g.) in methanol (250 c.c.), and the mixture electrolysed to yield 15-methylhexadecanoic acid (1.88 g.). It was purified by counter-current distribution (see Part III). It crystallised from acetone in plates, m. p. 60.3—60.5° (Weitkamp, *loc. cit.*, records m. p. 60.2°) (Found: C, 75.5; H, 12.7%; equiv., 270. Calc. for $C_{17}H_{34}O_2$: C, 75.5; H, 12.7%; equiv., 270).

Synthesis of (\pm)- α -Hydroxy-acids.—In the synthesis of (\pm)- α -hydroxy-acids by bromination of a fatty acid and subsequent hydrolysis, Bagard's method (*Bull. Soc. chim.*, 1907, 1, 30) which uses 0.33 mol. of phosphorus trichloride, gave a purer bromo-acid than did that of Clark and Taylor (*Org. Synth.*, 1941, Coll. Vol. I, p. 115).

Characterisation of the Hydroxy-acid Mixture by Fractionation of the Diols.—Wool-wax acids

(300 g.), after methylation, were reduced with lithium aluminium hydride in the usual way, to yield a mixture of monohydric and dihydric alcohols (258 g.). Acetylation yielded the corresponding acetates which were distilled at 1 mm. into two main fractions: (a) b. p. up to 158° and (b) b. p. 158—205°. These were separately saponified and chromatographed on alumina (cf. Part IV). The two glycol fractions obtained were combined (ca. 56 g., 80%), acetylated, and distilled at 1 mm. in the micro-spinning-band fractionating column to yield main fractions: (1) b. p. 154·5°; (2) b. p. 172°; (3) b. p. 185°; and (4) b. p. 187—192°.

n-Tetradecane-1 : 2-diol. Fraction 1 was hydrolysed and the *diol* produced crystallised from hexane, and then methanol, as plates (Found: C, 73·0; H, 13·1. $C_{14}H_{30}O_2$ requires C, 73·0; H, 13·1%).

n-Hexadecane-1 : 2-diol. Fraction 2, the *diacetate*, crystallised from methanol as long needles, f. p. 18·4°, n_D^{20} 1·4394 (Found: C, 70·1; H, 11·3; Ac, 25·0. $C_{20}H_{38}O_4$ requires C, 70·1; H, 11·2; Ac, 25·1%), and gave the diol, plates (from hexane and then acetone) (Found: C, 74·5; H, 13·4. Calc. for $C_{16}H_{34}O_2$: C, 74·4; H, 13·3%).

16-Methylheptadecane-1 : 2-diol. Fraction 3 was hydrolysed to the diol, plates (from hexane) (Found: C, 75·6; H, 13·5. Calc. for $C_{18}H_{38}O_2$: C, 75·5; H, 13·4%). The pure diol was oxidised with lead tetra-acetate to 15-methylhexadecanoic acid, plates, m. p. 60·2° (Found: C, 75·4; H, 12·7%; equiv., 270. Calc. for $C_{17}H_{34}O_2$: C, 75·5; H, 12·7%; equiv., 270). The infra-red spectra of both the diol and the degraded acid confirmed the view that they contain a terminal *isopropyl* group.

n-Octadecane-1 : 2-diol. Fraction 4 was hydrolysed and crystallised from hexane, ethanol, carbon tetrachloride and methanol, to give the *diol* as plates (Found: C, 75·3; H, 13·5. $C_{18}H_{38}O_2$ requires C, 75·5; H, 13·4%). The infra-red absorption spectrum was identical with that of synthetic (\pm)-octadecane-1 : 2-diol (see Part IV).

For other physical constants see the Table.

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