CCCXXII.—*Plant Cuticles. Part I (cont.). Modern Plant Cuticles.*

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IN continuation of our previous work (J., 1925, **127**, 1412), the behaviour of cutin from modern plants towards alkalis and oxidising agents, and on destructive distillation, has been studied for comparison with the behaviour of cutin separated from coal (see Part II; following paper). The wax associated with the cutin in modern plant cuticles has also been examined, in view of the probable contribution to coal of similar wax associated with the cuticles of the coal measure plants.

Our previous work was carried out with cuticle from *Agave americana*. Having exhausted our stock of this material, and being unable to obtain further supplies from the same source, we were fortunate enough to secure, through the kindness of the Public Works Department, Ashanti, a quantity of crude cuticle from another species of agave, namely, *Agave rigida*. This cuticle was resolved, in the manner previously described (*loc. cit.*, p. 1413), into (1) water-soluble material, 10%; (2) wax, 20%; (3) cellulose, 15%; and cutin, 55%.

Examination of the Cutin.—Treatment of the cutin from *Agave rigida* with alkalis resulted in the production of acid compounds substantially the same in quantity and kind as those from the cutin of *Agave americana*.

Treatment with dilute nitric acid as an oxidising agent gave a 60% yield of acid products, from which three types of acid have been isolated: (1) Insoluble in water, but readily soluble in cold alkali, corresponding on analysis with the formula $C_{11}H_{20}O_3$; (2) insoluble in water and in cold alkali, formula $C_{22}H_{42}O_3$; and (3) soluble in water. From the acids soluble in water, suberic acid was isolated in a pure state. A mixture of azelaic and sebacic acids in nearly equal proportion was also present.

Treatment of the cutin with hydrogen peroxide in the presence of glacial acetic acid yielded the (No. 2) acid, $C_{22}H_{42}O_3$.

On distillation in a vacuum, the cutin yielded 60% of condensable products containing approximately: saturated hydrocarbons, 38; unsaturated hydrocarbons, trace; aromatic hydrocarbons, 16; phenolic and acidic oils, 26; oxygenated compounds, 8; ether-soluble resin, 4; chloroform-soluble pitch, 7%. A notable feature of the distillation was the high yield of hydrocarbons, about one quarter of the total distillate consisting of pentatriacontane, $C_{35}H_{72}$, a hydrocarbon that has frequently been isolated, by extraction with solvents, from bituminous coals.

Examination of the Wax.—The wax extracted by solvents from the raw cuticle consisted mainly of monohydric alcohols, montanyl, $C_{28}H_{58}O$, and mellisyl, $C_{30}H_{62}O$, predominating. Smaller quantities of saturated and unsaturated fatty acids, hydrocarbons, and resins were also present.

EXPERIMENTAL.

Resolution of Cutin by Oxidation.—(1) *With dilute nitric acid.* 100 G. of cutin were boiled under reflux with 6 l. of 30% nitric acid during 2 hours. The evolution of acid fumes had then ceased and a brown oil, which floated on the surface of the hot acid solution, had been formed. On cooling, the acid solution, from which a quantity of crystalline material had separated, was decanted and evaporated to dryness (yield, 5%); the oil had hardened to a pale brown crust. This was washed free from acid and dried (yield, 55%).

Acids insoluble in cold water. The brown crust was treated with successive quantities of cold dilute alkali and filtered, until the filtrates were no longer coloured brown.

On acidifying the filtrates, a pale yellow flocculent precipitate was obtained. This was recovered from solution in warm ethyl ether as a pale yellow oil which formed a yellow translucent solid on cooling (soluble in ethyl ether, acetone, and ethyl alcohol; insoluble in light petroleum, chloroform, and benzene) (Found: C, 65·5; H, 10·0. $C_{11}H_{20}O_3$ requires C, 65·0; H, 10·0%). Yield, 43%. The copper salt was insoluble in alcohol. The *silver* salt was prepared in the usual manner from the neutral ammonium salt (Found: Ag, 34·9. $C_{11}H_{19}O_3Ag$ requires Ag, 35·1%).

The residue, insoluble in alkali, was a white solid. This was suspended in dilute hydrochloric acid, filtered off, washed free from acid, dried, and extracted in a Soxhlet apparatus with light petroleum (b. p. 60—80°). Nearly all the material was soluble in the light petroleum and could be crystallised from that solvent. M. p., after recrystallisation, 85—86° (Found: C, 75·0; H, 12·0. $C_{22}H_{42}O_3$ requires C, 74·6; H, 11·9%). Yield, 12%. The *silver* salt was prepared with some difficulty from a neutral solution of the ammonium salt, the slight solubility of the latter necessitating a large volume of solution (Found: Ag, 23·0. $C_{22}H_{41}O_3Ag$ requires Ag, 23·4%).

Acids sparingly soluble in cold water. The material remaining after evaporation of the nitric acid solution consisted of a yellow crystalline solution contaminated with a little dark oil. After several recrystallisations from boiling water, the mixture was resolved into two principal portions: a white crystalline solid, m. p. 137—139° (Found: C, 55·4, 55·4; H, 8·0, 8·0. Calc. for $C_8H_{14}O_4$, suberic acid: C, 55·2; H, 8·0%), and a pale yellow, crystalline powder of indefinite m. p. 90—95° (Found: C, 58·0; H, 8·4. Calc. for $C_9H_{16}O_4$, azelaic acid: C, 57·4; H, 8·5%, and for $C_{10}H_{18}O_4$, sebacic acid: C, 59·4; H, 8·9%).

(2) *With hydrogen peroxide.* 100 G. of cutin were kept in contact with a mixture of equal volumes of (100 vol.) hydrogen peroxide and glacial acetic acid at room temperature during 2 weeks. The acid solution was then decanted, and the residual cutin dried and exhaustively extracted with acetone under reflux. From the acetone solution a yellowish-green solid was obtained. On submitting the residual cutin to a second treatment with hydrogen peroxide, a further quantity of acetone-soluble material was obtained (total yield, 50%). This was mixed with sand and extracted with light petroleum in a Soxhlet apparatus. The extract, after several recrystallisations, was a pale yellow powder, m. p. 83° (Found: C, 74·4, 74·5; H, 11·9, 12·0. $C_{22}H_{42}O_3$ requires C, 74·6; H, 11·8%). The *silver* salt was prepared from a neutral solution of the ammonium salt (Found: Ag, 23·7. $C_{22}H_{41}O_3Ag$ requires Ag, 23·4%). The

ethyl ester, prepared from the silver salt and ethyl iodide, separated from light petroleum as a white powder, m. p. 63° (Found : C, 76·1; H, 12·1. $C_{26}H_{50}O_3$ requires C, 76·1; H, 12·2%).

Destructive Distillation of Cutin.—(1) *Condensable products.* 30 G. of dried cutin were distilled under high vacuum in a vertical glass retort with inverted neck (see Holroyd and Wheeler, J., 1928, 2669). The temperature was raised to 100° whilst the retort was being evacuated and thereafter, by increments of 50°, to 500°, the distillation lasting 10 days. A thin red oil condensed at 280°, but most of the condensable products were evolved between 300° and 350° as a yellow oil which partly solidified on cooling. The distillate was dissolved in chloroform, filtered and recovered as a pale brown semi-solid (yield, 60% of the ash-free, dry cutin). This was fractionated in the manner described by Holroyd and Wheeler (*loc. cit.*); there were then obtained : saturated hydrocarbons, 38; unsaturated hydrocarbons, trace; aromatic hydrocarbons, 16; phenolic and acidic oils, 26; oxygenated compounds, 8; ether-soluble resin, 4; and chloroform-soluble pitch, 7%.

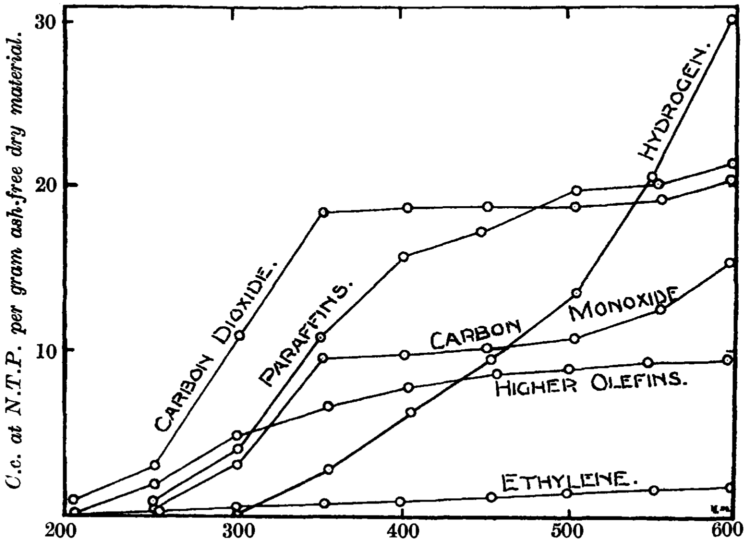
A portion of the original distillate was dissolved in chloroform and shaken successively with 10% sodium carbonate and 10% sodium hydroxide solutions, whereby acidic compounds were removed. The residue of neutral compounds, free from solvent, was treated with successive small quantities of cold acetone on a filter until the filtrate was no longer coloured. The material insoluble in cold acetone, a brown powder, was repeatedly crystallised from hot alcohol, whence it was obtained as a white solid, m. p. 70—72° (Found : C, 85·4; H, 14·5; *M*, ebullioscopic in benzene, 483, 496, 468. Calc. for $C_{35}H_{72}$, pentatriacontane, m. p. 75° : C, 85·4; H, 14·5%; *M*, 492). Yield, 20% of distillate.

(2) *Gaseous products.* 1 G. of dried cutin was distilled in a vertical glass retort, at 100° whilst the retort was being evacuated and thereafter at successive intervals of 50° up to 500°. The gases evolved over each temperature range were collected separately and analysed. The results (which should be compared with similar distillations of coal and coal-forming materials; see also Part II) are recorded graphically in the figure.

Examination of the Wax from Cuticle.—50 G. of the wax, a pale green solid, were mixed with sand and extracted with light petroleum (b. p. 60—80°) in a Soxhlet apparatus. The extract (yield, 42·5 g. or 85%) was resolved by the same solvent into sparingly soluble and readily soluble fractions.

The sparingly soluble fraction (37 g.), a white solid, m. p. 80—85°, was boiled during 2 hours with excess of 10% alcoholic potassium hydroxide. The solution was evaporated to dryness, and the residue mixed with sand and extracted with light petroleum. The extract, which proved to be a mixture of wax alcohols, was acetylated by means of acetic anhydride and the mixed acetates, after the removal of a small quantity of resinous matter by boiling alcohol, were then divided into two portions by fractional crystallisation from light petroleum. The alcohols recovered from each fraction appeared to be contaminated with traces of hydrocarbons, obstinately retained. They were therefore converted into the corresponding acids, by mixing them with 1½ times their weight of potash-lime, heating during 2 hours on an oil-bath at 260°, extracting the mixture (when cold) with light petroleum to remove hydrocarbons, and recovering the acids from the potassium salts in the usual manner. On recrystallisation from light petroleum, there were obtained: (1) A white crystalline powder (14 g.), m. p. 84—85° (Found: C, 79·3; H, 13·1. $C_{28}H_{56}O_2$, the acid corresponding to montanyl alcohol, requires C, 79·2; H, 13·2%); and (2) a pale yellow, crystalline powder (12 g.), m. p. 82—83° (Found: C, 79·4; H, 13·5.) $C_{30}H_{60}O_2$, the acid corresponding to mellisyl alcohol, requires C, 79·6; H, 13·3%). The acids (8 g.) recovered from the saponifiable portion were freed from traces of unsaturated compounds by means of their lead salts. After recrystallising from a mixture of alcohol and water, they melted at 53—57° (Found:

Distillation of modern plant cutin in a vacuum. Gases evolved in c.c. per gram ash-free dry material.



C, 75·9; H, 12·6. Calc. for $C_{16}H_{32}O_2$: C, 75·0; H, 12·5%. Calc. for $C_{18}H_{36}O_2$: C, 76·0; H, 12·7%).

The principal constituents of the portion of the wax sparingly soluble in light petroleum thus appear to be montanyl and mellisyl alcohols, both free and combined with small quantities of stearic and palmitic acids.

The portion of the wax readily soluble in light petroleum (5 g.) was a pale green semi-solid. This was boiled with excess of 10% alcoholic potassium hydroxide, the solution evaporated, and the residue extracted with light petroleum, from which most of the material was recovered unchanged. This was resolved by means of ferric chloride in concentrated hydrochloric acid solution into semi-solid hydrocarbons (3 g.) and oxygenated compounds (1 g.), which were not further examined.

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