181. *Occurrence of Betulinic Acid in the Bark of the Plane Tree.*

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The substance isolated from the shed bark of *Platanus acerifolia* by sublimation or extraction, m. p. 311—313° (decomp.), is identical with betulinic acid. Doubtless, "platanol" and "platanin", described by Dávid (*Magyar Gyógy. Társ. Ert.*, 1944, 8) and by Jaretzky (*Arch. Pharm.*, 1944, **282**, 75), respectively, and probably "platanolic acid", mentioned by Zellner and Ziffer (*Monatsh.*, 1925, **46**, 325), are also identical with betulinic acid.

Zellner and Ziffer (*loc. cit.*) isolated from the bark of the branches of young *Platanus orientalis* a hydroxy-carboxylic acid, to which they ascribed the formula $C_{24}H_{40}O_3$, calling it "platanolic acid". They observed that it was optically inactive and showed a positive Liebermann reaction (red ⟶ violet ⟶ blue ⟶ green), and they prepared derivatives recorded in the table. Their formula was supported only by a duplicate analysis and a molecular-weight determination by Rast's method. A methoxyl determination for the methyl ester gave a very high result.

Jaretzky (*loc. cit.*) isolated from the chloroform extract of the bark of *Platanus orientalis* a very similar substance, which he called "platanin", two analyses indicating the formula $C_{30}H_{50}O_4$ or $C_{30}H_{51}O_4$ (*sic*).

Dávid (*loc. cit.*) isolated an amorphous powder from the alcoholic extract of *Platanus acerifolia* bark and obtained crystalline needles by sublimation or by extraction with absolute methanol or ethanol. This product had m. p. 304—311° (decomp.), and formed a monoacetyl derivative, m. p. 287°. From analyses and molecular-weight determinations of this substance ("platanol") and its acetate, he deduced the formula $C_{28}H_{44}O_3$ or $C_{29}H_{46}O_3$. We have isolated and investigated this "platanol" and find it to be identical with betulinic acid (prepared by Ruzicka, Lamberton, and Christie, *Helv. Chim. Acta*, 1938, **21**, 1706, by oxidising betulin monoacetate), which was discovered shortly afterwards in the bark of *Cornus florida* (Robertson, Soliman, and Owen, *J.*, 1939, 1267) and in the seeds of *Zyzyphus vulgaris* (Kavaguchi and Kim, *J. Pharm. Soc. Japan*, 1940, **60**, 171).

Our conclusions are based on the following experimental data (see also table): (1) The physical constants, properties, and colour reactions of platanol are identical with those of betulinic acid; *e.g.*, pure platanol melts at 311—313° (decomp.), shows a positive Liebermann reaction (red ⟶ violet ⟶ blue ⟶ green), and is optically active ($[\alpha]_D^{22°}$ + 7·79° in pyridine). (2) Several analyses of platanol and its derivatives, and determination of molecular weights, especially the equivalent, support the formula $C_{30}H_{48}O_3$. (3) Platanol contains an easily acetylated hydroxyl group, and the acetate has m. p. 287—289°, regenerating the original substance on hydrolysis. (4) The presence of a carboxyl group in platanol was proved by titration in alcoholic solution and by esterification with diazomethane. (5) Platanol is unsaturated (yellow colour with tetranitromethane) and readily forms a dihydro-derivative, which can be acetylated and then esterified. (6) The hydroxyl group is secondary, since the dihydro-derivative can be oxidised to a ketone, forming an *oxime*. (7) Platanol yielded on bromination in chloroform solution at room temperature a *monobromo*-derivative, m. p. 250—251°. Although Robertson *et al.* (*loc. cit.*) found the bromination product of betulinic acid to be an "unseparable mixture of different bromo-derivatives", our substance was homogeneous, analysis being accurate for $C_{30}H_{47}O_3Br$. (This discrepancy is probably due to the different methods

used in the bromination.) The bromo-derivative forms an acetyl derivative, m. p. 280°, identical with that obtained by brominating the above acetate. This m. p. is 10° lower than that for the compound prepared by Robertson *et al.* (*loc. cit.*) by bromination of acetylbetulinic acid in ether–glacial acetic acid solution, but our product is not a lactone, for it gives acid reactions in alcoholic solution and can be esterified with diazomethane. (8) The isomeric lactone acetate, m. p. > 340°, results on treating platanol with glacial acetic acid–hydrochloric acid, or by various other methods. (9) M. p.s (and optical activity determined in several cases) of the platanol derivatives agree with those recorded for the corresponding derivatives of betulinic acid. (10) Selenium dehydrogenation yielded 1 : 2 : 5 : 6-tetramethylnaphthalene and a saturated hydrocarbon (not investigated, see Experimental).

We therefore conclude that " platanol " is identical with betulinic acid. Doubtless, Jaretzky's " platanin " (*loc. cit.*) is the same compound : his low analytical values can be explained by the fact that it readily retains small amounts of solvent. Zellner and Ziffer's " platanolic acid " too is probably the same as " platanol ", for it agrees in all its properties and reactions, and the m. p.s of their derivatives (except the bromo-compound) would have approximated to ours if they had been corrected, especially as betulinic acid slowly decomposes above 280°; moreover, their analyses are not in good agreement with the formula $C_{24}H_{40}O_3$, and our analyses of *impure* " platanol " agreed with theirs. Their statement that " platanolic acid " is optically inactive is not of much significance, for the specific rotatory power of betulinic acid is very low.

*Melting points of betulinic acid, platanol, platanolic acid, platanin, and their derivatives.**

	Betulinic acid.	Platanol (this paper).	Platanolic acid.	Platanin.
(I) Original substance	316—318° [b] 315—317 [c]	311—313°	281° [f]	313—315° [g] 321—323 [g]
(II) Acetyl deriv.	288—290 [a] 281—291 [b]	287—289	277 [f]	—
(III) Dihydro-deriv.	323—324 [a] 316—320 [b] 317—319 [c]	311—315	—	—
(IV) Acetyldihydro-deriv.	311—312·5 [e] 307—310 [b] 308—310 [c]	307	—	—
(V) Keto-dihydro-deriv.	256—257 [e, d]	255—258	—	—
(VI) Oxime of (V)	—	285	—	—
(VII) *Acetate* of (VI)	—	150—151	—	—
(VIII) Methyl ester of (I)	224—225 [a] 223—224 [b]	224—225	214—215 [f]	—
(IX) Methyl ester of (II)	200—202 [a] 201—202 [b]	203	—	—
(X) Methyl ester of (IV)	238·5—239 [a, e] 237—239 [c]	235	—	—
(XI) Monobromo-deriv. of (I)	—	250—251	211 [f]	—
(XII) Monobromo-deriv. of (II)	290 [b]	280	—	—
(XIII) *Methyl* ester of (XII)	—	138—140	—	—
(XIV) Lactone acetate	>350 [b] 357—360 [b] 344—346 [c]	>340	—	—

* M. p.s are corrected, except those of platanolic acid and its derivatives.

(*a*) Ruzicka, Lamberton, and Christie, *loc. cit.*
(*b*) Robertson, Soliman, and Owen, *loc. cit.*
(*c*) Kawaguchi and Kim, *loc. cit.*
(*d*) *Idem, ibid.*, p. 235.
(*e*) Ruzicka, Brenner, and Rey, *Helv. Chim. Acta*, 1941, **24**, 515.
(*f*) Zellner and Ziffer, *loc. cit.*
(*g*) Jaretzky, *loc. cit.* The first m. p. was obtained by rapid heating, and the second by using a Berl block preheated at 310°.

Experimental.

(Melting points are corrected.)

Isolation.—The bark (2000 g.) of *Platanus acerifolia* (shed in spring) was cut into small pieces and extracted for 9 hours with methanol (5·5 l.). The extract was concentrated to 500 ml. and cooled; the substance crystallised, and was filtered off, washed with cold methanol, and dried (36 g.). The dirty-green product, recrystallised from absolute methanol (charcoal), afforded colourless needles (14 g.). For analysis, it was recrystallised 5 times from absolute methanol, and dried at 130° in a vacuum; m. p. 311—313° (decomp.), $[\alpha]_D^{22°}$ + 7·79° in pyridine (Found : C, 78·6, 78·9; H, 10·4, 11·0. Calc. for

$C_{30}H_{48}O_3$: C, 78·85; H, 10·6%). The substance further purified through the acetate (below) gave: C, 78·8, 78·9, 78·8; H, 10·8, 10·7, 10·8; *M* (Rast), 451·8; *M* (by titration), 460·9 (Calc.: *M*, 458·7).

Acetate (II).—Platanol (2 g.) was dissolved in boiling acetic anhydride (10 ml.) or boiled for a short time with glacial acetic acid. The product precipitated by cooling was recrystallised from methanol. For analysis it was dried for 4 hours at 130° in a vacuum; m. p. 287—289° (decomp.), $[\alpha]_D^{22°}$ + 12·09° in pyridine [Found: C, 76·8, 77·2; H, 10·1, 10·0; *M* (ebullioscopic), 497·5; *M* (by titration), 498·2. Calc. for $C_{32}H_{50}O_4$: C, 77·1; H, 10·1%; *M*, 498·7). When this acetate (2 g.) was boiled with 10% methanolic potash (35 ml.) for 30 minutes, and then acidified, the product, recrystallised from absolute methanol, was identical with the original substance [m. p., mixed m. p., and analysis (see above)].

Acetyldihydrobetulinic Acid (IV).—This was prepared (*a*) by hydrogenation in presence of a platinum oxide catalyst of a solution of platanyl acetate (1 g.) in absolute ethanol (300 ml.) or ethyl acetate (40 ml.), and (*b*) by boiling dihydroplatanol (III) (0·5 g.) with anhydrous acetic acid (10 ml.). Crystallised from methanol or glacial acetic acid, it yields colourless plates, m. p. 307° (decomp.) (Found: C, 76·65, 76·85, 77·0; H, 10·5, 10·5, 10·5. Calc. for $C_{32}H_{52}O_4$: C, 76·75; H, 10·5%).

Dihydrobetulinic Acid (III).—Pure platanol (1 g.), dissolved in ethanol (500 ml.), was shaken in an atmosphere of hydrogen in the presence of prehydrogenated platinum oxide (0·3 g.). Absorption of 1 mol. of hydrogen was complete in 60 minutes. The solution was concentrated, and the substance crystallised. It retains ½ mol. of ethyl alcohol even after drying at 140° in a vacuum (Found: C, 77·3, 77·2, 77·1; H, 11·2, 10·9, 10·9. Calc. for $C_{30}H_{50}O_3,\frac{1}{2}C_2H_6O$: C, 77·3; H, 11·1%), which it loses on drying at 200° (Found: C, 78·35; H, 11·05. Calc. for $C_{30}H_{50}O_3$: C, 78·55; H, 11·0%); m. p. 311—315° (decomp.). Unlike the original substance, this shows no colour reaction with tetranitromethane.

Dihydrobetulonic Acid (V).—To a solution of dihydroplatanol (1 g.) in glacial acetic acid (80 ml.) a solution of chromic acid (0·23 g.) in glacial acetic acid was added, and the whole warmed on a steam-bath for 60 mins. The solution was then evaporated, and the residue digested with water, filtered off, and dried. Recrystallised from acetone, it yielded delicate needles, m. p. 255—258° (decomp.) (Found: C, 78·7, 78·55; H, 10·7, 10·9. Calc. for $C_{30}H_{48}O_3$: C, 78·85; H, 10·6%).

Oxime (VI). The substance (V) (0·4 g.) was dissolved in ethanol and an alcoholic solution of hydroxylamine hydrochloride (0·05 g.), and a 2% alcoholic solution of potassium hydroxide (2 ml.) was added. After 24 hours the precipitated *oxime* was recrystallised from ethanol; needles, m. p. 285° (decomp.) (Found: C, 76·5, 76·25; H, 10·5, 10·55. $C_{30}H_{49}O_3N$ requires C, 76·4; H, 10·5%). The oxime (100 mg.) was boiled with acetic anhydride (5 drops), and the product cooled and mixed with excess of saturated potassium carbonate solution; the precipitated *acetate* (VII), crystallised from 70% ethanol and dried at 70° in a vacuum, had m. p. 150—151° (Found: C, 74·3, 74·0; H, 10·2, 10·5. $C_{32}H_{51}O_4N$ requires C, 74·5; H, 10·4%).

Methyl Betulinate (VIII).—Platanol (2 g.) was dissolved in methanol (350 ml.), and an ethereal solution of diazomethane added at 0°. The ester was precipitated on concentration of the solution; recrystallised from benzene (10 ml.), it formed colourless prisms, m. p. 224—225° (Found: C, 79·3; H, 10·8. Calc. for $C_{31}H_{50}O_3$: C, 79·1; H, 10·7%).

Methyl Acetylbetulinate (IX).—This was prepared from (II) (0·5 g.) by means of diazomethane. Recrystallised from methanol, and dried at 100° in a vacuum, it yielded small prisms, m. p. 203°, $[\alpha]_D^{6°}$ + 16·86° in chloroform (Found: C, 77·7; H, 10·3. Calc. for $C_{33}H_{52}O_4$: C, 77·3; H, 10·2%).

Methyl Acetyldihydrobetulinate (X).—The acetyl compound (IV) (0·2 g.) was dissolved in methanol (40 ml.) and treated with diazomethane. The crystalline substance obtained by concentrating the solution was recrystallised from methanol and dried at 100° in a vacuum, affording prisms, m. p. 235° (Found: C, 76·85; H, 10·65. Calc. for $C_{33}H_{54}O_4$: C, 77·0; H, 10·6%).

Bromobetulinic Acid (XI).—To a solution of platanol (2 g.) in cold chloroform a 4% chloroform solution of bromine (0·7 g.) was added in small portions. After separation of a small gelatinous precipitate, the solution was concentrated under reduced pressure at room temperature until crystallisation took place. The substance was filtered off, washed with ice-cold chloroform, and dried over paraffin in a vacuum at room temperature; colourless needles, m. p. 250—251° (decomp.) (Found, for crude substance: C, 67·1, 67·5; H, 8·6, 8·7. Found, for substance recrystallised from benzene, and dried at 60° in a vacuum: C, 67·5; H, 8·7. $C_{30}H_{47}O_3Br$ requires C, 67·3; H, 8·8%).

Acetylbromobetulinic Acid (XII).—(*a*) The foregoing substance (XI) was boiled with excess of acetic anhydride, and the precipitated acetyl derivative recrystallised from ligroin. (*b*) Platanol (1·6 g.) was dissolved in boiling glacial acetic acid (100 ml.), cooled to 40°, and a solution of bromine (0·6 g.) in glacial acetic acid added. The resulting solution was concentrated under reduced pressure, and the residue dried for 24 hours over potassium hydroxide in a vacuum at room temperature and finally recrystallised from benzene. (*c*) Substance (II) (1 g.) was dissolved in cold chloroform (20 ml.) and a 4% solution of bromine (0·35 g.) in chloroform was added. After evaporation at room temperature under reduced pressure, the residue was dissolved in a small amount of chloroform and diluted with 6—7 vols. of methanol; the substance crystallised in analytically pure state. The three products were identical; m. p. 280° (decomp.) [Found: (*a*) C, 66·6; H, 8·6; (*b*) C, 66·4, 66·5; H, 8·65, 8·9; (*c*) C, 66·3, 66·6; H, 8·7, 9·0. Calc. for $C_{33}H_{49}O_4Br$: C, 66·5; H, 8·55%].

Methyl Acetylbromobetulinate (XIII).—The foregoing substance (XII) (0·15 g.) was dissolved in cold methanol (20 ml.), and a solution containing 0·5 mol. of diazomethane (10 ml.) added at 0°. Concentration of the solution afforded a crystalline *ester*, which was recrystallised from methanol, and dried at 80° in a vacuum, giving delicate needles, m. p. 138—140° (Found: C, 66·5; H, 9·0. $C_{33}H_{51}O_4Br$ requires C, 67·0; H, 8·7%).

Acetylbetulinic Acid Lactone (XIV).—(*a*) Platanol (2 g.) was boiled with 98% formic acid (70 ml.). The greater part dissolved at first and then afforded a precipitate. Recrystallised from xylene, this yielded long needles, m. p. 330° (decomp.), which did not react with diazomethane or with bromine. As the analyses for this product were not satisfactory it was converted into its acetyl derivative by means of acetic anhydride, and this was recrystallised from xylene, yielding needles, m. p. > 340° (decomp.). (*b*) Substance (II) (1·5 g.) was boiled with 98% formic acid for 1 hour; the crystals which separated on cooling were washed with water, dried (1·1 g.), boiled with benzene (10 ml.), and recrystallised from

xylene, yielding needles, m. p. > 340° (decomp.). (*c*) Powdered platanol (5 g.) was heated with glacial acetic acid (35 ml.) containing 10% of hydrogen chloride for 90 minutes at 100° in a sealed tube. Water (70 ml.) was added, and the precipitate was washed with water, dried, and boiled for 10 minutes with acetic anhydride (60 ml.). The substance precipitated on cooling was boiled with benzene (40 ml.) and finally recrystallised from xylene; needles, m. p. > 340° (decomp.). (*d*) Powered platanol (5 g.) was refluxed with a mixture of glacial acetic acid (150 ml.) and hydrochloric acid (*d* 1·19; 150 ml.) for 2 hours. Only part dissolved, and the residue was filtered off; the filtrate was diluted with water (300 ml.), and the precipitate washed with water, dried, boiled with benzene, and recrystallised finally from xylene; needles, m. p. > 340° (decomp.). (*e*) Into a solution of platanol (1 g.) in ether–ethanol (2 : 1; 300 ml.), dry hydrogen chloride was passed for 2 hours. Concentration of the solution to 70 ml. precipitated an amorphous product, which was acetylated with acetic anhydride. The product crystallised from xylene in needles, m. p. > 340° (decomp.) [Found: (*a*) C, 76·9; H, 10·2; (*b*) C, 77·2; H, 10·3; (*c*) C, 77·1; H, 10·3. Calc. for $C_{32}H_{50}O_4$: C, 77·1; H, 10·1%].

Dehydrogenation of Platanol.—Platanol (30 g.) was heated in an atmosphere of nitrogen in a metal-bath to 240°, selenium (17 g.) was added, and the temperature increased to 265°; more selenium (7 g.) was then added and the temperature increased during 40 minutes to 335°. Evolution of hydrogen selenide began at 310°. After $1\frac{1}{2}$, $2\frac{1}{2}$, and 8 hours further portions of selenium (7 g.) were added, and heating was continued for another 30 hours at 335—340°. The dark residue was cooled and extracted with ether (250 ml.), and the extract filtered and evaporated. A dark brown fluorescent oil (24 g.) remained, which was fractionated under reduced pressure: (1) <120°/17 mm., 0·15 g. of lemon-yellow oil; (2) 120—140°/4 mm., 5·6 g. of yellow oil; (3) 140—240°/4 mm., 0·2 g. of brown oil which crystallised; (4) > 240°/4 mm., 1·0 g. of brown oil solidifying to a glass. Only fractions (2) and (3) were investigated.

Fraction (2). This was dissolved in absolute ethanol (15 ml.), and a solution of picric acid (2 g.) in ethanol (20 ml.) added. The red solution thus obtained was boiled for 10 minutes, then evaporated, and the residue collected on a fritted-glass filter and washed with cold ethanol. The crude picrate (2·4 g.) was recrystallised from absolute ethanol (20 ml.), yielding red needles (1·6 g.), m. p. 134—135°, raised by another six crystallisations to 143°, and by a seventh to 148° (Found: C, 58·0; H, 4·5; N, 10·2. Calc. for $C_{20}H_{19}O_7N_3$: C, 58·1; H, 4·6; N, 10·2%). Ruzicka *et al.* (*Helv. Chim. Acta*, 1933, **16**, 320) give m. p. 154—154·5° for pure 1 : 2 : 5 : 6-tetramethylnaphthalene picrate.

One part of the crude picrate, m. p. 134—135°, was treated with dilute ammonia, and the regenerated hydrocarbon was crystallised from ethanol and dried in a vacuum at 50—60°, forming colourless needles, m. p. 111—112° (Found: C, 91·5; H, 8·9. Calc. for $C_{14}H_{16}$: C, 91·25; H, 8·75%). As Ruzicka *et al.* (*loc. cit.*) give m. p. 116° for 1 : 2 : 5 : 6-tetramethylnaphthalene, we assume that our substance was a not quite pure specimen of this hydrocarbon.

Fraction (3). Dissolved in hot absolute ethanol (20 ml.), this yielded on cooling a small amount of colourless needles which, recrystallised from methanol, formed needles, m. p. 44—45°. The substance was saturated, for it gave no reaction with picric acid, showed no colour reaction with tetranitromethane, and did not discolour a solution of potassium permanganate or bromine. Analyses agreed with that for a C_{30} paraffin or a saturated hydrocarbon C_nH_{2n}. Molecular-weight determination (Rast) gave values between 420 and 480. We think it very improbable that by dehydrogenation a paraffin of this high carbon number could be obtained, but lack of material prevented further investigation (Found: C, 85·0, 85·6, 85·3; H, 14·6, 14·5, 14·7. Calc. for $C_{30}H_{62}$: C, 85·2; H, 14·8%. Calc. for C_nH_{2n}: C, 85·6; H, 14·4%).

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