CHEMICAL EXAMINATION OF THE LEAVES OF *RHODODENDRON CINNAMOMEUM*

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Received June 7, 1961

THE results of the chemical examination of the leaves of *R. falconeri*, *R. nilagiricum* and *R. campanulatum* have been reported in earlier communications from these laboratories.[1-3] *R. cinnamomeum* is a tree growing in the Darjeeling region of the Himalayas. This is a distinct species which is likely to be confused with *R. arboreum*, but can be easily recognised by the dense rusty tomentum on the lower surface of the leaves. Further, the flowers of *R. cinnamomeum* are not so bright red as those of *R. arboreum*.

A survey of the literature showed that the plant has not been examined so far.

The powdered leaf was extracted with petroleum ether, chloroform and alcohol in succession. The petroleum ether extract yielded crystal mixtures which, by fractional crystallization followed by chromatography over alumina, could be separated into three components. One melting at 280–82° was identified as epifriedelanol. Another melting at 258–60° was identified as friedelin.[4-6] A minor component, m.p. 74–76°, was not examined further. The chloroform extract yielded ursolic acid. The alcohol extract, when examined as described in the experimental section, yielded quercetin and a minor component.

Ursolic acid and quercetin were identified by their properties and those of their acetates and by mixed melting points.

Friedelin has so far been reported to occur in only one Rhododendron species, *viz.*, *R. westlandii*.[7] The identity of the substance obtained in the present investigation was deduced from its properties and by the preparation of its 2:4-dinitrophenylhydrazone[8] and enol-benzoate[6] and by reduction to friedelan-3α-ol.[6]

The identity of epifriedelanol (friedelan-3 β-ol) was established by the preparation of its acetate and by oxidation to friedelin (m.p. and mixed m.p.).

EXPERIMENTAL

The coarsely powdered leaf (3 kg.) was extracted with petroleum ether, then with chloroform and then with alcohol in the cold using 3×8 l. of each solvent. These were followed by a single extraction with hot alcohol.

Petroleum ether extract.—The dark-green extract was concentrated to 250 ml. and left overnight at room temperature when a heavy crystalline material deposited. This was filtered and washed with warm petroleum ether (fraction A, 3 g.). The filtrate was further concentrated, the syrupy residue treated with hot acetone (150 ml.) and the mixture allowed to stand for a few days at room temperature, when crystals deposited along with some waxy matter. This was filtered and the wax washed off with warm acetone leaving behind fraction B (3 g.). The combined mother liquor, on removal of the solvent, yielded a dark-green residue (residue X, 76 g.) whose examination is described later.

Fractions A and B.—These two fractions had similar properties. They were united and crystallized from excess of chloroform, when clusters of colourless glistening plates slowly separated. These were filtered and recrystallized from chloroform, m.p. 280–82° (epifriedelanol, 2·8 g.; for characterisation *see* later). The residue (3 g.) from the combined mother liquor was chromatographed over alumina (90 g.) employing 200 ml. portions of solvents for elution. The course of the chromatogram is shown in Table I.

Fractions 1 and 2 were united and crystallized twice from benzene-acetone when colourless nodules were obtained, m.p. 74–76° (minor component, 0·15 g.).

Fractions 3 to 6 were united and crystallized twice from benzene, when long colourless needles were obtained, m.p. 258–60° (friedelin, 1·4 g.; for characterisation *see* later).

Fractions 7 to 9 were united and crystallized from chloroform as colourless plates, m.p. 278–80° (epifriedelanol, 0·4 g.).

Residue X (mentioned earlier) was saponified with 10% sodium hydroxide in benzene-alcohol (1 : 9) solution and the unsaponifiable matter (30 g.) was extracted in the usual way. It was dissolved in warm acetone (100 ml.) and left at room temperature for a week. The crystals that deposited were filtered, washed and recrystallized from chloroform, when colourless plates were obtained, m.p. 278–80° (epifriedelanol, 0·2 g.). $[\alpha]_D^{28} = +21{\cdot}1° \pm 2°$ ($c = 0{\cdot}824$ in chloroform). Mixed m.p. with epifriedelanol obtained above was undepressed. The rest of the unsaponifiable matter was rejected.

TABLE I

Fraction No.	Eluate	Residue	
		Weight in mg.	M.p. after a single crystallization
1.	Petroleum ether ..	7	70–72°
2.	Petroleum ether-benzene (19 : 1) ..	160	72–73°
3.	„ (9 : 1) ..	340	248–55°
4.	„ (4 : 1) ..	540	250–54°
5.	„ (1 : 1) ..	830	250–54°
6.	Benzene ..	210	254–56°
7.	Benzene-chloroform (19 : 1) ..	150	265–67°
8.	„ (9 : 1) ..	220	266–69°
9.	„ (4 : 1) ..	120	268–70°
10.	„ (1 : 1) ..	50	Amorphous
11.	Chloroform ..	20	„

Minor component.—M.p. 74–76°. No colour in the Salkowski or Liebermann-Burchard test or with tetranitromethane. $[\alpha]_D^{29} = +21{\cdot}2° \pm 3°$ ($c = 0{\cdot}6141$ in chloroform). Found: C, 82·4; H, 14·6. $C_{29}H_{60}O$ (probable formula) requires: C, 82·0; H, 14·2%.

Friedelin.—Colourless needles from benzene, m.p. 258–60°. Salkowski, Liebermann-Burchard and tetranitromethane reactions were negative. $[\alpha]_D^{28} = -23{\cdot}5° \pm 2°$ ($c = 1{\cdot}023$ in chloroform). Found: C, 84·7; H, 12·3. $C_{30}H_{50}O$ (friedelin) requires: C, 84·5; H, 11·8%.

The 2 : 4-dinitrophenylhydrazone[8] crystallized from chloroform-alcohol as shining orange-red plates, m.p. 301–05° (decomp.). Found: N, 9·7. $C_{36}H_{54}O_4N_4$ (friedelin 2 : 4-dinitrophenylhydrazone) requires: N, 9·2%.

The enol-benzoate, prepared as described by Corey and Ursprung[6] crystallized from ethyl acetate-chloroform as colourless plates, m.p. 262–65°.

$[\alpha]_D^{29} = +59{\cdot}3° \pm 2°$ ($c = 0{\cdot}922$ in chloroform). Found: C, 83·2; H, 10·7. $C_{37}H_{54}O_2$ (friedelin enol-benzoate) requires: C, 83·7; H, 10·3%.

Preparation of friedelan-3 α-ol[6].—A boiling solution of friedelin (0·3 g.) in *n*-amyl alcohol (30 ml.) was treated with sodium (0·6 g.) over a period of 15 minutes. Refluxing was continued until all the sodium had dissolved and the solvent was removed by distillation with steam. The resulting suspension was filtered, the precipitate washed, dried and crystallized from benzene-ethyl acetate, when colourless plates, m.p. 298–300°, were obtained. $[\alpha]_D^{28} = +17{\cdot}7° \pm 2°$ ($c = 1{\cdot}262$ in chloroform). Found: C, 84·7; H, 12·8. $C_{30}H_{52}O$ (epifriedelanol) requires: C, 84·1; H, 12·2%.

Epifriedelanol (*friedelan-3 β-ol*).—Colourless plates from chloroform, m.p. 280–82°. In the Salkowski reaction it gave a yellow colour gradually changing to orange-red. A red colour changing to deep pink was obtained in the Liebermann-Burchard reaction. No colour was obtained with tetranitromethane reagent. $[\alpha]_D^{28} = +25{\cdot}2° \pm 2°$ ($c = 0{\cdot}862$ in chloroform). Found: C, 84·5; H, 12·7. $C_{30}H_{52}O$ (epifriedelanol) requires: C, 84·1; H, 12·2%.

The acetate, prepared with pyridine and acetic anhydride, crystallized from benzene-acetone as colourless plates, m.p. 282–84°. $[\alpha]_D^{28} = +29{\cdot}3° \pm 3°$ ($c = 0{\cdot}724$ in chloroform). Found: C, 82·2; H, 11·8. $C_{32}H_{54}O_2$ (epifriedelanol acetate) requires: C, 81·6; H, 11·6%.

Preparation of friedelin from epifriedelanol[9].—Epifriedelanol (isolated from the leaves) was oxidised by reacting with chromic acid (just over one oxygen equivalent) in glacial acetic acid at 60–70° for half an hour. The product, on repeated crystallization from chloroform-benzene, yielded long colourless needles, m.p. 256–59°. $[\alpha]_D^{29} = -20{\cdot}8° \pm 2°$ ($c = 0{\cdot}983$ in chloroform). Found: C, 84·9; H, 11·9. $C_{30}H_{50}O$ (friedelin) requires: C, 84·5; H, 11·8%. The mixed m.p. with friedelin (isolated from the leaves) was undepressed.

Chloroform extract.—The extract was evaporated and the resulting dark-green semi-solid residue (*ca.* 150 g.) was extracted with hot acetone (750 ml.) when the wax and colouring matter went into solution. The insoluble material was filtered, washed, dissolved in excess of boiling alcohol (1·5 l.) (charcoal) and filtered. On leaving overnight the filtrate deposited a small quantity of wax which was filtered and rejected. The filtrate, on leaving aside for a number of days, slowly deposited a mass of colourless needles. The mother liquor, on further concentration, yielded two more crops of the same material (total yield: 18·1 g.). Crystallization from alcohol yielded

colourless needles, m.p. 282–83° (ursolic acid, 15·5 g.). $[\alpha]_D^{28} = +64·7° \pm 2°$ ($c = 1·12$ in absolute alcohol). Found: C, 78·7; H, 10·9. $C_{30}H_{48}O_3$ (ursolic acid) requires: C, 78·9; H, 10·6%. Colour in Liebermann-Burchard test: red-violet-blue-green. The acetate crystallized from alcohol as colourless needles, m.p. 281–82°. $[\alpha]_D^{28} = +68·8° \pm 3°$ ($c = 0·936$ in chloroform). Found: C, 76·7; H, 10·7. $C_{32}H_{50}O_4$ (ursolic acid acetate) requires: C, 77·1; H, 10·1%. Mixed melting points of the acid and its acetate with the respective authentic samples[2] were undepressed.

Alcohol extract.—The united dark red solution was concentrated under reduced pressure to 1 l. with occasional addition of water and left in the ice-chest for a few days, when a large amount of resin separated. The clear reddish-brown supernatant liquid was decanted, extracted with ether containing 3% alcohol (10 × 200 ml.) and the aqueous layer rejected. The united yellow ethereal extract was evaporated to a syrupy residue (5 g.) which was dried in a desiccator and treated with warm acetone (50 ml.), when most of it went into solution leaving behind a small quantity of yellow powder. Crystallization from absolute alcohol gave pale yellow needles (10 mg.). The substance did not melt below 350° and gave a negative reaction for anthoxanthins. It gave a yellow solution with aqueous sodium carbonate and a blue colour with alcoholic ferric chloride.

The residue obtained by evaporation of the acetone solution described above could not be crystallized. It was hydrolysed by boiling with 7% alcoholic sulphuric acid. The aglycone which was extracted with ether, on repeated crystallization from dilute alcohol and dilute acetone, yielded yellow needles, m.p. 304–07° (decomp.) (quercetin, 0·2 g.). It answered the colour reactions described for quercetin in the literature. Found: C, 59·9; H, 3·8. $C_{15}H_{10}O_7$ (quercetin) requires: C, 59·6; H, 3·3%. The acetate crystallized from alcohol as colourless needles, m.p. 196–98°. Found: C, 58·9; H, 4·3. $C_{25}H_{20}O_{12}$ (quercetin penta-acetate) requires: C, 58·6; H, 3·9%. Mixed m.p. with an authentic sample of penta-acetyl quercetin[1] was undepressed.

Summary

The examination of the leaves of *Rhododendron cinnamomeum* is described. The petroleum ether extract yielded epifriedelanol, friedelin and a minor component while the chloroform extract yielded ursolic acid. Quercetin and a minor component were obtained from the alcoholic extract.

Acknowledgments

We thank Dr. V. S. Rao, Conservator-General of Forests, West Bengal, and Mr. D. K. Dey, Divisional Forest Officer, Darjeeling Division, West

Bengal, for supplying the material used in this investigation along with its botanical identity. We thank Mr. R. V. Krishna Rao for the microanalyses.

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