117. *Chemistry of the* Coprosma *Genus. Part II. The Colouring Matters from* Coprosma areolata.

By Lindsay H. Briggs, (Miss) Margot R. Craw, and Jack C. Dacre.

The bark of *Coprosma areolata* is shown to contain the phenomenally high yield of 23% of two anthraquinone derivatives, rubiadin-1 methyl ether and a new compound, *areolatin*, $C_{15}H_{10}O_6$, which is probably 1 : 5 : 6 : 7-tetrahydroxy-2-methylanthraquinone.

Coprosma areolata is a shrub or small tree 6—15 feet high, endemic to New Zealand and occurring commonly in lowland forests throughout the North and the South Island. A preliminary investigation was made by Aston (*New Zealand J. Sci. Tech.*, 1918, **1**, 264, 346) who observed that the deep brown inner bark gave a violet coloration with sodium hydroxide solution and dyed wool either as a substantive dye or with mordants. No attempt was made to isolate the dye.

An exhaustive acetone extract of the air-dried bark when subjected to chromatographic adsorption on a magnesium oxide column produced a bluish-black lake and a light-orange band readily eluted with acetone. Removal of the solvent used to elute this band furnished rubiadin-1 methyl ether, characterised by its melting point, colour reactions, and preparations of its acetate, benzoate and methyl ether. This compound has already been isolated from *C. australis* (Part I, previous paper) and *C. rubra* (forthcoming publication).

Acidification of the lake produced a brownish-red water-insoluble product crystallising from glacial acetic acid in needles or plates which shrink at 298° without melting and gradually carbonise above this temperature. Analyses agreed with the formula $C_{15}H_{10}O_6$, and the colour reactions and its derivatives indicated that it is a new compound for which the name *areolatin* is proposed.

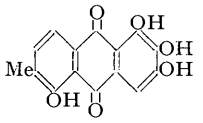
The two compounds can be obtained in quantity by extraction with acetone, removal of solvent, and treatment with calcium or barium hydroxide solution, rubiadin-1 methyl ether forming a red soluble calcium or barium salt, and areolatin, blackish insoluble salts from which the colouring matters can be recovered by acidification.

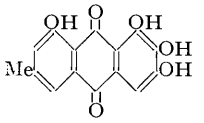
By using the chromatographic procedure, the same two colouring matters were separated from the root-bark but in considerably less yield. In both cases the compounds are present in approximately equal amount.

Areolatin gives colour reactions typical of anthraquinones, and its reduction product with hydriodic acid the reactions of an anthranol. It forms a tetra-acetate and a tetrabenzoate, and must therefore contain four hydroxyl groups. Since it forms a tetramethyl ether, all four groups must be phenolic. It furnishes one molecule of acetic acid in the Kuhn–Roth oxidation with chromic acid, and the methyl group thus indicated is in β-position, since zinc dust distillation produced β-methylanthracene. At least one phenolic group is adjacent to the methyl group, since the tetra-acetate could not be converted into the corresponding carboxylic acid by oxidation with chromic acid (cf. Anslow and Raistrick, *Biochem. J.*, 1940, **34**, 1124). Areolatin

is a strong mordant dye, indicative of phenolic groups in 1 : 2-position, while 1 : 4-groups are absent as shown by lack of fluorescence in acetic acid solution (Raistrick, Robinson, and Todd, *ibid.*, 1934, **28**, 559). The absorption spectra in alcoholic and concentrated sulphuric acid solutions indicated a relationship to both morindone and anthragallol, while the colours produced on mordanted wool were almost indistinguishable from those of anthragallol. The presence of two phenolic groups in β-positions was indicated by the isolation of a dimethyl ether as a by-product in the methylation to the tetramethyl ether, while the constitution of the other half of the molecule was confirmed by the isolation (in small yield) of 3-methoxy-4-methylphthalic anhydride on oxidation of the tetramethyl ether with chromic acid.

From these facts it appeared that areolatin is 1 : 5 : 6 : 7-tetrahydroxy-2-methylanthraquinone (I). A 1 : 8-structure (II) is not rigidly excluded, but is less likely on spectrochemical

(I.) 

 (II.)

as well as phytochemical evidence, since there are no records of 1 : 8-compounds in the *Rubiaceæ* although they occur in the *Rhamnaceæ* and the lower plants. A final decision will be attempted by synthesis.

This is the first record of an anthraquinone with four phenolic groups occurring in the higher plants, although two, cynodontin (Raistrick, Robinson, and Todd, *loc. cit.*) and catenarin (Anslow and Raistrick, *ibid.*, 1941, **35**, 1006), occur in species of moulds. This fact and the occurrence of rubiadin-1 methyl ether and areolatin together in the same plant are further exceptions to the empirical rules of Mitter and Biswas (*J. Indian Chem. Soc.*, 1928, **5**, 769) proposed for natural anthraquinones.

Experimental.

We are indebted to the Auckland City Council for their permission to collect the bark used in this investigation from mature trees at Smith's Bush near Auckland.

The finely broken air-dried bark was exhaustively extracted with acetone. On concentration, a completely crystalline mixture of lemon-yellow needles and dark-red plates separated. For chromatographic adsorption, however, a complete acetone solution was employed and adsorbed on a magnesium oxide column, 8 in. × ½ in. A bluish-black lake formed on the top of the column followed by a light-orange band. Development of the column with acetone failed to affect the lake but removed the lower band completely. Removal of the solvent afforded bright yellow needles, m. p. 302°, identified by m. p. and mixed m. p. with rubiadin-1 methyl ether. This was confirmed by the preparation of the acetate, m. p. 174°, methyl ether, m. p. 159°, and benzoate, m. p. 159·5° (for details see Part I, *loc. cit.*), all undepressed by authentic specimens.

The black lake was extruded, suspended in water, and decomposed with dilute hydrochloric acid. The dark-red precipitate, after crystallisation from glacial acetic acid, formed plates or needles of *areolatin* which shrank at 298° without melting and gradually carbonised above this temperature. Further recrystallisation from glacial acetic acid or alcohol did not change this characteristic shrinking point (Found : C, 62·9, 63·3; H, 3·8, 3·6. $C_{15}H_{10}O_6$ requires C, 62·9; H, 3·5%). The *C*-Me value was determined by the Kuhn–Roth method as modified by Barthel and La Forge [*Ind. Eng. Chem.* (*Anal.*), 1944, **16**, 434] (Found : *C*-Me 6·1. $C_{15}H_{10}O_6$ requires for 1 *C*-Me, 6·1%).

The two compounds could be conveniently obtained in quantity by treating the material obtained by acetone extraction with dilute ammonia solution followed by excess of calcium hydroxide solution. The black insoluble calcium salt of areolatin was washed free from the red soluble salt of rubiadin-1 methyl ether, and the free compounds were regenerated by acidification.

Areolatin is readily soluble in acetone, dioxan, and pyridine, less soluble in alcohol, glacial acetic acid, ethyl acetate, and ether, difficultly soluble in methyl alcohol, and insoluble in water, chloroform, and hydrocarbon solvents. With concentrated sulphuric acid it gives a dark reddish-purple coloration deepening to purple on addition of boric acid. Dilute sodium hydroxide solution gives a reddish solution changing quickly to reddish-brown. It is insoluble in sodium carbonate but dissolves in dilute ammonia solution to a brown solution changing quickly to greenish-blue. In concentrated nitric acid it is insoluble in the cold but dissolves on warming with a cherry-red–orange yellow–orange-brown coloration. The calcium, barium, and magnesium salts are purplish-black.

Areolatin Tetra-acetate.—Areolatin (85 mg.) was heated under reflux for 2 hours with acetic anhydride (5 c.c.) and fused sodium acetate (500 mg.). The hot solution was poured into water (100 c.c.) and the yellow precipitate repeatedly crystallised from glacial acetic acid to give long light-yellow needles of the *acetate* (yield 80 mg., 60%), m. p. 238° (Found : C, 60·5, 60·7; H, 4·0, 4·1. $C_{23}H_{18}O_{10}$ requires C, 60·8; H, 4·0%).

Areolatin Tribenzoate.—Areolatin (72 mg.), pyridine (3 c.c.), and benzoyl chloride (1 c.c.) were warmed for a few minutes and poured into ice-cold water (100 c.c.). The reddish-black sticky oil produced was separated by decantation, washed with 5% sodium carbonate solution, and dried. The *tribenzoate* then separated from glacial acetic acid in short, thick, orange-yellow crystals (yield 63 mg., 42%), m. p. 220°, unchanged on further recrystallisation (Found : C, 72·1, 71·8; H, 3·7, 3·9. $C_{36}H_{22}O_9$ requires C, 72·2; H, 3·7%).

Areolatin Tetrabenzoate.—Areolatin (140 mg.), pyridine (4 c.c.), and benzoyl chloride (2 c.c.) were shaken and allowed to stand overnight. Then, after being warmed for 10 minutes, the mixture was poured into ice-cold water (100 c.c.). The reddish-black oil formed solidified on trituration with hot alcohol, and after repeated crystallisation from glacial acetic acid the *tetrabenzoate* formed very pale greenish-yellow diamond-shaped plates, m. p. 281° (Found: C, 73·4, H, 3·7. $C_{43}H_{26}O_{10}$ requires C, 73·5; H, 3·7%).

Methylation of Areolatin.—Areolatin (210 mg.) dissolved in dry acetone (25 c.c.), anhydrous potassium carbonate (4 g.), and methyl sulphate (2 c.c.) were heated under reflux for 1 hour. Further potassium carbonate (2 g.) and methyl sulphate (1 c.c.) were added, and heating was continued for 8 hours. The yellow acetone solution was decanted and poured into ice–water. The yellow needles of *areolatin tetramethyl ether*, which separated immediatley (yield, 240 mg., 95%), after recrystallisation from glacial acetic acid had m. p. 175° (Found: C, 66·6; H, 5·4; OMe, 34·3. $C_{19}H_{18}O_6$ requires C, 66·7; H, 5·2; 4OMe, 36·2%).

The *dimethyl ether* could be obtained as a by-product by working up the red potassium salt from the above experiment, but was preferably prepared under milder conditions. Areolatin (250 mg.) dissolved in dry acetone (30 c.c.), anhydrous potassium carbonate (4 g.), and methyl sulphate (2 c.c.) were heated under reflux for ½ hour. The dark red salt was separated from the acetone, suspended in water, and acidified with dilute hydrochloric acid. The orange-yellow precipitate after repeated crystallisation from glacial acetic acid (yield 100 mg., 40%) formed plates, m. p. 220° (Found: C, 65·2; H, 4·4; OMe, 18·2. $C_{17}H_{14}O_6$ requires C, 65·0; H, 4·4; 2OMe, 19·7%). The dimethyl ether failed to dye wool mordanted with potash alum or stannous chloride.

Reduction of Areolatin.—A 1 : 20 mixture of areolatin (200 mg.) and zinc dust was heated in a stream of hydrogen. The bright yellow product (yield 20 mg., 15%) which sublimed in the cooler part of the tube, was repeatedly recrystallised from alcohol, and then formed almost colourless leaflets or plates, m. p. 203°, undepressed by pure β-methylanthracene, for an authentic sample of which we are indebted to Dr. Siegfried, Zofingen.

Areolatin (464 mg.) dissolved in glacial acetic acid (10 c.c.), hydriodic acid (2·5 c.c., *d* 1·35), and red phosphorus (500 mg.) were heated under reflux for 4 hours. On cooling, the mixture was poured into water (200 c.c.), giving a greenish-black precipitate and a reddish-brown solution which was decolorised by the addition of a slight excess of sodium hydrogen sulphite solution. A portion of the solid material on sublimation at 180°/0·005 mm. formed yellow plates, m. p. 229°, which were insufficient for further characterisation but gave the characteristic tests for an anthranol.

Oxidation of Areolatin Tetramethyl Ether.—A solution of the tetramethyl ether (2·5 g.) in acetic anhydride (25 c.c.) and glacial acetic acid (15 c.c.) was heated to 100° and vigorously stirred during ½ hour while chromic acid (6 g.) dissolved in glacial acetic acid (25 c.c.) and water (4 c.c.) was added. The solution was then reduced to *ca.* 10 c.c. under reduced pressure and extracted with ether (8 × 25 c.c.) which on evaporation yielded long white needles (20 mg.). These crystallised in the same form from toluene, m. p. 134—135°, and had the same m. p. and solubilities as those recorded for 3-methoxy-4-methylphthalic anhydride (Simonsen and Rau, *J.*, 1921, 1345). The analysis, although not good, did not agree with any other possible structure derivable from a tetramethoxymethylanthraquinone (Found: C, 63·1; H, 5·4. Calc. for $C_{10}H_8O_4$: C, 62·5; H, 4·2%).

Absorption Spectra.—The absorption spectra of the undermentioned compounds were measured in the visible region using a Coleman model II Universal Spectrophotometer. We are indebted to Dr. W. Gilmour of the Auckland Public Hospital for permission to use this instrument and to Mr. J. B. Brown for assistance in the measurements. The analytically pure compounds were measured in alcohol, concentrated sulphuric acid, and, where possible, N/20-sodium hydroxide solution.

	Alcohol.		Sulphuric acid.		N/20-Sodium hydroxide.	
	log ε.	λ, A.	log ε.	λ, A.	log ε.	λ, A.
Areolatin	4·438	4375	4·751	5480	—	—
	3·842	5610	4·026	7020	—	—
Morindone	4·425	4540	4·711	5610	4·508	5450
	4·218	5510	—	—	—	—
Anthragallol	4·258	4210	4·481	4780	—	—
	—	—	3·606	7040	—	—
Morindin	4·171	4425	4·373	5620	4·158	5020
β-Morindin	4·208	4430	—	—	4·193	5010
Rubiadin-1 methyl ether	3·616	3980	4·452	4560	4·175	4710
	2·820	5050	—	—	—	—
Rubiadin dimethyl ether	—	—	4·432	4590	—	—

Dyeing Properties of Areolatin.—The colours produced by areolatin on wools mordanted with the following compounds are practically indistinguishable from those of anthragallol. Iron, uranium, blackish-brown; tin, reddish brown; bismuth, molybdenum, rich brown; chromium, titanium, brown; strontium, cadmium, brownish-yellow; boron, nickel, zinc, manganese, tungsten, chrome alum, dull yellow; zirconium, dull yellowish-brown; lead, copper, dark fawn; aluminium, light fawn.

The analyses are by J. Mills, University of Adelaide, Drs. Weiler and Strauss, Oxford, and R. N. Seelye of this Department.

We are indebted to the Chemical Society, the Department of Scientific and Industrial Research, New Zealand, and the Royal Society of New Zealand for grants, and to Imperial Chemical Industries Ltd. for gifts of chemicals.

AUCKLAND UNIVERSITY COLLEGE, NEW ZEALAND. [*Received, May 14th,* 1947.]