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RESEARCH NOTE

Chemistry of *Heterodermia microphylla*, a lichen new to Sri Lanka

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Lichens are biologically distinct symbiotic entities composed of an algal or cyanobacterial photobiont and a fungal mycobiont[1]. Lichens synthesize a great variety of metabolites with different structures and potential biological activities[2,3]. In our continuing research on the chemistry of tropical lichens, we have investigated a foliose type lichen belonging to the family Physiaceae, *Heterodermia microphylla* (Kurok.) Skorepa (Figure 1), a lichen which has not been reported from Sri Lanka prior to this study. The thallus is appressed and marginally ascendant; its upper surface, and sometimes both surfaces, may contain a cortex[4]. The lower surface is corticated with rhizinae which are found only along the margin[5]. The photobiont of *Heterodermia* is a green alga, which is a chlorococcoid. The thallus of *H. microphylla* has lobes slightly disjunct or adjunct, more or less plane, not ascending, with short lateral lobes[6]. Underside of the thallus is non-corticulate and pale. *H. microphylla* is present in Asia and New Zealand. This lichen can be found at altitudes of 1500-2300 m growing on tree trunks in sheltered but open woodland and occasionally on rock. The genus *Heterodermia* is useful in determining the ecological continuity of forest ecosystems[7].

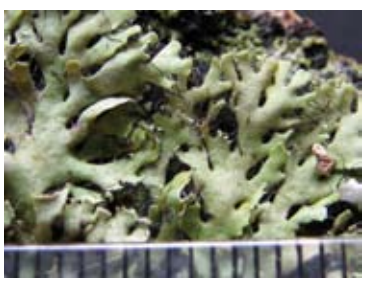

Figure 1: A part of the thallus (upper surface) of *H. microphylla*

H. microphylla growing on a rock beside a small stream was collected from Labukelle, Central Province. Dried, cleaned specimens of *H. microphylla* (26.05 g) were sequentially extracted with 2.5 L of hexane and CH_2Cl_2 (2.5 L).

The hexane extract (242 mg) when subjected to flash chromatography (eluent: 50% hexane/CH_2Cl_2) yielded the following: Methyl-5-chloro-3-formyl-2,4-dihydroxy-6-methylbenzoate **(1)** :6mg[8]); Methyl-β-orcinolcarboxylate **(2)** :8 mg; m.p. 144-143 °C, lit.[9]140-141 °C; Zeorin **(3)** :42 mg; m.p. 236-242 °C lit.[10] 240-243) °C.

The CH_2Cl_2 extract (460 mg) when subjected to Medium Pressure Liquid Chromatography (MPLC) (hexane to CH_2Cl_2) followed by flash chromatography (eluent: 1% MeOH/CH_2Cl_2) yielded: Atranorin **(4)** :43 mg; m.p. 194-196 °C, (lit m.p.[11] 196 °C); and Chloroatranorin **(5)** :6 mg; m.p. 207-210 °C (lit. m.p.[12] 208-208.5 °C).

Upon close examination, the Nuclear Magnetic Resonance (NMR) data (^{1}H and^{13} C) of compound **(1)** were identical to methyl chlorohematommate, isolated from the lichen *Lethariella canariensis*, its only other reported source in nature[8]. The hexane extract also yielded methyl-β-orcinolcarboxylate **(2)** and the triterpenoid zeorin **(3).** In addition to chloroatranorin **(5)**, the CH_2Cl_2 extract yielded atranorin **(4)**. The latter

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exhibited moderate larvicidal activity against the 2nd instar larvae of *Aedes aegypti* (90 and 100 % mortality at 100 ppm after 24 and 48 h, respectively). [13]

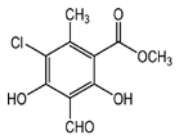
The Thin Layer Chromatography (TLC) bioassay [14] indicated the antifungal activity of compound **(4)** against *Colletotrichum gloeosporioides*. A quantitative spore germination assay[15] corroborated the above result (Table 1) indicating that atranorin **(4)** was comparable in activity to the standard benlate (methyl-1-(butylcarbonyl)-2-benzamidizolecarbamate) against *C. musae*.

From a chemotaxonomic point of view, this is the first report of the occurrence of compound **1** in the genus *Heterodermia*. and the family Physiaceae.

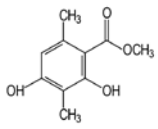
Table 1: Spore germination results for atranorin

Fungus	Compound	% Germination
C. gloeosporioides	**4**[a]	58.2 ± 3.3
	control[b]	74.2 ± 1.4
	benlate[a]	33.3 ± 1.1
C. musae	**4**	26.2 ± 2.3
	control	66.8 ± 1.1
	benlate	32.4 ± 3.3

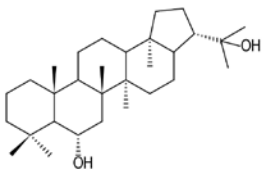
[a] Concentration of both compound **4** and benlate was10 ppm
[b] Negative control with no compound



(1)



(2)



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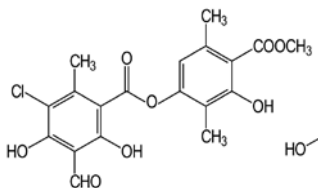
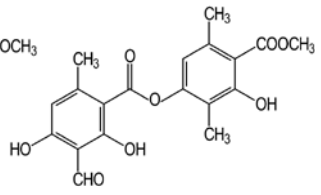


(4)



(5)

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