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Studies on Chilean Lichens. XXI. Secondary metabolites from the Antarctic species *Hypogymnia lugubris*

MARISA PIOVANO[1], WANDA QUILHOT [2], MARÍA CHAMY[1], PETER FIEDLER[1]
and JUAN A. GARBARINO[2]

SUMMARY

From Hypogymnia lugubris *(Pers.) Krog, a lichen collected in Antarctica, atranorin, ergosterol peroxide, and physodic, physodalic, protocetraric and usnic acids were isolated.*

Key word: Antarctic lichen, secondary metabolites, depsides, depsidones, usnic acid, steroids.

Estudios sobre líquenes chilenos. XXI. Metabolitos secundarios de la especie antártica *Hypogymnia lugubris*

MARISA PIOVANO[1], WANDA QUILHOT [2], MARÍA CHAMY[1], PETER FIEDLER[1]
y JUAN A. GARBARINO[1]

RESUMEN

De Hypogymnia lugubris *(Pers.) Krog, un líquen recolectado en la Antártica, se aisló atranorina, peroxiergosterol y los ácidos fisódico, fisodálico, protocetrárico y úsnico.*

Palabras claves: líquen antártico, metabolitos secundarios, dépsidos, depsidonas, ácido úsnico, esteroles.

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[1] Departamento de Química. Facultad de Ciencia, Universidad Técnica Federico Santa María, Casilla 110-V, Valparaíso, Chile.

[2] Escuela de Química y Farmacia, Facultad de Medicina, Universidad de Valparaíso, Casilla 92-V, Valparaíso, Chile.

INTRODUCTION

The lichen forming fungi produces secondary metabolites very infrequently found in other organisms. Although most species produce a constant secondary chemistry, some are differentiated into chemotypes which may correlate with geography and, where sympathetic, with ecology (Culberson *et al.*, 1989). The chemical characteristics are also important due to their function as screening compounds and their possible action in photosynthesis (Lawrey, 1986; Quilhot *et al.*, 1992). Continuing the systematic chemical study of Antarctic lichens, this paper informs about a new chemotype from *Hypogymnia lugubris* (Pers.) Krog, collected on King George Island, South Shetland Islands, during the XXVIII Antarctic Scientific Expedition, supported by Instituto Antártico Chileno (INACH).

The genus *Hypogymnia*, included in the Hypogymniaceae family, contains about 45 species, distributed in the northern and southern hemispheres (Galloway, 1985). In numerous taxa atranorin, chloroatranorin, the physodic acid complex, physodalic, and protocetraric acids occur (Culberson, 1969, 1970; Culberson *et al.*, 1977; Galloway, 1985).

EXPERIMENTAL

General procedures: The general experimental details have been published previously (Quilhot *et al.*, 1989; Piovano et al., 1991).

Extraction of *Hypogimnia lugubris* (Pers.) Krog: The lichen material was collected on King George Island, South Shetland Islands, in January 1992. It grows on rocks and is protected by *Neuropogon* spp. *thalli*. A voucher specimen is deposited in the herbarium of Escuela de Química y Farmacia, Universidad de Valparaíso. The air-dried and ground lichen thalli (138 g) were extracted, successively with dichloromethane and acetone, each time for a period of 48 h, at room temperature. Extracts were concentrated at reduced pressure.

The dichloromethane extract was fractionated on a silica gel column by elution with naphta-ethyl acetate-HCOOH (35:15:1).

In order of elution there appeared atranorin, usnic acid, ergosterol peroxide, physodic and physodalic acids. From the acetone extract, fractionated on a silica gel column by elution with naphta-ethyl acetate-HCOOH (35:5:1), atranorin and usnic, physodic, physodalic and protocetraric acids were obtained. (See Fig. 1).

The compounds were identified by physical and spectroscopic methods and compared with authentic samples.

Atranorin, 140 mg, 0.10% (Quilhot et al., 1989).

Physodic acid (1), 3.20 g, 2.32% (Hirayama *et al.*, 1975) mp 205° d (methanol-water).

IR $^{KBr}_{max}$ cm^{-1} : 3550, 3180, 1690, 1675, 1630, 1215.

^{1}H-nmr [CD_3OD] δ : 0.87 (3H, t, J = 6, CH_3); 0.93 (3H, t, J = 6, CH_3); 1.10-2.00 (12H, m, CH_2 $(CH_2)_3$ CH_3); 2.51 (2H, t, J = 7, CH_{2b}); 3.16 (2H, m, CH_{2c}); 3.94 (2H, s, CH_{2a}); 6.49 (1H, s, Ar-H_3); 6.60 (2H, d, J = 2, Ar-$H_{3,5}$).

MS m/z: 470 (M^+, 8%), 426 (25), 425 (84), 399 (100), 371 (20), 357 (40), 355 (64), 301 (8), 299 (42), 283 (78), 247 (60), 195 (79), 194 (100).

Physodalic acid (2), 225 mg, 0.16% (Elix *et al*., 1987) mp 260° d (ethyl ether).

IR $^{KBr}_{max}$ cm^{-1}: 3250, 1740, 1685, 1640, 1620, 1540, 1340, 1250.

^{1}H-nmr [$(CD_3)_2$ CO/ $(CD_3)_2SO$] δ: 1.99 (3H, s, OA_c); 2.50 (3H, s, Ar-CH_3); 2.56 (3H, s, Ar-CH_3); 5.19 (2H, s, CH_2); 6.84 (1H, s, Ar-H); 10.65 (1H, s, CHO); 12.10 (2H, brs, OH).

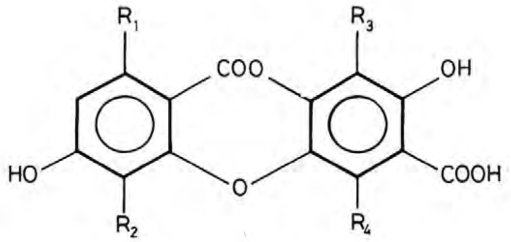
Protocetraric acid (3), 61 mg, 0,04% (Sala and Sargent, 1981), mp 245 - 250° d (acetone).

IR $^{KBr}_{max}$ cm^{-1}: 3200, 1740, 1690, 1640, 1250.

^{1}H-nmr [$(CD_3)_2$ CO] δ: 2.51 (3H, s, Ar-CH_3-6); 2.70 (3H, s, Ar-CH_3-6'); 4.79 (2H, s, CH_2); 6.80 (1H, s, Ar-H); 10.79 (1H, s, CHO).

Usnic acid, 160 mg, 0.12% (Quilhot *et al.,* 1989).

Ergosterol peroxide, 25 mg, 0,02% (Quilhot *et al.,* 1989).



	R_1	R_2	R_3	R_4
[1]	$\underset{a}{C}H_2CO\underset{b}{C}H_2C_3H_7$	H	H	$\underset{c}{C}H_2C_4H_9$
[2]	CH_3	CHO	CH_2OAc	CH_3
[3]	CH_3	CHO	CH_2OH	CH_3

Fig. 1: Depsidones from *Hypogimnia lugubris*

RESULTS AND DISCUSSION

Hypogimnia lugubris accumulates atranorin (depside), physodic, physodalic, protocetraric acids (depsidones), usnic acid, and ergosterol peroxide.

For a sample collected in South Georgia the presence of atranorin has been reported, as well as of physodalic acid and a non identified compound (Lindsay, 1973).

H. lugubris from New Zealand produces atranorin, chloroatranorin and physodic, physodalic, protocetraric acids, as well as (±) - 2' -0-methylphysodic, 3-hydroxyphysodic, alectoronic and vittatolic acids (Galloway, 1985). The capacity of lichens for producing characteristic secondary metabolites permits the recognition of chemotypes which have similar morphologic features (Tabacchi, 1984). Our results make the existence of a new chemotype of *H. lugubris* evident.

Relatively few lichens have been analyzed for steroids: one reason for this is that these metabolites are present in only very small amounts in the plant material (Tabacchi *et al*., 1987). In the majority of the Antarctic species analysed ergosterol peroxide is present in amount of traces and up to concentrations fluctuating between 0.003% (Quilhot *et al*., 1989; Piovano *et al*., 1991; Garbarino *et al*., 1991). From *H. lugubris* 25 mg (0.02%) of this phytosterol was isolated.

CONCLUSION

In lichens chemical races are related with, among other factors, their geographic location, which involves changes in the microhabitat in which the species develops its life cycle.

The stressing conditions of the Antarctic environment, in particular light and temperature, might influence the synthesis of secondary products in this species.

In *H. lugubris*, as well as in other Antarctic species, atranorin and usnic acid coexist. These metabolites filter UV radiation which is harmful to the photobiont. The fluorescent emission of atranorin might be used in photosynthetic work (Quilhot *et al*., 1991), thus increasing the productivity of this species.

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