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Thin-layer chromatographic study of the phenolics of the *Pleurocladula* species (*Hepaticae*).

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Abstract

Phenolic compounds in *Pleurocladula albescens* (Hook.) Grolle and *Pleurocladula islandica* (Nees) Grolle were studied by two-dimensional thin-layer chromatography. Consistent differences between both taxa were found.

INTRODUCTION

In recent years, thin-layer chromatography has been widely used for description of the population structure (Weimarck, 1974) and elucidation of taxonomic problems. This technique is suitable to supplement the cytological and morphological studies in higher plants, e.g. in *Lotus* (Grant, Whetter, 1966), *Mentha* (Olsson, 1967), *Saxifraga* (Jaworska, Nybom, 1967), *Secale* (Dedio et al., 1969a), *Triticale* (Dedio et al., 1969b), *Prunus* (Muszyński, Nybom, 1969), *Potentilla* (Asker, Fröst, 1970 a, b), *Aegilops* (Kaltsikes, Dedio, 1970 a, b), *Hordeum* (Fröst, Holm, 1972), *Avena* (Asker Fröst, 1973), *Aconitum* (Szweykowski, Krzakowa, 1977a, b), *Dryopteris* (Widen et al., 1975), as well as in liverworts: *Pellia* (Szweykowski, Krzakowa, 1977c). However, from the biochemical points of wiew some liveworts have been investigated earlier (Markham, Porter, 1969; Markham et al., 1977 and the literature cited there; Mues, Zinsmeister, 1976).

This paper comprises a study of the variation of nonidentified phenolic compounds in both *Pleurocladula* (Grolle, 1979) species: *P. albescens* (Hook). Grolle and *P. islandica* (Nees) Grolle with the aim to find new characters discriminating these two taxa.

MATERIAL AND METHODS

For this study herbarium samples collected in 1965 and coming from natural populations growing in the Tatra Mountains were used. For the detailed list and description of these populations see Krzakowa (1971). Over 1000 dry stems of *P. islandica* and 800 stems of *P. albescens* were taken with tweezers singly out of each carpet. Together 100 mg of dry plants of each species were analysed. The procedure of phenolic compounds extraction and chromatography was the same as described earlier (Szweykowski, Krzakowa, 1977a, 1979). The similarity index S_{phi} was calculated according to the formula (Leuschner, 1974):

$$S_{phi} = \frac{(a \times d) - (b \times c)}{\sqrt{(a+b)(c+d)(a+c)(b+d)}}$$

The letters a through d denote the numer of spots present or absent in particular parts according to the diagram:

	+	—
+	a	b
—	c	d

where "+" means spot present, and "—" means spot absent.

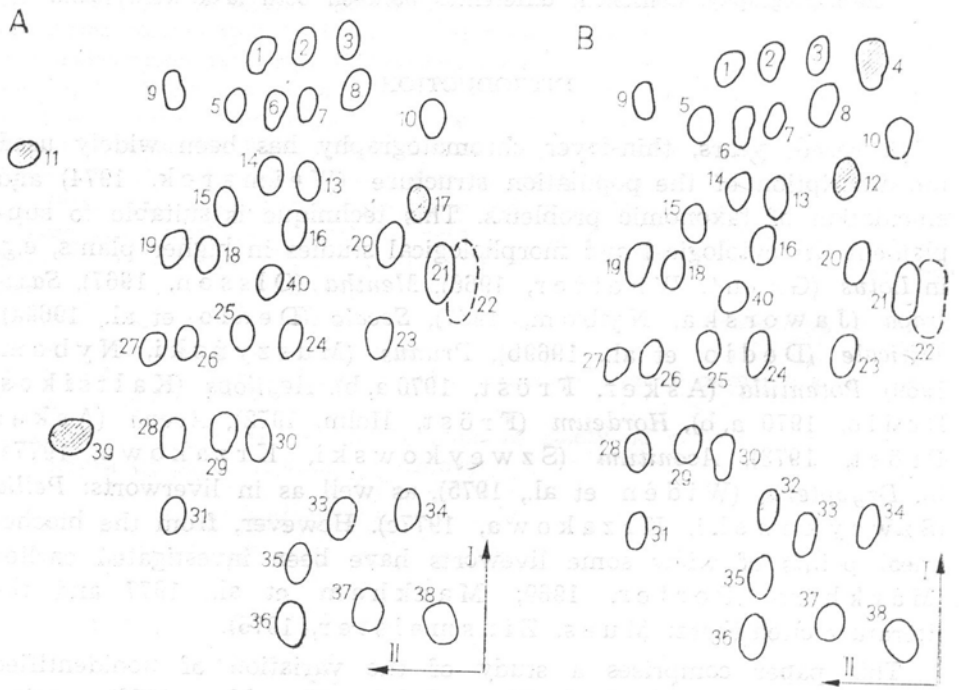

Fig. 1. Two-dimensional thin-layer chromatograms of extracts from *Pleurocladula albescens* (A) and *Pleurocladula islandica* (B) species. Characteristic spots are shaded, the rest are common to both species

On the basis of the similarity indices matrix a Wrocław diagram (=dendrite) was constructed (Florek et al., 1951; Sneath, Sokal, 1973).

Table 1

Distribution of phenolic compounds in the *Pleurocladula* species

Spot No.	Colour in UV $^{+}NH_3$	Populations													
		Pleurocladula albescens							*Pleurocladula islandica*						
		1	2	3	5	6	7	10	4	8	9	11	12	13	14
1	B	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	B	+	+	+	+	−	+	+	+	+	+	+	+	+	+
3	B	+	+	+	+	+	+	+	−	+	+	+	+	−	+
5	Y	−	+	+	+	+	+	−	+	+	+	+	−	+	+
6	B	+	+	+	+	+	+	+	+	+	+	+	+	+	+
7	B	+	−	+	+	+	+	+	+	+	−	+	+	+	+
8	B	−	+	+	−	−	+	+	−	+	+	+	+	+	+
9	B	+	+	+	−	+	+	−	+	−	−	−	−	+	−
10	Y	+	−	−	+	+	+	+	−	+	−	+	−	+	−
13	B	+	+	+	+	−	+	+	+	+	+	−	+	+	+
14	B	+	+	+	+	+	+	+	−	+	+	−	+	+	+
15	B-V	+	+	+	+	+	+	−	+	+	−	+	+	−	+
16	Y	+	+	−	+	+	+	+	+	+	+	+	+	+	+
18	C	+	−	−	+	+	+	+	−	+	+	+	+	+	+
19	C	−	+	+	−	+	+	+	+	+	−	+	−	−	−
20	C	+	+	+	+	+	+	+	+	+	+	+	+	−	+
21	Y	+	+	+	+	+	+	+	+	+	+	+	+	+	+
22	Y	−	−	−	+	−	+	+	+	+	−	+	−	+	+
23	B	+	+	+	+	+	+	+	+	+	+	+	+	+	+
24	C	−	−	−	+	−	+	−	−	+	−	+	+	−	−
25	Y	+	+	+	−	+	+	+	−	+	+	+	−	−	+
26	Y	−	−	−	−	+	+	+	−	+	−	+	+	−	+
27	Y	−	−	+	−	+	+	+	−	−	+	−	+	+	+
28	C	+	−	+	+	−	+	+	+	+	+	+	+	+	+
29	C	−	−	+	−	+	+	+	+	+	+	+	+	−	−
30	Y	−	−	−	−	−	+	+	−	−	+	+	+	−	−
31	Y	+	+	−	+	+	+	+	−	+	−	+	−	+	−
33	C	−	−	+	−	−	+	−	+	+	−	−	+	−	+
34	C	+	−	+	+	−	+	+	+	+	+	−	+	−	+
35	Y	−	−	−	−	−	+	−	+	+	+	−	+	−	−
36	B-V	+	+	+	+	+	+	+	+	+	−	−	+	+	−
37	B	−	−	+	+	+	+	+	+	+	+	+	+	+	+
38	Y	−	−	+	+	+	+	+	+	+	+	+	+	+	+
40	C	+	−	+	−	−	−	−	+	−	+	−	+	+	−
4	T	+	+	+	+	+	+	+	−	−	−	−	−	−	−
12	B	+	+	+	+	+	+	+	−	−	−	−	−	−	−
32	V	+	+	+	+	+	+	+	−	−	−	−	−	−	−
17	Y	−	−	−	−	−	−	−	+	+	+	+	+	−	+
39	B	−	−	−	−	−	−	−	+	+	+	+	+	+	+
11	V	−	−	−	−	−	−	−	−	+	−	−	+	+	−

Colour key: B — blue, C — celladon, T — turquoise, Y — yellow, *A* — violet.

RESULTS AND DISCUSSION

Forty different spots were detected in both species (Fig. 1). Thirty four of them were common to both, but each species has its own characteristic spots. The spots nos 11, 17, 39 occured in *P. albescens* only, another three spots (4, 12, 32) were found exclusively in *P. islandica* (Table 1).

The dendrite constructed on the basis of the S_{phi} similarity indices matrix (Table 2) clusters all populations. The groups are separated by the lowest taxonomic distance i.e. 1.85. The first of them (in the upper part of Fig. 2) comprises samples belonging to *P. islandica*. The second one (lower part of the same figure) is occupied by samples belonging to *P. albescens* exclusively.

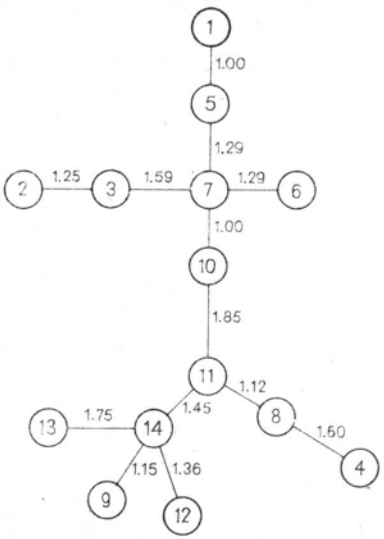

Fig. 2. Wrocław diagram showing the interpopulational variability of 14 natural populations of *Pleurocladula islandica* (upper part) and *Pleurocladula albescens* (lower part) and the taxonomic distance between them

Thus, the chromatographic patterns complete and confirm very well the morphological distinction between *Pleurocladula albescens* and *Pleurocladula islandica* described previously (K r z a k o w a, 1971).

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Table 2

Matrix of taxonomic distances, d_{ij}. Lower half $d_{ij}=1-S_{phl}$. Upper half values obtained when to the shortest distance (0.4223) was assigned arbitrarily the value 1.0

Populations														
	1	2	3	5	6	7	10	4	8	9	11	12	13	14
1		1.0613	1.5243	1	1.5065	1.8042	1.5307	2.3680	2.1137	1.3680	2.0585	2.0888	1.9732	2.2666
2	0.4482		1.2555	1.5354	1.2868	1.5382	1.7521	2.3374	2.2666	2.3374	2.3237	1.6152	2.2711	2.2811
3	0.6437	0.5302		1.8804	1.6095	1.5929	1.7713	1.7009	1.9941	1.9678	1.8241	2.3680	2.3152	2.0296
5	0.4223	0.6484	0.7941		1.5612	1.2922	1.3647	2.1757	1.8617	1.3038	1.9837	2.2247	1.7599	1.8215
6	0.6362	0.5434	0.6797	0.6593		1.2922	1.3647	2.0476	2.2209	2.0476	1.9837	1.6514	2.2666	2.3419
7	0.7619	0.6496	0.6727	0.5457	0.5457		1.0007	2.1641	2.2122	2.1641	2.0732	2.2084	2.1262	2.1198
10	0.6464	0.7399	0.7480	0.5763	0.5763	0.4226		1.8738	2.2780	2.0149	1.8572	2.2100	1.8840	1.9380
4	1	0.9871	0.7183	0.9188	0.8647	0.9139	0.7913		1.6036	1.7682	2.0744	1.7327	1.8688	1.6630
8	0.8926	0.9572	0.8421	0.7862	0.9379	0.9342	0.9620	0.6772		1.9254	1.1288	1.5584	1.9863	1.2084
9	1	0.9871	0.8310	0.9729	0.8647	0.9139	0.8509	0.7467	0.8131		1.8132	1.1679	1.8688	1.1501
11	0.8693	0.9813	0.7703	0.8377	0.8377	0.8755	0.7843	0.8760	0.4767	0.7657		2.1489	2.1615	1.4537
12	0.8821	0.6821	1	0.9395	0.6974	0.9326	0.9333	0.7317	0.6581	0.4932	0.9075		2.0888	1.3647
13	0.8333	0.9591	0.9777	0.7432	0.9572	0.8979	0.7956	0.7892	0.8388	0.7892	0.9128	0.8821		1.7599
14	0.9572	0.9633	0.8571	0.7692	0.9890	0.8952	0.8184	0.7023	0.5103	0.4857	0.6139	0.5763	0.7432	

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Zastosowanie chromatografii cienkowarstwowej związków fenolowych do badań zmienności dwóch gatunków rodzaju Pleurocladula (Hepaticae)

Streszczenie

Badania wykazały, że *Pleurocladula albescens* (Hook.) Grolle i *Pleurocladula islandica* (Nees) Grolle charakteryzują się obecnością 40 plam fenolowych, z których 34 występuje u obu gatunków. Każdy z nich charakteryzuje się oprócz tego obecnością trzech plam, które u drugiego nie występują. Wyniki analiz przedstawiono za pomocą dendrytu opartego na uporządkowanej macierzy współczynnika podobieństwa S_{phi}. Jak widać na rysunku 2, populacje utworzyły dwa skupienia, a hiatus między gatunkami jest wyraźny.