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PHENOTYPIC VARIABILITY OF LEAFLETS  
IN *THALICTRUM MINUS* - COMPLEX  
(*RANUNCULACEAE*)

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The variability of 8 characters of leaflets in the *Thalictrum minus* - complex from Yugoslavia is shown with special regard to *Thalictrum velebiticum* Deg. 1937, and to their possible distinction by use. Data have been processed by classical statistics and a comparison of variation coefficients for each sample and character has been made. These characters show significant variability, a continuity of connection between their extreme values and a low diagnostic value due to considerable overlapping between the samples. The sample representing *T. velebiticum* shows certain populational characteristics.

Introduction

A series of characteristics is related to the genus *Thalictrum*: the phenomenon of parthenogenesis (Winkler 1920), the successive passage from entomophily to anemophily (Daumann 1969, Damboldt and Zimmermann 1974), the high contents of specific alkaloids (Raffauf 1977, Wu Wu Nan et al. 1980, and others), many polyploidic forms with a large number of multiplications of the basic chromosome number ( $x=7$ ) even up to 22 (Bolkhovskikh et al. 1969, Kuzmanov 1986, Kuzmanov et al. 1987), the presence of apomixis and the existence of particular populations in a clonic form, hybridization (Tutin 1964, Damboldt and Zimmermann 1974), large areas of most European genus species (Meusel 1964) and important phenotypic variability.

None of the numerous classifications of this genus in to sections, series, etc. (Jančhen 1949, Ly Thi Ba 1981, and others) has been generally accepted, and the number of species is in fact a matter of guesswork, the nomenclature being accompanied by constant classification difficulties, in this case especially due to the large synonymy. For Yugoslavia, the most optimistic number of species is 17, and the most reduced list contains only 7 of them while many previous forms are treated as intraspecies categories. However, the distinction of one group of plants such as *Thalictrum minus*-complex (Damboldt and Zimmermann 1974), the size and the extent of which are explained differently, is quite usual. The aim of the present study was to define variability limits for some characters related to the morphological characteristics of leaflets (foliolae), to determine differences in the variability level between particular characters and to compare the species *T. velebiticum* (Degen 1937) gathered on locus classicus at Velika and Mala Paklenica with the remaining part of widely spread *T. minus*-complex on the territory of Yugoslavia. As a matter of fact, the species *T. velebiticum* if it really exists, represents an endemic, but it is seldom mentioned in floras as an independent species (Degen 1937, Trinajstić 1973), and when mentioned it is given an inferior taxonomic rank (Hayek 1927, Soó 1966).

### Material and Methods

The material was descended from collections of Department of Botany (HbZ), I. Trinajstić (Hb Trin.) and my personal, enriched by specimens from Velika and Mala Paklenica (specimens no. 53—103). Material was determined and classified in two groups: *Thalictrum minus* — complex and *T. velebiticum*. It's important to mention that same specimens can not be easily classify in this manner (Damboldt et Zimmermann 1974, Tutin 1964), and they bring some error in samples treatment. The material was collected mostly in NW Croatia: Velebit — Oštarije, Šugarska Duliba (specimens no. 39—48), Senjsko bilo (specimens no. 1, 9, 10), Bakar, Antovo, Ladvić specimens no. 19, 23, 36, 37, 38, 45), Gorski Kotar (specimens no. 3, 15, 26, 31), Plitvice lakes (specimens no. 12, 49), Plješevica (specimens no. 5, 8), Lika and Dinara (specimens no. 4, 6, 29, 30, 32, 33, 34). Same specimens was collected in Oštrc (no. 13, 20, 24) and Radoboj (no. 25) and some in Slovenija — Soča spring (no. 18), Dvor (no. 51, 52) and Crna Gora — Biogora lakes (no. 17).

All measurements were made on 48 specimens of *T. minus*-complex (sample 1) and 50 specimens from Paklenica (sample 2) (Fig. 1). Both samples were also treated as one of 98 specimens (sample 3), i. e. as part of the same complex.

The general morphological characteristics of leaflets were divided in several more precisely defined characters:

- 1 — the minimum width of leaflets (min W in mm)
- 2 — the maximum width of leaflets (max W in mm)
- 3 — the minimum length of leaflets (min L in mm)
- 4 — the maximum length of leaflets (max L in mm)

Each of these characters relating to the dimensions of the leaflets was registered for each specimen separately, thus defining the limits for the appearance of all leaflets on a single individual regardless of their distribution.

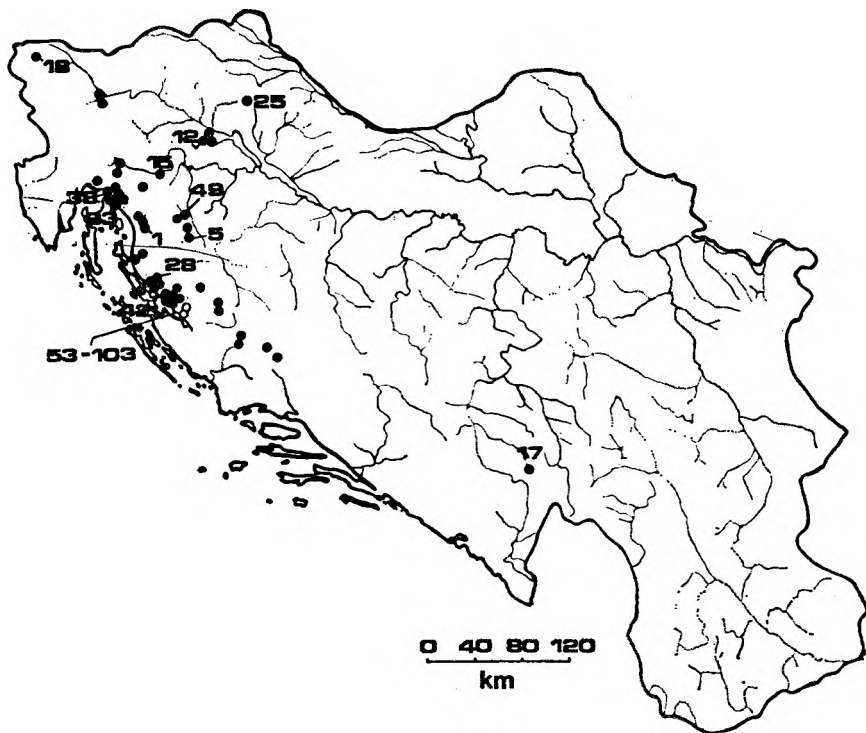


Fig. 1. Locations: • sample 1; ○ sample 2; Ordinal numbers of locations are accidental.

5 — the minimum number of leaflet tips (min no. T)

6 — the maximum number of leaflet tips (max no. T)

These characters define the range of the total number of leaflet tips for each individual regardless of their distribution.

7 — the ratio max L / maximum leaflet incision (max I).

The max I was measured (in mm) on each individual and it represents the deepest incision found in a particular pattern. This character is as relative indicator in relation to the max L.

8 — the shape of the base of leaflets is the character presented in of 3 possible states:

a) state (1) ... a wedge-shaped, cuneiform, leaflet base

b) state (0) ... a heart-shaped, cordate, leaflet base

c) state (0.5) ... an ellipsoidal ovate, leaflet base, the category comprising the transitional shapes between 1 and 0.

Statistical data processing comprised all characters except the characters number 8 and calculated:  $\bar{x}$  = the arithmetic mean, S = standard deviation, V = coefficient of variation,  $S\bar{x}$  = error  $\bar{x}$ , E = error  $\bar{x}$  in

%,  $S_x$  = error S,  $S_v$  = error V (Sokal and Rohlf 1981) critical ratios  $t_{\bar{x}}$ ,  $t_x$  and  $t_v$  for  $\bar{x}$ , S and V (with  $P = 0.01$ ) (Marinković et al. 1982) as well as  $t$  — test (with  $P = 0.01$ ) and  $F$  — test (with significance level of 5%) for each sample (Petz 1985).

## Results and Discussion

The results of statistical data processing are shown in Table 1. The calculated critical ratios  $t_{\bar{x}}$ ,  $t_s$  and  $t_v$  compared with the values in the table indicate that the data for samples are good representatives of populations.

### Characters 1, 2, 3 and 4

The use of leaflets dimensions for the purpose of diagnosis, i.e. as key parts for the determination of particular shapes and for a better description of the taxon is relatively frequent. Thus, for example, to distinguish *T. minus* L. var. *majus* Cr. and var. *minus* the leaflets of 20 — 30 mm and less than 20 mm are indicated (Gajić 1970), to distinguish many varieties and forms of *T. minus* L. ssp. *minus*: leaflets 8—12 mm long for f. *roridum* (Wallr.) Koch. are indicated, for f. *glaucomedium* Nyár. leaflets 15—20 mm long, 4—8 mm long for f. *microphyllum* (Borb.) Soó, 8—12 mm long for var. *minus* Sch. et K., 4—8 mm long for f. *minutifolium* (Borb.) Soó, etc (Soó 1966), for ssp. *minus* leaflets 5—15 mm long, and for ssp. *majus* 10—30 mm long (Damboldt and Zimmermann 1974, Tutin 1966) and similarly for many other taxa of various ranks of the complex (Hayek 1927, Nyárády 1953, Panov 1970, Oberdorfer 1979).

A statistically significant difference exist between  $\bar{x}_1$  and  $\bar{x}_2$  with regard to character 1 and statistically significant is also the difference between  $S_1$  and  $S_2$ , showing that this differences are not the result of variations between the samples but that very likely they exist between real populations too, i.e. the narrowest leaflets of the population from Paklenica on the average are wider than the narrowest leaflets of the specimens of *T. minus*-complex. The statistically significant differences between samples 1 and 2 were not found for other characters relating to the dimensions of the leaflets (characters 2, 3, 4). For illustration in fig. 2 the selected 13 patterns are presented (out of 98) by a rectangle with dimensions defined by min L and max L on y — axis and by min W and max W on x — axis. Inside this rectangle there are all dimensions of the leaflets available on the respective individual with unknown distribution inside it.

Between the markedly small — leaf shape (specimen no. 76) and markedly large — shape (specimen no. 5) there is a number of intermediate dimensions of the leaflets. By increasing the number of specimens the clarity of the overview is lost and consequently the continuity of connection grows. The distribution between the limit values (Fig. 2, A), regarding all data, shows right assymetry with significantly fewer large — leaflet specimens in the sample.

It is evident that any grouping of the plants inside a particular sample into taxonomically separate groups according to these characters is unacceptable and that it would inevitably lead to errors in determination.

Table 1. Results of the statistical data processing

Character Parameter	1	2	3	4	5	6	7
Sample 1 N = 48							
$\bar{x}_1$	3.02	13.20	5.08	13.79	1.47	5.72	3.10
min	1	6	2	6	1	3	1.83
max	7	32	13	33	3	9	5
$S_1$	1.21	5.35	2.48	5.74	0.54	1.55	0.83
$V_1$	40.06	40.53	48.82	41.62	36.73	27.09	26.77
$S\bar{x}_1$	0.17	0.77	0.35	0.82	0.08	0.22	0.12
$Ss_1$	0.12	0.54	0.25	0.58	0.05	0.16	0.08
$Sv_1$	4.09	4.14	4.98	4.25	3.75	2.76	2.73
$E_1$	5.78	5.85	7.04	6.04	5.32	3.91	3.36
$t\bar{x}_1$	17.27	17.08	14.18	16.62	18.80	25.54	25.85
$ts_1$	9.76	9.97	9.98	9.81	9.73	9.77	9.78
$tv_1$	9.79	9.79	9.79	9.79	9.79	9.79	9.79
Sample 2 N = 50							
$\bar{x}_2$	4.99	14.48	6.20	13.60	2.42	7.14	3.21
min.	2	6	3	6	1	3	1.6
max	11	31	10	27	4	12	5.6
$S_2$	1.96	4.82	2.03	4.22	0.73	2.23	1.01
$V_2$	39.28	33.29	32.74	31.03	30.16	31.23	31.42
$S\bar{x}_2$	0.27	0.68	0.28	0.59	0.10	0.31	0.14
$Ss_2$	0.19	0.48	0.20	0.42	0.07	0.22	0.10
$Sv_2$	3.93	3.33	3.27	3.10	3.02	3.12	3.14
$E_2$	5.57	4.71	4.63	4.39	4.27	4.42	4.44
$t\bar{x}_2$	17.95	21.22	21.59	22.74	23.41	22.62	22.50
$ts_2$	9.97	10.00	10.00	9.96	10.01	10.00	10.00
$tv_2$	10.00	9.99	9.99	10.00	9.99	9.99	10.00
t — test	6.00	1.23	2.43	0.18	7.24	3.64	0.60
F — test	2.63	1.23	1.49	1.84	1.79	2.06	1.47
Sample 3 N = 98							
$\bar{x}_3$	4.02	13.85	5.65	13.69	1.96	6.44	3.16
min	1	6	2	6	1	3	1.6
max	11	32	13	33	4	12	5.6
$S_3$	1.90	5.10	2.32	5.00	0.79	2.04	0.92
$V_3$	47.26	36.82	41.06	36.52	40.31	31.68	29.11
$S\bar{x}_3$	0.19	0.52	0.23	0.50	0.08	0.20	0.09
$Ss_3$	0.13	0.36	0.16	0.35	0.07	0.14	0.06
$Sv_3$	3.38	2.63	2.93	2.61	2.88	2.26	2.08
$E_3$	4.78	3.72	4.14	3.69	4.09	3.20	2.96
$t\bar{x}_3$	20.88	26.87	24.12	27.09	24.40	31.19	33.78
$ts_3$	14.03	13.98	13.97	14.01	14.01	14.01	14.02
$tv_3$	14.00	13.99	13.99	14.00	13.99	14.00	14.00

The method of describing the intraspecies forms by nomination of forms, varieties and the like such as *macrophyllum*, *minutifolium*, *emicrophyllum* etc. does not seem flexible enough. The population from Paklenica, as the representative of the species *T. velebeticum* does not show the diagnostically relevant differences.

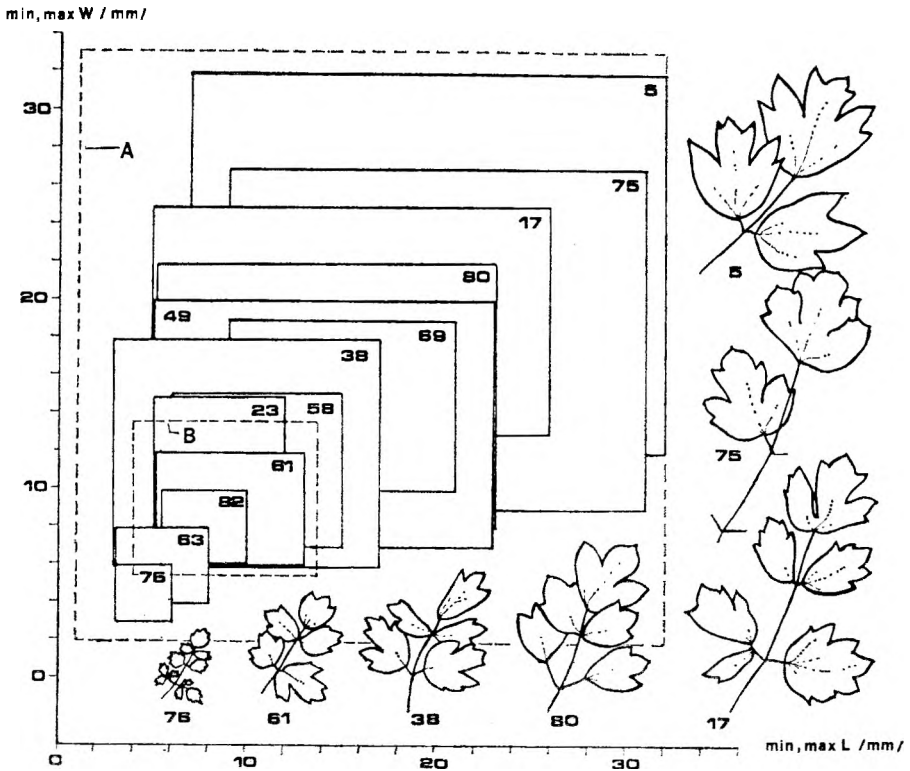


Fig. 2. Variability of dimensions for leaflets in part of the sample; the patterns marked by ordinal number; A — limits values; B — arithmetical means for the sample 3.

Characters 5 and 6

The number of leaflet tips is indicated as the characteristics for particular taxa: for *T. minus* L. three — raglike leaflets in front, for *T. elatum* Jacq. 3 — 5 times raglike (Hayek 1927), 3 — 5 blades for particular shapes of *T. minus* ssp. *minus* (Soó 1966) and the like (Trinajstić 1973, Gajić 1970).

The divergence between  $\bar{x}_1$  and  $\bar{x}_2$  is statistically significant for min T and max T too, the divergence between  $S_1$  and  $S_2$  being statistically significant for the max T only. The total individual variability (Fig. 3) ranges from 2 — 3 to 3 — 12 tips on the leaflets. The number of tips in the sample grows continuously without two or more gap separated categories.

As can be seen a clear distinction of the population from Paklenica is impossible, the overlapping being important, it is evident that the plants from Paklenica tend to wards a wider total range of tip number for a particular individual and to a higher top bottom limit for the min T and max T (Tab. 1, t — test).

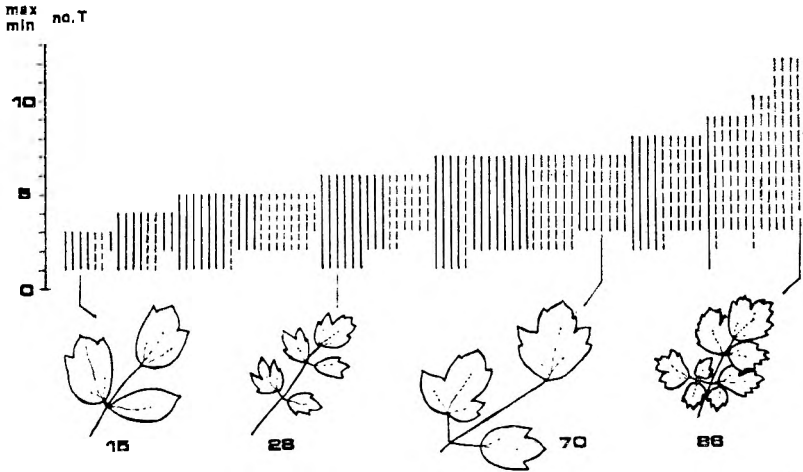


Fig. 3. Individual variability of number leaflets tips expressed by min — max value for particular pattern; full line — sample 1; dotted line — sample 2.

### Character 7

The incision depth of the leaflets greatly affects the general morphological appearance of the leaflets and, although not frequently, some shapes were given more or less separate taxonomic status based on it: *T. minus* ssp. *minus* f. *multipartitum* Nyár., f. *subintegrum* Nyár., f. *palmatifidum* (Borb.) Soó (Soó 1966). Most floras, however, do not mention the incision of leaflets as a separate character (Tutin 1964, Damboldt and Zimmermann 1974, Hayek 1927, and others).

The F-test and t-test do not show any statistically significant differences either between  $\bar{X}_1$  and  $\bar{X}_2$  or between  $S_1$  and  $S_2$ . The range of the ratio max L / max I is relatively wide, 1.6 — 5.6, making the marginal patterns of the distribution very different in aspect (Fig. 4). The population from Paklenica does not show divergences, although precisely in sample 2, from the point of view of max I, the shapes with the least incised leaflets have been registered.

### Character 8

Efforts to describe the shape of the base of leaflets in floras (Hayek 1927, Degen 1937, Soó 1966, Trinajstić 1973, Martinčić and Sušnik 1974, and others) have resulted in such terms as ovate, triangulate, cuneate and similar, and for *T. velebiticum* orbiculate or slightly cordate. About 80% of the patterns of *T. minus* — complex have the wedge — shaped base of leaflets (Fig. 5) and most plants gathered in Paklenica (about 90%) have a heart — shaped base of leaflets suggesting the morphological distance of this population.

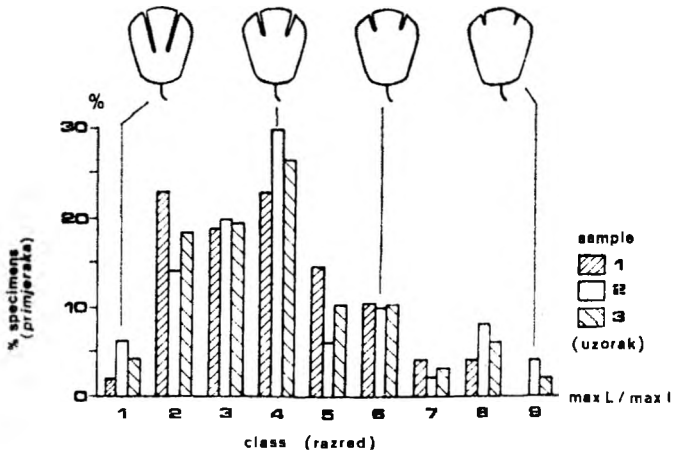


Fig. 4. Distribution of samples with regard to max L/max I; the hypothetical examples given in scale for the middle of the corresponding class; the class ranges: (1) 1.5—1.9, (2) 2.0—2.4, (3) 2.5—2.9, (4) 3.0—3.4, (5) 3.5—3.9, (6) 4.0—4.4, (7) 4.5—4.9, (8) 5.0—5.4, (9) 5.5—5.9.

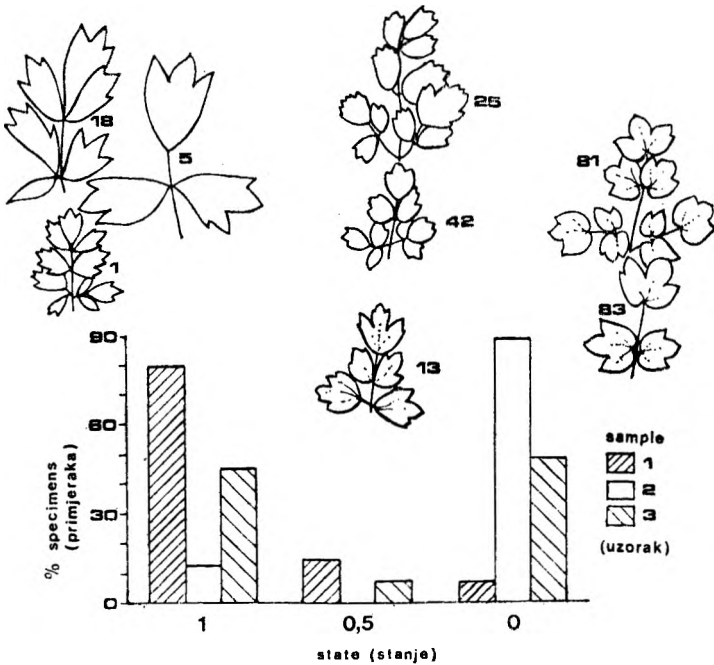


Fig. 5. Distribution of samples according to the shape of the base of leaflets.



This conclusion is rendered rather difficult because of several facts, namely:

— the presence of such intermediate shapes of the leaflets bases that cannot be classified unambiguously in either the shape 1 or shape 0 (0.5);

— a high degree of subjectivity in the classification of dubious shapes into one of the proposed states; and

— as a result of the above, that this group of plants in nature does not exist in the 3 proposed or even more states, but, it seems, in a much more continuous series the verbal description of which most often causes difficulties in determination. Anyway, it is evident that the population from Paklenica shows certain particularities with regard to the shape of the base of leaflets.

#### Coefficient of variations

A comparison of the coefficients of variations V, as comparable relative indications for variability (Fig. 6) shows:

— the highest variability of the characters relating to the dimensions of leaflets (characters 1, 2, 3 and 4) with  $V > 30\%$  and the statistically significant difference between  $S_1$  and  $S_2$  for min W of the leaflets;

— a slightly lower variability, especially for sample 1, of characters 6 and 7 and his statistically significant difference between  $S_1$  and  $S_2$  for the character 6.

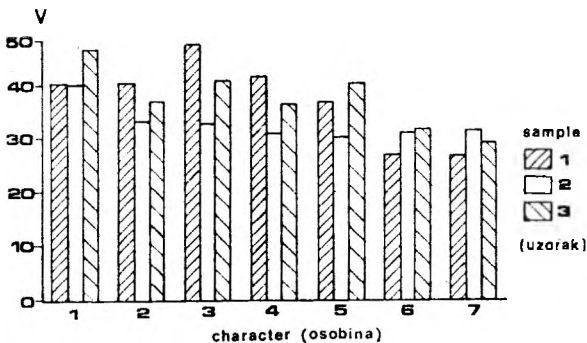


Fig. 6. Coefficients of variability for the samples 1, 2 and 3 for the characters 1—7.

#### Conclusion

None of the characters treated shows diagnostically relevant characteristics. This particularly holds for the dimensions of leaflets which are so frequently used for such purposes.

All characters show continuity in shapes between the limit values. The sample from Paklenica, the representative of the species *T. velebicum* Deg. 1937, is not distinctive by these morphological features of leaflets, although it shows characteristics of the population, especially in min W of leaflets, in min and max I of leaflets and in shape of the base of leaflets. It must be said, however, that these same characters in correlation with some other features may, perhaps be of taxonomical importance.

The high phenotypic variability is obviously one of the principle causes of taxonomical and determinational difficulties (Tutin 1964, Damboldt and Zimmermann 1974). However, the evolutionary strategy and paths of divergence in the genus and complex are still insufficiently known.

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SAŽETAK

FENOTIPSKA VARIJABILNOST LISAKA U *THALICTRUM MINUS* — KOMPLEKSU  
(*RANUNCULACEAE*)

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Prikazana je varijabilnost 8 osobina lisaka na *T. minus*-kompleksu iz Jugoslavije s osobitim osvrtom na *T. velebiticum* Deg. 1937, te na mogućnost njihovog razlikovanja upotrebom navedenih osobina. Podaci su obrađeni klasičnom statistikom uz primjenu  $t$  i  $F$ -testova, a komparirani su i koeficijenti varijabilnosti za svaki uzorak i osobinu. Osobine pokazuju značajnu varijabilnost, a zbog velikog preklapanja među uzorcima i slabu dijagnostičku vrijednost. Uzorak koji reprezentira *T. velebiticum* pokazuje stanovite populacijske osobitosti.

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