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**Tom XXVI-XXVII, Beograd, 1992-1993.**

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## **ГЛАСНИК**

**ИНСТИТУТА ЗА БОТАНИКУ И БОТАНИЧКЕ  
БАШТЕ УНИВЕРЗИТЕТА У БЕОГРАДУ**

**Tom XXVI-XXVII**

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1994.**

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**Institut za botaniku i botanička bašta „Jevremovac” – Beograd, Takovska 43**  
**Jugoslavija**

**ГЛАСНИК ИНСТИТУТА ЗА БОТАНИКУ И БОТАНИЧКЕ  
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TATJANA STEVIĆ, BRANKA VUKOVIĆ-GAČIĆ, JELENA  
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## DETECTION OF NATURAL BIOANTIMUTAGENS BY BACTERIAL SHORT-TERM TESTS

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Stević, T., Vuković-Gačić, B., Knežević-Vukčević, J., Jankov, R.M. and Simić, D. (1992-1993): *Detection of natural bioantimutagens by bacterial short-term tests*. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 1 - 6.

Antimutagenic activity of CO<sub>2</sub> reextract of the cultivated sage containing terpenoid fraction, detected in *E. coli* repair proficient strain, was tested using *S. typhimurium*/microsome assay (Ames test). There was no effect on spontaneous and UV-induced mutations in TA98 strain. The reduction of UV-induced mutation frequency was observed when tested with TA100 strain with metabolic activation. Possible reasons for the discrepancy between results obtained with *E. coli* and *S. typhimurium* test systems are discussed.

Key words: short-term tests, Ames test, antimutagenesis, sage extract.

Ključne reči: kratkoročni testovi, Ames-ov test, antimutagenaza, ekstrakt žalfije.

## INTRODUCTION

Evaluation of different biological activities of extracts of various plants is directed to several principles: antimicrobial activities, antimutagenic/anticarcinogenic activities (Kuroda et al., 1990) and the direct effect on some specific tissues. The present data on mechanisms of inhibitors of mutagenesis/carcinogenesis, although still fragmentary, provides an extremely useful scientific premise for the primary prevention of mutation-related diseases.

According to Kada et al., (1982, 1986) antimutagenic and anticarcinogenic effect can be achieved by means of three mechanisms: (i) by increasing the fidelity of DNA replication, (ii) by favoring error-free repair of DNA damage, (iii) by inhibiting error-prone repair systems. For some antimutagenic agents mechanisms of mutagenesis inhibition are known: cobaltous chloride increases the fidelity of DNA replication and enhances recombination repair, sodium arsenite inhibits the *umuC* gene expression and enhances error-free repair in bacteria (Nunoshiba & Nishioka, 1987). The antimutagenicity of cinnamaldehyde, coumarin, umbelliferone, vanillin and tannic acid is ascribed to promotion of error-free DNA damage repair (Kada et al., 1986; Shimoi et al., 1985). There is also considerable evidence for antimutagenic effect of many naturally-occurring compounds in bacterial (Kuroda & Inoue, 1988; Kuroda, 1990) and mammalian test systems (Bootman et al., 1988), although the underlying mechanisms are still obscure.

The bacterial short-term tests, routinely used to detect environmental mutagens, are recommended for identifying antimutagens. In addition to their rapidity and low costs they provide considerable information about cellular and molecular mechanisms of mutagenesis and antimutagenesis. Combined with activation systems, they can even provide information about the kind of metabolic activation or detoxification that the agent may undergo *in vivo*.

In this work we tested antimutagenic effect of CO<sub>2</sub> reextract of cultivated sage (*Salvia officinalis* L.) containing terpenoids in the *Salmonella typhimurium* mutagenicity test (Ames test). This test is recommended by OECD (Organization for Economic Co-operation and Development, 1986) and is widely used in environmental mutagenicity testing.

## MATERIAL AND METHODS

Strains. *Salmonella typhimurium* strains were TA98 (*hisD3052rfaΔuvrB*/pKM101) and TA100 (*hisG46 rfa ΔuvrB*/pKM101) (Ames et al., 1975).

*S. typhimurium* mutagenicity assay medium was 1.5% Difco bacto agar and 2% D-glucose in Vogel-Bonner medium E (Maron & Ames, 1983). Top agar containing Difco bacto agar (6 mg/ml) and NaCl (5 mg/ml) was supplemented with 0.05 mM biotine and 0.05 mM histidine.

S9 mix. S9 fraction was isolated from the liver of Albino Wister male rats (170-180 g) induced with pheno-barbital/β-naphtho flavone (Garner et al., 1972). S9 mix contained 4% (v/v) S9 fraction, 33 mM KCl, 8 mM MgCl<sub>2</sub>, 5 mM glucose-6-phosphate and 4 mM NADP in 0.1 M phosphate buffer pH 7.4.

*S. typhimurium* assay. The overnight culture of *S. typhimurium* strain was washed by centrifugation and resuspended in 0.01 M MgSO<sub>4</sub> giving a similar titer. UV-irradiation conditions were the same as described previously (Simić et al., 1985). Samples

(0.1 ml) of UV-irradiated cells were added in 2 ml of molten top agar with or without S9 mix (0.5 ml), mixed and poured onto minimal glucose agar plates with or without sage extract. After incubation at 37°C for 48 h the number of His<sup>+</sup> revertants was determined and the presence of the background lawn on all plates was confirmed.

Preparation of sage extract. *Salvia officinalis* L. cultivated in Bački Petrovac (Vojvodina), collected during the flowering period 1992, was dried, ground and subjected to extraction and CO<sub>2</sub> reextraction as described previously (Đarmati et al., 1993). The CO<sub>2</sub> extract was dissolved in ethanol just before use and diluted with distilled water.

## RESULTS AND DISCUSSION

In our previous work (Vuković-Gačić & Simić, 1993) we designed *Escherichia coli* K12 assay-system for detection of bioantimutagens, factors which reduce the apparent spontaneous and induced mutation frequency by interfering with cellular processes of mutation fixation (Kada et al., 1985; Kuroda, 1990). The set of tester strains consists of a) SY252 repair proficient strain for detection of induced reversions, b) isogenic mutator strains deficient in methyl-directed mismatch repair (*mutH*, *mutL*, *mutS* and *uvrD*) for detection of spontaneous mutations, c) isogenic repair proficient strain carrying *sfiA::lacZ* fusion for measuring the level of SOS induction (induction of mutagenic SOS repair). The inhibition of spontaneous and UV-induced mutagenesis was studied by reversion of *argE3* ochre mutation which can occur by base substitution, mostly at A:T sites (Todd et al., 1979).

To exclude desmutagens, factors which act directly on mutagens or their precursors and inactivate them (Kada et al., 1982), we used UV-irradiation as mutagen. The assay-system was validated using model bioantimutagens, cobaltous chloride and tannic acid.

With a set of newly constructed *E. coli* strains we have carried out a comparative screening for natural antimutagens from various medicinal plants. The refractory antimutagenic capacity was obtained with nontoxic concentrations of 9 extracts (St. John's wort, thyme, aloe, camomile, nettle, mint, lime-tree, sage and X-tea), depending on the bacterial strain used and the concentration of the extract applied (Vuković-Gačić & Simić, 1993) illustrating the complex situation which is expected for the whole extract.

Further study was performed with CO<sub>2</sub> reextracts of cultivated and wild sage containing terpenoids. Among three extracts tested only extract of cultivated sage, prepared without steam distillation prior to ethanolic extraction, suppressed UV-induced mutagenesis in *E. coli* repair proficient strain (Simić et al., 1994).

To test the sage extract with antimutagenic activity in *E. coli* we used *S. typhimurium* tester strains which detect frameshift mutations (TA98) or base substitutions mostly at G:C sites (TA100) (Maron & Ames, 1983). The experiments were carried out with or without addition of microsomal fraction of rat liver (S9). The presence of metabolic activation enzymes in the test system enables transformation of extract compounds and mimics the situation in mammalia.

Tab. 1. – Effect of sage extract on spontaneous and UV-induced mutations in TA98 strain

Sage extract (µg/plate)	His <sup>+</sup> revertants/plate					
	-S9			+S9		
	-UV	+UV	% I	-UV	+UV	% I
0	41	213		26	307	
50	30	227	-6	34	233	12
75	28	251	-17	36	303	2
100	36	232	-8	38	307	0
150	40	241	-13	34	275	11
200	64	222	-4	29	291	5

UV dose was 6 J/m<sup>2</sup>.

The numbers represent the average of duplicate plates.

%I = 1 - (Nt/Nc) x 100

Nt - number of mutants/plate with sage extract;

Nc - number of mutants/plate without extract.

The effect of sage extract on spontaneous and UV-induced His<sup>+</sup> revertant colonies in TA98 strain is shown in Tab. 1. The extract is without significant effect on spontaneous and UV-induced frameshift mutations, neither with S9 fraction nor without.

Tab. 2. – Effect of sage extract on spontaneous and UV-induced mutations in TA100 strain

Sage extract (µg/plate)	His <sup>+</sup> revertants/plate					
	-S9			+S9		
	-UV	+UV	% I	-UV	+UV	% I
0	172	1636		126	2312	
50	134	1744	-7	126	1530	34
75	181	1644	0	133	1684	27
100	171	1560	5	131	1563	34
150	142	1388	15	131	1455	37
200	124	1510	8	132	1320	43

UV dose was 6 J/m<sup>2</sup>.

The numbers represent the average of duplicate plates.

%I was calculated as in Table 1.

Moreover, in TA100 strain, detecting base substitution as SY252 stain, UV-induced mutations were not reduced in the absence of S9 fraction (Tab. 2). This result may be due to lack of excision repair in *S. typhimurium* strains which would prevent detection of antimutagenic agents enhancing excision repair. Further study with repair proficient *S. typhimurium* strains could validate this hypothesis.



Interestingly, in TA100 strains UV-induced mutations were reduced about 40% when extract was exposed to metabolic activation (Tab. 2). It is already established that this sage extract contains a variety of terpenoids (Darmati et al., 1993) and it is possible that in *S. typhimurium* TA100 strain we detect different compounds which require metabolic transformation for antimutagenic activity. The study with purified terpenoids is under the way.

### ACKNOWLEDGEMENTS

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### Rezime

TATJANA STEVIĆ, BRANKA VUKOVIĆ-GAČIĆ, JELENA  
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### DETEKCIJA PRIRODNIH BIOANTIMUTAGENA POMOĆU KRATKOROČNIH BAKTERIJSKIH TESTOVA

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U divljem soju *E. coli* otkrivena je antimutagena aktivnost CO<sub>2</sub> reekstrakta kultivisane žalfije koji sadrži frakciju terpenoida. U ovom radu za detekciju antimutagene aktivnosti CO<sub>2</sub> reekstrakta žalfije korišćen je *S. typhimurium*/mikrozom test (Ames-ov test). U soju TA98 nije utvrđen efekat na spontanu i UV-indukovanu mutagenezu, a u TA100 soju utvrđeno je smanjenje frekvence UV-indukovanih mutacija u prisustvu mikrozomalne frakcije ćelija jetre pacova. Navedeni su i diskutovani mogući razlozi koji su doveli do različitih odgovora u *E. coli* i *S. typhimurium* test sistemima.

UDC 582.951.8 : 588.7 : 57.086.83 (497.11)  
Original scientific paper

MILICA MANOJLOVIĆ, BRANKA VINTERHALTER, DRAGAN  
VINTERHALTER

***IN VITRO* PROPAGATION OF *CATALPA OVATA* G. Don AND *CATALPA*  
*BIGNONIOIDES* Walt.**

Institute for Biological Research, „Siniša Stanković,” Belgrade

Manojlović, M., Vinterhalter, B., Vinterhalter, D. (1992-1993): *In vitro propagation of Catalpa ovata* G. Don and *Catalpa bignonioides* Walt – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 7 - 14.

Shoot cultures of *Catalpa ovata* and *C. bignonioides* were started from epicotyls of aseptically germinated seeds and maintained on Murashige and Skoog (1962) based medium. Optimal BA concentration for shoot multiplication was  $0.5 \text{ mg}^{-1}$  for *C. ovata* and  $1.0 \text{ mg}^{-1}$  for *C. bignonioides*. IBA had little effect on shoot multiplication and  $0.1 \text{ mg}^{-1}$  was considered optimal for both species. Shoots of *C. bignonioides* were more than twice longer than shoots of *C. ovata* on all BA concentrations exceeding  $0.1 \text{ mg}^{-1}$ . Shoots 1.5-2.0 cm long could be easily rooted, media supplemented with  $0.5 \text{ mg}^{-1}$  IBA was considered optimal. Rooted plantlets were adapted in glasshouse after planting in mixtures of peat, sand and organic manure.

Key words: *in vitro*, propagation, shoot cultures, *Catalpa ovata* G. Don., *Catalpa bignonioides* Walt.

Ključne reči: *in vitro*, razmnožavanje, kulture izdanaka, *Catalpa ovata* G. Don., *Catalpa bignonioides* Walt.

## INTRODUCTION

Genus *Catalpa* of the Bignoniaceae family contains about 10 species of trees which are planted as ornamental plants or in timber plantations (Bailey, 1949). *C. bignonioides* Walt. is native to North America and *C. ovata* Don, to East Asia (China, Japan). They can be distinguished by color of flowers; *C. ovata* has yellow flowers striped orange and spotted dark violet whilst *C. bignonioides* has white flowers with two yellow stripes inside and spotted purplish-brown. Both species are characterized by very large, hart shaped leaves. Decorative appearance of these specimens is enhanced by fruits which are large, up to 40 cm long pods. Leaves, flowers and fruits are known to contain pharmacologically active substances like catalpol and catalposide which have diuretic properties. Bruised leaves of *C. bignonioides* emit an unpleasant odor.

In this paper we present results on the investigation of *in vitro* methods suitable for clonal propagation of *C. ovata* and *C. bignonioides*. *In vitro* propagation of *C. bignonioides* has been elaborated by Wysłokinska and Swiatek, 1989.

## MATERIAL AND METHODS

Seeds of both species were collected from trees growing in Botanical garden, Belgrade. Seeds were surface sterilized for 20 min in 10% solution of commercial bleach (4.5% NaOCl) to which few drops of liquid detergent have been added. Seeds were thoroughly washed in autoclaved water and then germinated on moist filter paper in small petri dishes, 5 seeds per plate. After first signs of germination, seeds were transferred to agar solidified medium, individually in 10 x 100 test tubes. This medium was either hormone-free- or supplemented with 1.0 mg l<sup>-1</sup> BA and 0.2 mg l<sup>-1</sup> IBA. After proper germination, epicotyls were excised and first true subculture was performed.

Medium used in all experiments comprised Murashige and Skoog (1962) inorganic salts, vitamins and inositol. It was modified to contain 0.4 mg l<sup>-1</sup> vitamin B<sub>1</sub>, 2.0% sucrose and 0.62% agar. Media was autoclaved for 20 min on 114-115°C, Ph was adjusted to 5.5-5.7 prior to autoclaving. Culture vessels used for shoot multiplication were 100 ml wide neck Erlenmeyer flasks containing 40 ml of medium or 150 ml transfusion bottles containing 50 ml medium. Both types of culture vessels were closed by cotton wool plugs. Conditions in the growth room were 16/8 hours light to darkness, irradiance 5.0-7.2 (10.) Wm<sup>-2</sup> provided by cool white fluorescent lamps. Temperature in the growth room was adjusted to 25 ± 2°C.

Multiplication index – parameter used to evaluate propagation rate corresponds to the number of new propagula (shoots explants) produced by a single shoot explant during subculture lasting for 4 weeks, which can be used for further subculturing.

Rooting was performed with shoots 1.5-2.0 cm long. Rooting results were scored after 4 weeks. Planting substrate was prepared from mixture of peat, sand and „Beohumus” (organic manure). Adaptation and further cultivation of rooted plantlets were performed in glasshouse. Transferred plantlets were frequently sprayed with 0.3% Venturin to prevent damage by fungi.

All experiment were repeated at least twice with no less than 24 replicates per treatment.

## RESULTS

Experiments were started in spring 1989. Surface sterilization of seeds was successful and the number of seeds found to be contaminated on sterile filter paper was practically nil. Some inborn bacterial contamination were found later specially in *C. ovata*. Upon transfer to agar solidified medium germination was fast and first true subculture which included excision of epicotyl was performed after three weeks. On hormone free medium epicotyls were long but terminal buds often perished from necrosis. On medium with  $1.0 \text{ mg l}^{-1}$  BA and  $0.2 \text{ mg l}^{-1}$  IBA epicotyls were short but healthy, and therefore they were used for establishment of shoot cultures.

From second subculture epicotyl explants of both species were transferred to medium supplemented with  $0.5 \text{ mg l}^{-1}$  BA and  $0.1 \text{ mg l}^{-1}$  IBA, which was previously in our laboratory found to be suitable for maintenance of shoot cultures of a number of dicotyledonous tree species including; apple, quince, mulberry, carob, various *Prunus* species and other. Preliminary results showed marked differences in the growth of two *Catalpa* species. Whilst growth of *C. ovata* was satisfactory, shoot multiplication of *C. bignonioides* was low and shoot length unusually high. To investigate the optimal BA concentration for maintenance of shoot culture stocks of both species, experiments were performed in which BA concentration was varied ( $0\text{-}2.0 \text{ mg l}^{-1}$ ) and IBA concentration was at constant concentration  $0.1 \text{ mg l}^{-1}$  (Fig. 1).

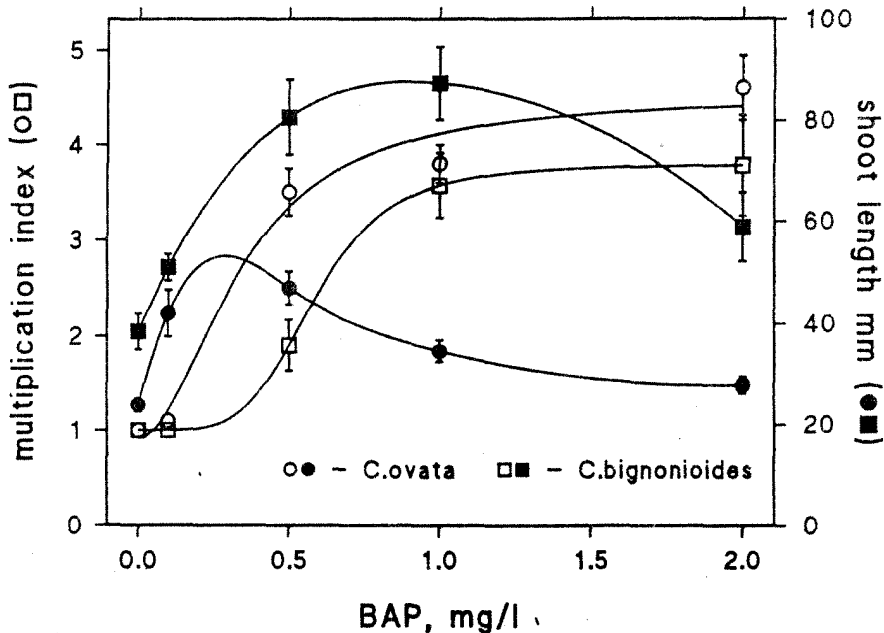


Fig. 1. - Effect of  $0\text{-}2.0 \text{ mg l}^{-1}$  BA on shoot multiplication and shoot elongation in *Catalpa ovata* and *C. bignonioides*.

According to Fig. 1, shoot multiplication of *C. ovata* increased faster with increase of BA concentration than in *C. bignonioides*. In *C. ovata* plateau for shoot multiplication was reached at  $0.5 \text{ mg l}^{-1}$  BA. Thus at  $0.5 \text{ mg l}^{-1}$  BA shoot multiplication of *C. ovata* was nearly twice higher than in *C. bignonioides*. The maximum values for shoot length in *C. ovata* is somewhere between  $0.1$  and  $0.5 \text{ mg l}^{-1}$  BA and for *C. bignonioides* at  $1.0 \text{ mg l}^{-1}$ . In the range  $0.5$ - $2.0 \text{ mg l}^{-1}$  BA shoots of *C. bignonioides* were twice longer than shoots of *C. ovata* which was an obvious and striking difference between the two *Catalpa* species (Fig. 2). Considering both shoot multiplication and shoot length, the optimum BA concentration for maintenance of shoot cultures was  $0.5 \text{ mg l}^{-1}$  for *C. ovata* (as initially provided) and  $1.0 \text{ mg l}^{-1}$  for *C. bignonioides*. In general *C. bignonioides* at all BA concentration exhibited unusually high shoot elongation not only in relation to *C. ovata* but also in relation to other dicotyledonous tree species.

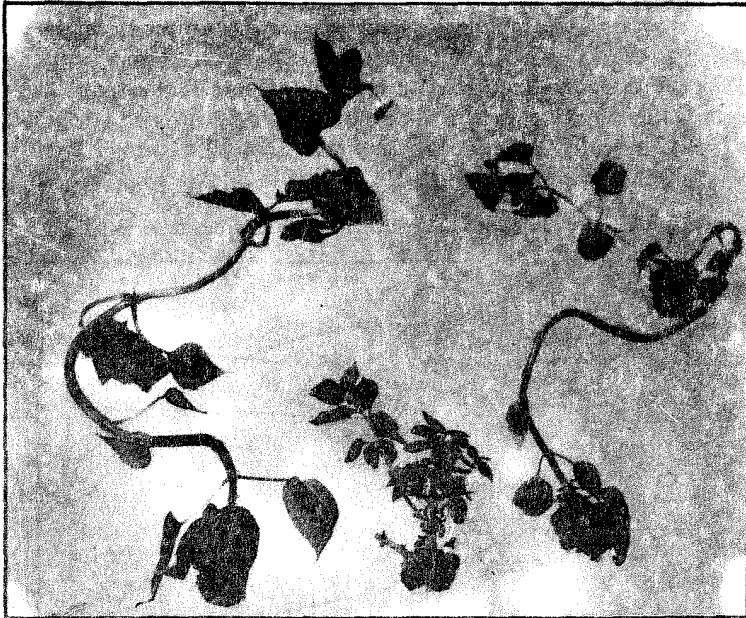


Fig. 2. – Shoot cultures of *Catalpa ovata* (center) and *C. bignonioides* (left and right) after 4 weeks on medium containing  $0.5 \text{ mg l}^{-1}$  BA and  $0.1 \text{ mg l}^{-1}$  IBA. Compare difference in shoot length.

Since preliminary results showed that IBA in  $0$ - $1.0 \text{ mg l}^{-1}$  had little effect on shoot multiplication except that it stimulated callusing, and that  $0.1 \text{ mg l}^{-1}$  IBA already gave good results in the maintenance of shoot culture stocks, effect of IBA concentration on shoot multiplication was not further investigated.

Single isolated roots of both species could be easily rooted on hormone-free or media containing auxins. The effect of  $0$ - $1.0 \text{ mg l}^{-1}$  IBA on the number of roots per

rooted shoot and length of longest root is presented in Fig. 3. It is evident that for both species results were nearly identical. Root length decreased with increase of IBA in the medium whilst the number of roots per rooted shoot increased.

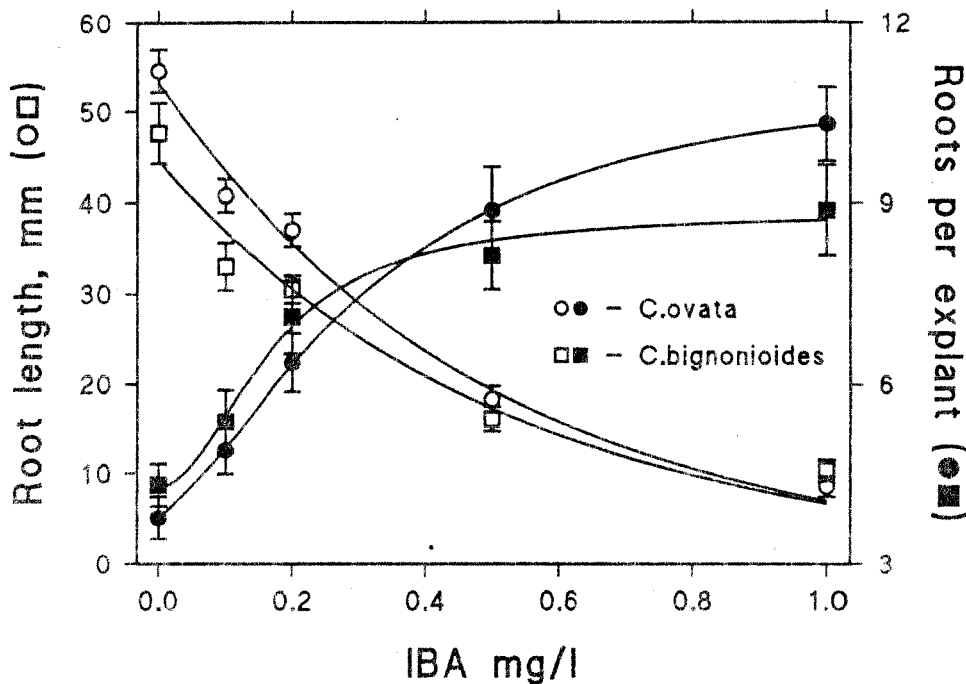


Fig. 3. – Effect of 0-1.0 mg<sup>l</sup> IBA on the average number of roots per rooted culture and root length.

Rooting could also be achieved in a two-stage rooting procedure in which shoots were first placed on media supplemented with 2.0 mg<sup>l</sup> IBA and then transferred to hormone free medium after 1, 2, 4 or 7 days. The number of roots per rooted shoot was very high, Fig. 4. In *C. ovata* it increased with increased duration of the first stage of rooting but in *C. bignonioides* it was constant in all treatments. Also root length in *C. ovata* decreased with prolonged duration of auxin treatment whilst in *C. bignonioides* values were erratic and roots in general were short.

Adaptation of plantlets after transition to *ex vitro* conditions was affected by the rooting method. In plants rooted in continuous contact with medium supplemented with IBA, success in adaptation was 62-80% for *C. ovata* and 86-100% for *C. bignonioides*. Adaptation of plantlets rooted in the two-stage procedure was less successful, 10-66% for both species. Further growth of transferred plantlets in the glasshouse was fast.

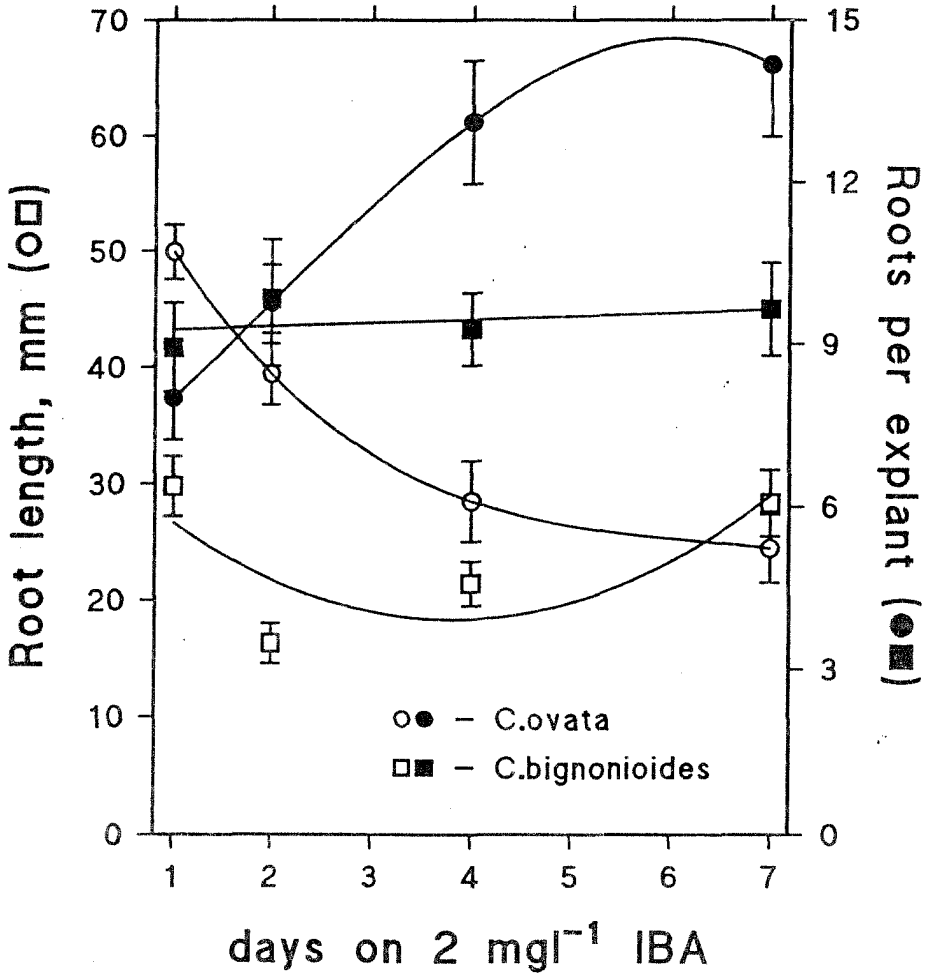


Fig. 4. - Effect of duration of the auxin treatment (1, 2, 4 and 7 days on 2.0 mg l<sup>-1</sup> IBA) followed by transfer to hormone-free medium on the average number of roots per rooted culture and root length.

#### DISCUSSION

The technique of *in vitro* propagation of dicotyledonous tree species from epicotyls of aseptically germinated seeds has proved in our laboratory to be a simple and reliable method for fast establishment of *in vitro* cultures. This method has been specially useful when clonal propagation was not the primary goal of investigation and when preliminary culture requirements for shoot cultures of certain species where



sought. In this way conditions were determined for maintaining shoot cultures of *Ceratonia siliqua* (Vinterhalter et al., 1992; Vinterhalter & Vinterhalter, 1992) and *Monia alba* (Vinterhalter & Grubišić, 1990). Although the use of seedling epicotyls can not be considered as true clonal propagation (Krikorian, 1982) it can be used in species propagated by open pollinated seeds. Wysockinska and Swiatek (1989) presented a multiplication scheme for *C. bignonioides* in which shoot cultures were established by differentiation of callus originating from hypocotyls of two week old seedlings. Reasons for which this propagation scheme is based on the use of callus are not clear. We however propose the use of epicotyls and a significant shortcut in the *in vitro* propagation scheme for both *Catalpa* species.

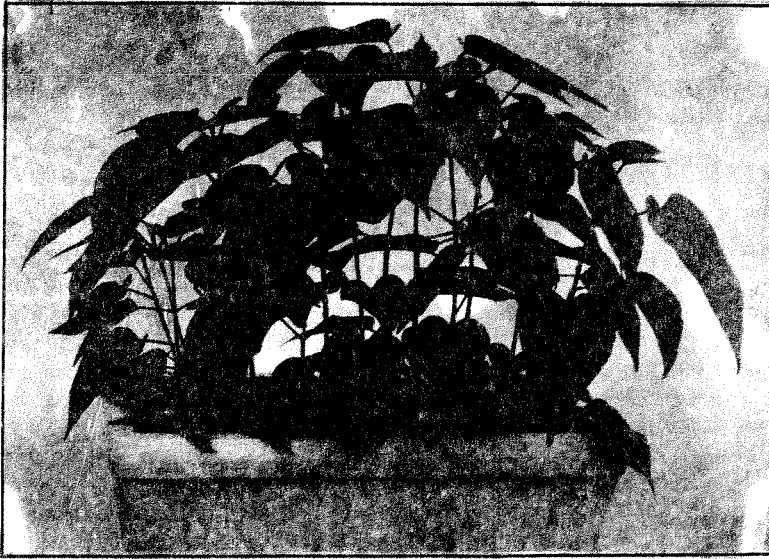


Fig. 5. – Plantlets of *C. bignonioides* after successive adaptation.

Hormonal balances which we used for shoot multiplication and rooting of *Catalpa ovata* are well known to give good results with many dicotyledonous species including almost all top fruit species (apples, pears, quinces, plums, peaches, cherries etc.). *C. bignonioides* required higher cytokinin concentration ( $1.0 \text{ mg l}^{-1}$  BA) for satisfactory shoot multiplication. Wysockinska and Swiatek (1989) recommend even  $2.0 \text{ mg l}^{-1}$  BA as optimal for shoot multiplication which according to our findings already induced some vitrification and fasciation.

Our results in general support the findings of Wysockinska and Swiatek (1989) and show that other *Catalpa* species like *C. ovata* can also be propagated by *in vitro* methods.

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## Rezime

MILICA MANOJLOVIĆ, BRANKA VINTERHALTER, DRAGAN VINTERHALTER

***IN VITRO* RAZMNOŽAVANJE *CATALPA OVATA* G. DON I *CAPALTA BIGNONIOIDES* WALT.**

Institut za biološka istraživanja „Siniša Stanković“, Beograd

Kulture izdanaka *Catalpa ovata* i *C. bignonioides* pokrenute iz epikotila aseptično iskljajalog semena gajene su na podlozi po Murashige i Skog-u (1962). Optimalna koncentracija BA za multiplikaciju izdanaka bila je  $0.5 \text{ mg}^{-1}$  za vrstu *C. ovata* i  $1.0 \text{ mg}^{-1}$  za *C. bignonioides*. IBA je vrlo malo uticala na multiplikaciju pa je koncentracija  $0.1 \text{ mg}^{-1}$  smatrana optimalnom za obe vrste. Izdanci vrste *C. bignonioides* bili su više nego dva puta duži nego izdanci vrste *C. ovata* na svim podlogama koje su sadržavale više od  $0.1 \text{ mg}^{-1}$  BA. Izdanci dužine 1.5-2.0 cm su se lako ožiljavali, a podloga sa  $0.5 \text{ mg}^{-1}$  IBA je bila optimalna. Adaptacija ožiljenih biljaka nakon presađivanja u smesu treseta, peska i organskog đubriva bila je uspešna a biljke su dalje gajene u staklari.

UDC 581.1 : 582.475.4 (497.11)  
Original scientific paper

MILA BOGDANOVIĆ, GORDANA DRAŽIĆ, MILICA VUČKOVIĆ

**SEASONAL CHANGES OF PIGMENT CONTENT IN THE NEEDLES OF  
DOUGLAS FIR (*PSEUDOTSUGA TAXIFOLIA* BRITT.) IN NATURAL  
CONDITIONS**

INEP, Institute of Biotechnological Research, University of Belgrade,  
Zemun-Beograd

Bogdanović, M., Dražić, G., Vučković, M. (1992-1993): *Seasonal changes of pigment content in the needles of Douglas fir (Pseudotsuga taxifolia Britt.) under natural conditions*. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 15 - 20.

The results of investigations have shown that the age of the tree of the yellow form of Douglas fir may not be involved in chlorophyll and carotenoid contents of its needles. In all trees examined, needles of the past year had more chlorophyll and carotenoids. In the yellow form of Douglas fir there were only slight variations of pigment content in the needles during the year. Green variety needles, however, have a twofold pigment content than the yellow form, accompanied by greater variations. Interdependence of chlorophyll and carotenoid metabolism is discussed.

Key words: *Pseudotsuga taxifolia*, chlorophyll, carotenoids, seasonal changes.

Ključne reči: *Pseudotsuga taxifolia*, hlorofil, karotenoidi, sezonsko variranje.

## INTRODUCTION

First investigations of conifer photosynthetic pigments date back to the past century. Most data to be found in the literature refer to *Pinus* (Gerhold, 1959a, 1959b; Bogdanović et al., 1981; Bogdanović & Mančić, 1989), and are likely to produce a conclusion that the seasonal changes of the amount and composition of conifer photosynthetic pigments in natural conditions may reflect their metabolism.

From theoretical aspects, investigations of conifers lead to the understanding of metabolism control mechanism of leaf pigments throughout the year. On the practical side these investigations are interesting because many coniferous trees are used for the industry or landscaping of human settlements. In our country, Douglas fir (*Pseudotsuga taxifolia* Britt.) is widely spread both as a culture and the ornamental plant. Most commonly encountered are varieties *viridis* and *glauca*. The variety *viridis* occurs in its yellow form (Vidaković, 1982).

The objective of the present study was to examine the nature of the seasonal changes of pigment contents and their ratios in the needles of green and yellow Douglas firs in natural conditions.

## MATERIAL AND METHODS

Needles of the first and second vegetation year of the green Douglas fir, age 30 years, and yellow forms aged 30 and 5 years of age were used as experimental material. All trees were grown in the garden of the Institute for the Application of Nuclear Energy in Zemun.

Needles for pigment analysis were sampled monthly for a year, always from the same shoots due to variability of material.

Chlorophylls were determined using spectrophotometry according to Vernon (1960), and carotenoids according to Holm (1954). The results are the means of four replications.

## RESULTS

The seasonal variation of chlorophylls and their ratios in the needles of Douglas fir are shown in Fig. 1. The content of total chlorophyll in the current needles of a young yellow tree remains almost unchanged. A slight raise may be noticed in May and June, and then again in February (Fig. 1A). The needles of the preceding year contain more chlorophylls, however, with a content stable throughout the year. The ratio of Chl.a/Chl.b shows similar variations in both the current and preceding needles. Beginning summer, this ratio drops and then raises again as a consequence of the variations in the content of Chl.b.

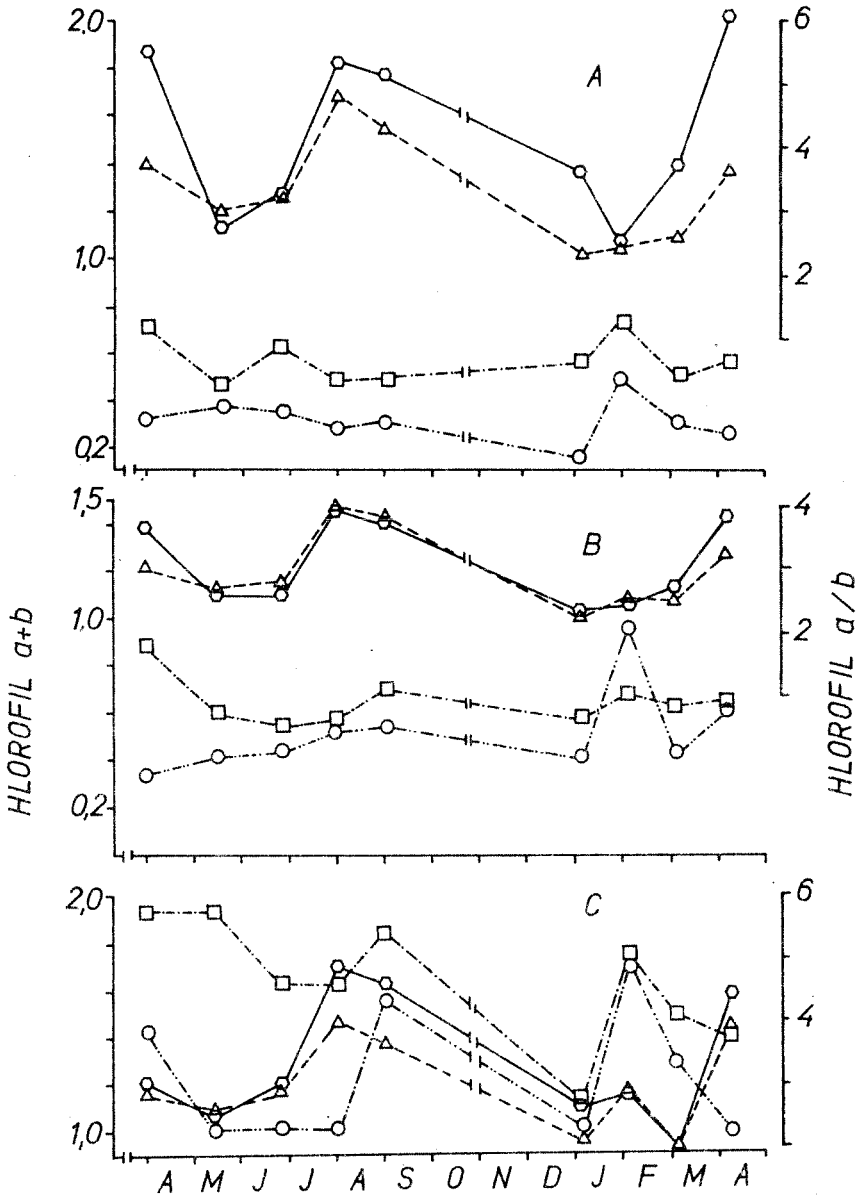


Fig. 1. - Seasonal variations of total chlorophylls (mg/g fresh matter) and Chl a/b ratio in the needles of: A - young yellow tree, B - adult yellow tree, and C - adult green tree.

- - - - ○ Chlorophyll content of current needles.
- - - - □ Chlorophyll content of a year-old needles.
- - - - ○ Chl a/b ratio in current needles.
- △ - - - △ Chl a/b ratio in a year-old needles.

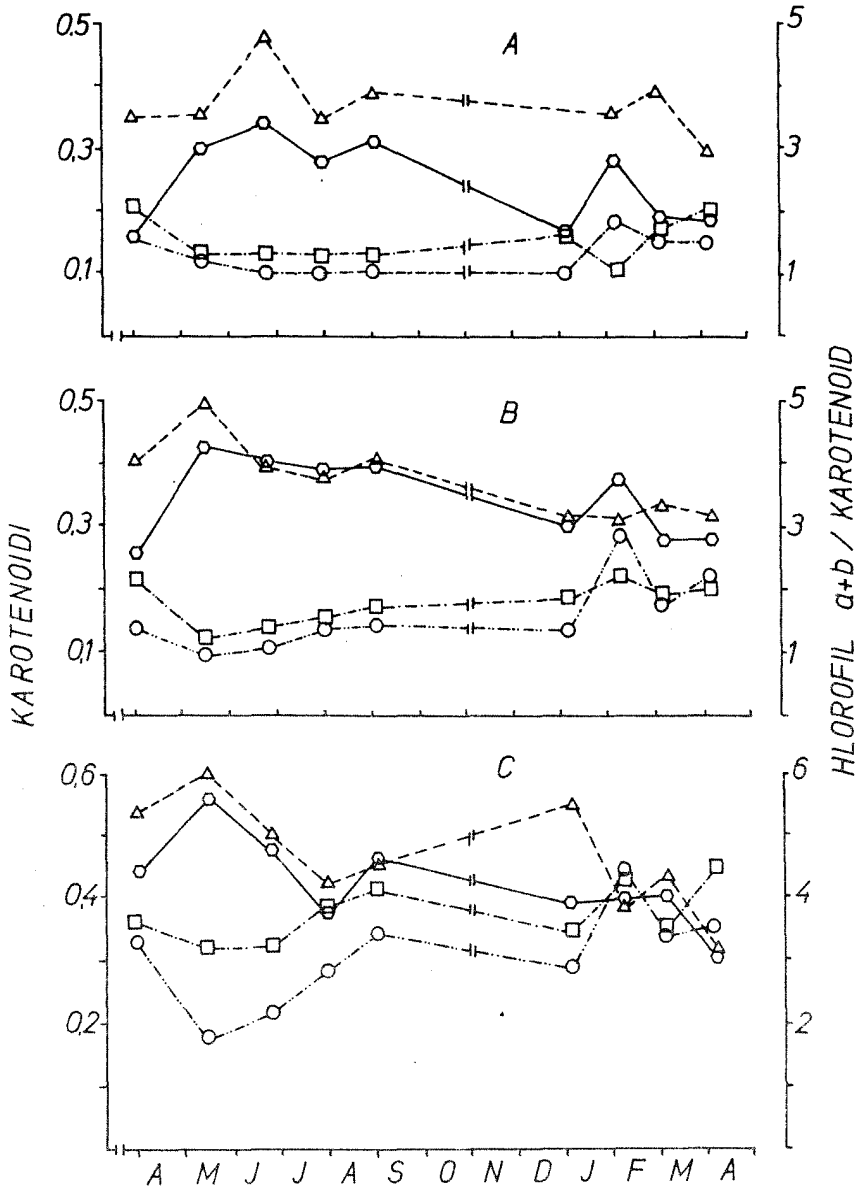


Fig. 2. – Seasonal variations of total carotenoid content (mg/g fresh matter) and of total chlorophyll/total carotenoid ratio in the needles of: A – young yellow tree, B – adult yellow tree, and C – adult green tree

- - - - ○ Carotenoid content of current needles.
- - - - □ Carotenoid content of a year-old needles.
- - - - ○ Chl/carotenoid ratio in current needles.
- △ - - - △ Chl/carotenoid ratio in a year-old needles.

The needles of the old yellow tree follow the same chlorophyll variation pattern, however, the content of chlorophyll in its needles is somewhat higher (Fig. 1B), and concerns needles of both the current and the preceding year. On the other hand, Chl a/Chl b ratio varies less than in a young tree needles, and throughout the year shows values typical of mature leaves of the majority of green plants.

The needles of the full-grown green tree have double chlorophyll content whose variation pattern is similar to that of an old yellow tree. The ratio Chl a/Chl b is most often about 3. In spring it tends to increase and remain at a higher level, to drop during summer months. The tendency is the same in the current and a year-old needles.

Figure 2 shows variations of carotenoid contents and varying ratios of total chlorophyll/total carotenoid. Carotenoid content is mostly stable throughout the period of investigation, with slight oscillations noticed in the spring.

Older needles contained slightly more carotenoids than the current needles. No significant differences could be noticed between needles of the young and the old yellow tree (Fig. 2A, B). A slight variation of the ratio total chlorophyll/total carotenoid was noticeable during summer months. The raise of this ratio was the consequence of the increased chlorophyll content, while carotenoids remained at the same level.

The needles of the green Douglas fir had an almost twofold carotenoid content than the needles of yellow trees. A slight raise of carotenoids could be noticed in summer (Fig. 2C). The ratio Chl a+b/carotenoid in the needles of a mature Douglas fir tends to vary during the year, showing a sudden increase in the spring and a gentle drop in the summer.

## DISCUSSION

It was shown that the content of chlorophyll and carotenoids tended to vary during the year in the young seedlings of Scotch pine. These variations depended on the temperature (Bogdanović & Mančić, 1989), leading to reddening of the needles in the winter and their greening in the spring. However, needles of the adult Douglas fir are green or yellowish throughout the year. This would suggest a genetical determination of the colour which tempted us to investigate the metabolism of chlorophyll and carotenoids in the needles of both yellow and green varieties of Douglas fir over a year period.

Khodasevich et al. (1978) reported that the yellow form of *Pinus sylvestris* had more carotenoids than the green one. However, our results have shown less chlorophyll and carotenoids in the needles of the yellow form than that in the needles of green variety. It may, therefore, be suggested that certain photobleaching of chlorophyll occurred in our experimental conditions, leading to the prevalence of yellow colour of the needles.

The investigations of Krivosheeva and Shavnin (1988) showed that oxygen evolution system of the needles of Siberian pine operated until November and was followed by the inactivation of primary photosynthetic processes. Two-year old needles proved more stable at low temperatures than the current needles. A stable pigment ratio of Douglas fir during the year suggested a normal photosynthesis process in the yellow form regardless of the temperature. In the green variety, though, it may be assumed that photosynthetic processes were more active in the summer.

In both the yellow and green variety of Douglas fir, needles of the preceding year had a higher pigment content. Similar results were obtained by Godnev et al. (1969)

who studied seasonal variations of pigments in spruce needles. It was noted that the Chl a/b ratio varied to a greater extent in the needles of a young yellow tree. This would suggest a greater variation of chlorophyll b in the needles of the yellow Douglas fir, without significant differences between the current and a year-old needles. These results are likely to point out to a similar metabolism of pigments in young and old needles of the same tree regardless of its age, thus supporting the idea of a genetically determined characteristic.

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## Rezi me

MILA BOGDANOVIĆ, GORDANA DRAŽIĆ, MILICA VUČKOVIĆ

**SEZONSKO VARIRANJE PIGMENATA U ČETINAMA DUGLAZIJE  
(*PSEUDOTSUGA TAXIFOLIA* BRITT.) U PRIRODNIM USLOVIMA**

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Zemun-Beograd

Ispitivano je sezonsko variranje pigmenta u četinama žute i zelene forme duglazije (*Pseudotsuga taxifolia* Britt.). Rezultati istraživanja su pokazali da starost stabala žute forme duglazije nema uticaja na sadržaj hlorofila i karotenoida u četinama. Četine prethodne godine sadrže više hlorofila i karotenoida kod svih ispitivanih stabala. Variranje sadržaja pigmenta u toku godine u četinama duglazije je neznantno kod žute forme. Četine zelenog varijeteta sadrže dvostruko više pigmenta od četina žute forme i više variraju. Diskutovana je međusobna zavisnost metabolizma hlorofila i karotenoida.



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Original scientific paper

PETAR D. MARIN

**NUTLET SCULPTURING OF SELECTED SPECIES FROM  
AJUGOIDEAE, SCUTELLARIOIDEAE AND STACHYOIDEAE  
(LAMIACEAE)**

Institute of Botany and Botanical Garden „Jevremovac”, Faculty of Biology,  
Belgrade

Marin, P. (1992-1993): *Nutlet sculpturing of selected species from Ajugoidea, Scutellarioideae and Stachyoideae (Lamiaceae)*. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 21 - 29.

Nutlet ornamentation of four species of *Ajuga* (*Ajugoideae*), two species of *Scutellaria* (*Scutellarioideae*), two species of *Marrubium*, *Prasium majus* and *Sideritis hyssopifolia* (*Stachyoideae*) were analysed, using scanning electron microscopy. The study provides additional stable micromorphological characters to include in classification at tribal and generic levels within the *Lamiaceae*.

Key words: *Lamiaceae*, *Ajuga*, *Prasium*, *Sideritis*, *Marrubium*, *Scutellaria*, nutlet sculpturing, SEM, taxonomy.

Ključne reči: *Lamiaceae*, *Ajuga*, *Prasium*, *Sideritis*, *Marrubium*, *Scutellaria*, skulpturiranost orašica, SEM, taksonomija.

## - INTRODUCTION

*Lamiaceae* is a large family with about 220 genera and more than 3500 species. The family has an almost cosmopolitan distribution and wide range of morphological diversity. The species are mainly herbaceous or shrubby growing usually in warm open habitats. Areas of major diversity of *Lamiaceae* species are Mediterranean region, Asia and Northern America, including Mexico (Hedger, 1992).

Many species are of economic importance (aromatic, medicinal or culinary herbs). A number of *Labiatae* species were investigated by botanists, from different aspects.

Taxonomy of this interesting and important family is still unresolved. First of all, there is not clear difference between *Lamiaceae* and *Verbenaceae* within the order *Lamiales* (Cronquist, 1981; Olmstead et al. 1992). In spite of several major reviews dealing with infrafamilial classification of *Lamiaceae* (Bentham, 1876; Briquet, 1895-97; Erdtman, 1945; Wunderlich, 1967; Cantino, 1992), many problems still remains. Those classifications are based on different characters (morphological, palynological, embryological). An attempt of application of chloroplast DNA sequences in *Asteridae* as well as *Lamiales* has been done (Olmstead et al. 1992).

In earlier reviews of any group of *Lamiaceae*, nutlet microcharacters were ignored or seldom mentioned in spite of their stability (Davis & Heywood, 1963). Recent studies of selected taxa, have shown that nutlet microcharacters can be used as additional taxonomic parameters in delimitation of such taxa (Krestovskaja, 1988; Husain et al. 1990; Marin et al. 1994).

In this survey nutlet ornamentation of selected genera from the *Ajugoideae*, *Scutellarioideae* and *Stachyoideae* as a potential valuable taxonomical character is described.

## MATERIAL AND METHODS

Plant material was collected from different parts of Yugoslavia. Some samples were collected from herbarium specimens from the Herbarium of Botanical Institute and Garden „Jevremovac“ in Belgrade, where voucher specimens are deposited. Places of collection of specimens are given in Tab. 1.

Tab. 1. – Places of collection of analysed species

Taxa	Place of collection
Subfam. <i>Ajugoideae</i>	
<i>Ajuga reptans</i> L.	Beograd (Yu)
<i>Ajuga genevensis</i> L.	Kukavica (Yu)
<i>A. laxmanii</i> (L.) Benth	Lisac planina (Yu)
<i>A. chamaepitys</i> (L.) Schreber	Bistrica (Yu)
Subfam. <i>Scutellarioideae</i>	
<i>Scutellaria alpina</i> L.	Košice (Czech Republic)
<i>Scutellaria altissima</i> L.	Fruška Gora (Yu)
Subfam. <i>Stachyoideae</i>	
<i>Prasium majus</i> L.	Barcelona (Spain)
<i>Sideritis hyssopifolia</i> L.	Bern (Swiss)
<i>Marrubium incanum</i> Desr.	Barcelona (Spain)
<i>Marrubium vulgare</i> L.	Barcelona (Spain)

For scanning electron microscopy (SEM) at least three samples of mature nutlets were used. The samples were previously observed by light microscope. The nutlets were mounted on stubs and coated with 30 nm of gold - palladium (85:15) in a JEOL JEE 4B vacuum evaporator and observed with a JEOL JSM T.35 scanning electron microscope.

## RESULTS AND DISCUSSION

The nutlet sculpturing of 10 species from selected genera of *Ajugoideae*, *Scutellarioideae* and *Stachyoideae* subfamilies are presented (Figs 1-27). Intrafamilial classification was according to Wunderlich (1967).

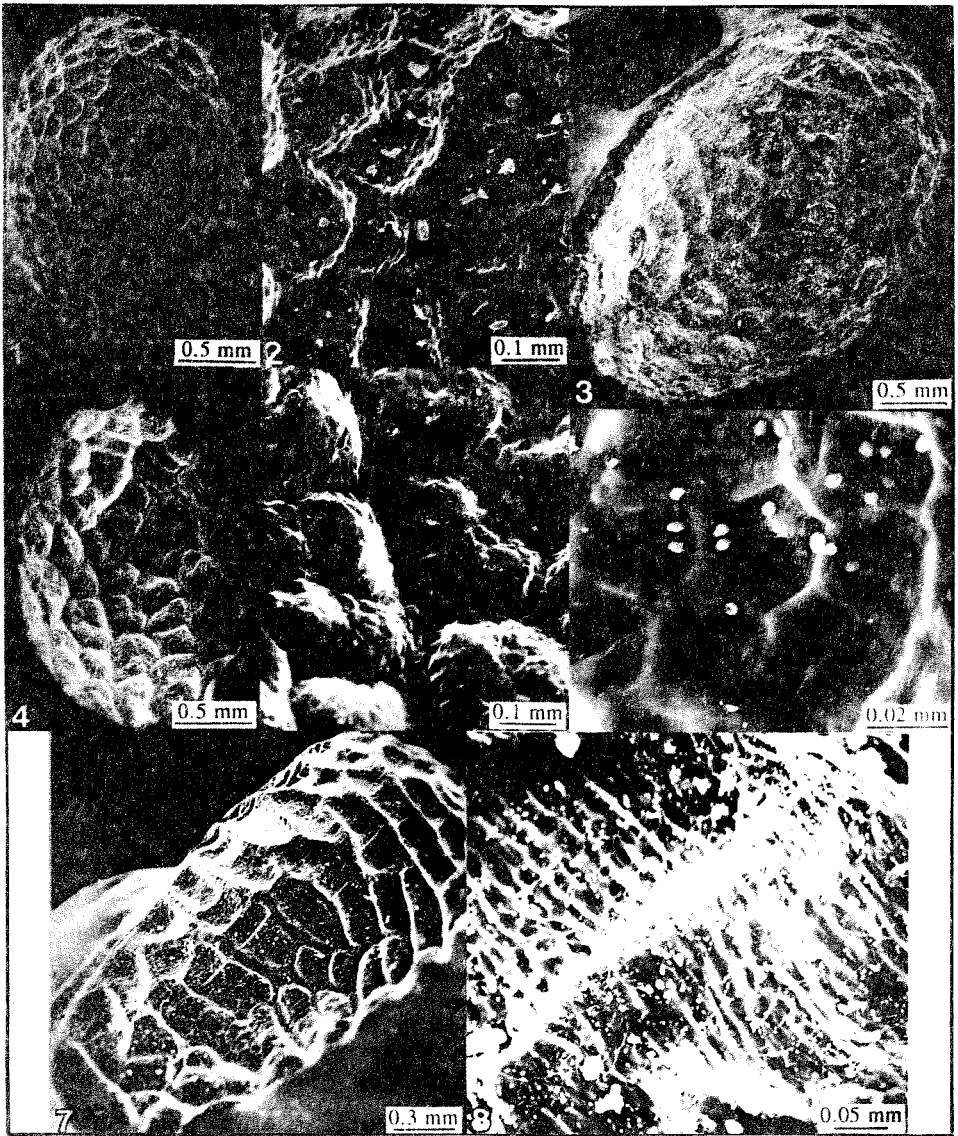
From the *Ajugoideae* subfamily, four *Ajuga* species are analysed (Figs 1-8). The *Ajuga* nutlets showed quite high similarities in size, shape and ornamentation between the species. Especially, *Ajuga genevensis* (Figs 1, 2), *A. laxnanii* (Fig. 3) and *A. reptans* (Figs 4-6) were very similar with characteristic more or less ovate or angular sunken fields. The epicarp cells were 5-6 angular with thick walls. The ornamentation of *A. chamaepitys* (Figs 7, 8) is also with sunken fields, but they are transversally elongated. The epicarp cells are striate. Species from the first group showed similarities with some *Teucrium* species (*T. polium*, *T. bracteatum*) (Marin et al., 1994). *Teucrium* is close genus to *Ajuga*, also with some similarities in nutlet micromorphology. But, according to micromorphological, as well as other characters, *Teucrium* is very heterogenous. In number of species oil glands were found on nutlets, and some of them consists of trichomes. Those structures were not found in any *Ajuga* species analysed in this paper. Some authors suggest to establish a new subfamily *Teucroioideae*, which includes *Teucrium*, some genera from *Ajugoideae* (but not *Ajuga*) and some genera which were so far placed in *Verbenaceae* family (Cantino et al., 1992). Micromorphological data can not resolve completely these problems. However, there are some similarities in general microcharacters between these to genera which shows possibility of their close relationships.

Recent investigation of fatty acid composition in *Ajugoideae* nutlets showed high similarities of *Teucrium* and *Ajuga* (Marin et al., 1991). The main constituents in both genera were octadecadienoic (18:2) and octadecatrienoic acid (18:3).

From the *Scutellarioideae* subfamily, two *Scutellaria* species were analysed (Figs 9-16). According to nutlet ornamentation, *Scutellaria* species are very distinguishable from all other analysed genera. *Scutellaria altissima* possess very characteristic bumps, which are multicelled (Figs 9-12). The cells forming those bumps are very small, with more or less expressed apices. In the center of bumps small trusses of trichomes were present. In *Scutellaria alpina* similar pattern was found. The bumps were not expressed, but a number of trichome trusses were dispersed all over the nutlets (Figs 13-16). Trichomes were long and unicellular. In addition, very small papillae were distributed on epicarp cells of nutlets. (See higher magnification, Figs 15, 16).

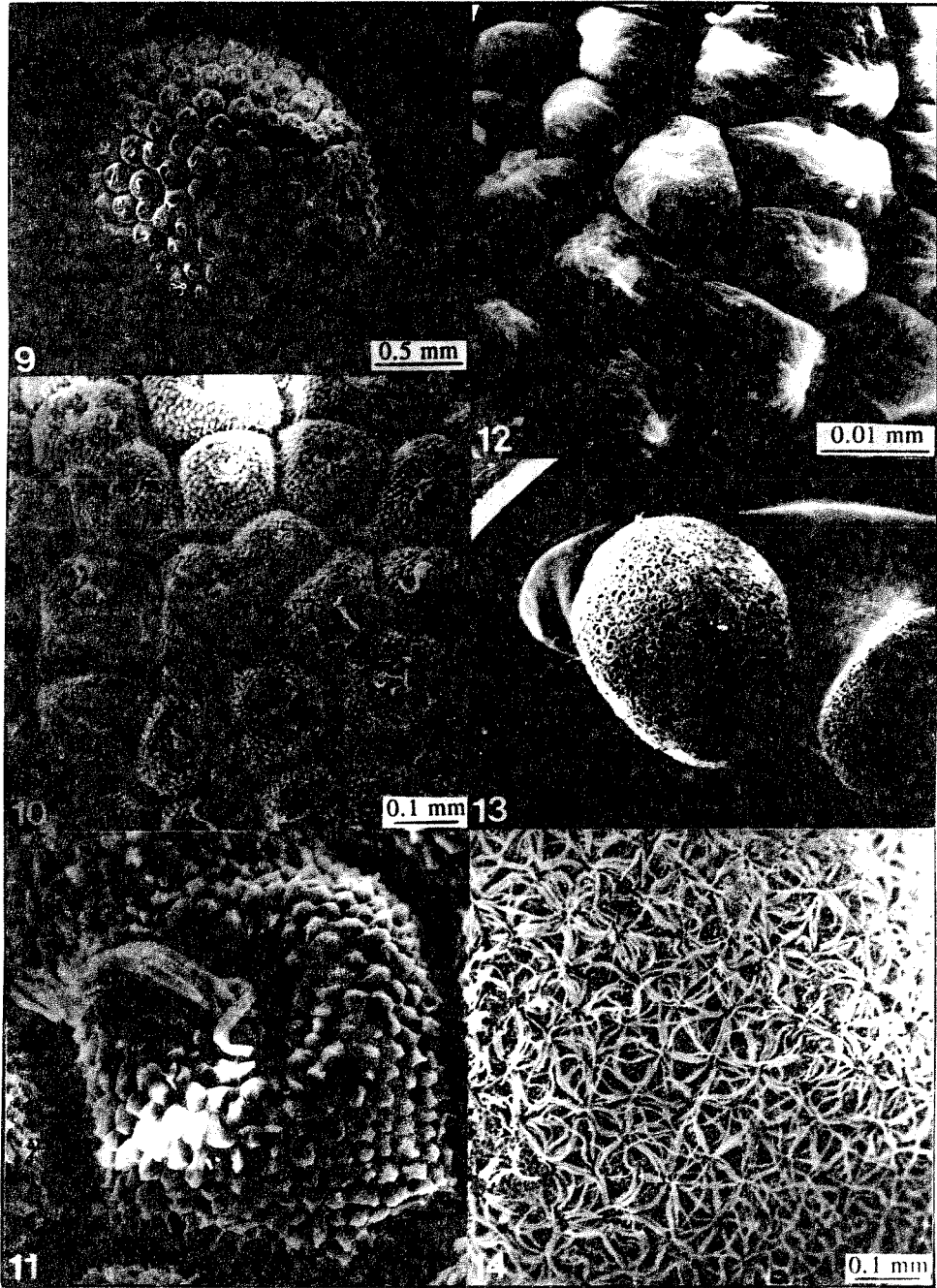
Nutlet lipid analysis in four *Scutellaria* species showed quite high uniformity in fatty acid composition (Marin et al., 1991). The octadecadienoic and octadecenoic were dominant fatty acids. Content of the octadecatrienoic acid was very low.

Recent investigation of global taxonomy of *Scutellaria* showed that nutlet morphology varies enormously within the sections (Paton, 1990). Some of species showed papillatae nutlets, in others the papillae were fused into a basal wing. In some species the hooks on the nutlet surface are present. Paton (1992) was hypothesized that the morphology of the fruiting calyx in *Scutellaria* is subjected to two main selection pressures, to the protection of immature fruit and the dispersal of the mature nutlets. According to this author, nutlet ornamentation probably plays a role in dispersal (adaptation to water, wind or animal dispersal). He suggested that nutlet sculpturing is more variable than calyx morphology because fewer constraints are placed on its evolution during development.

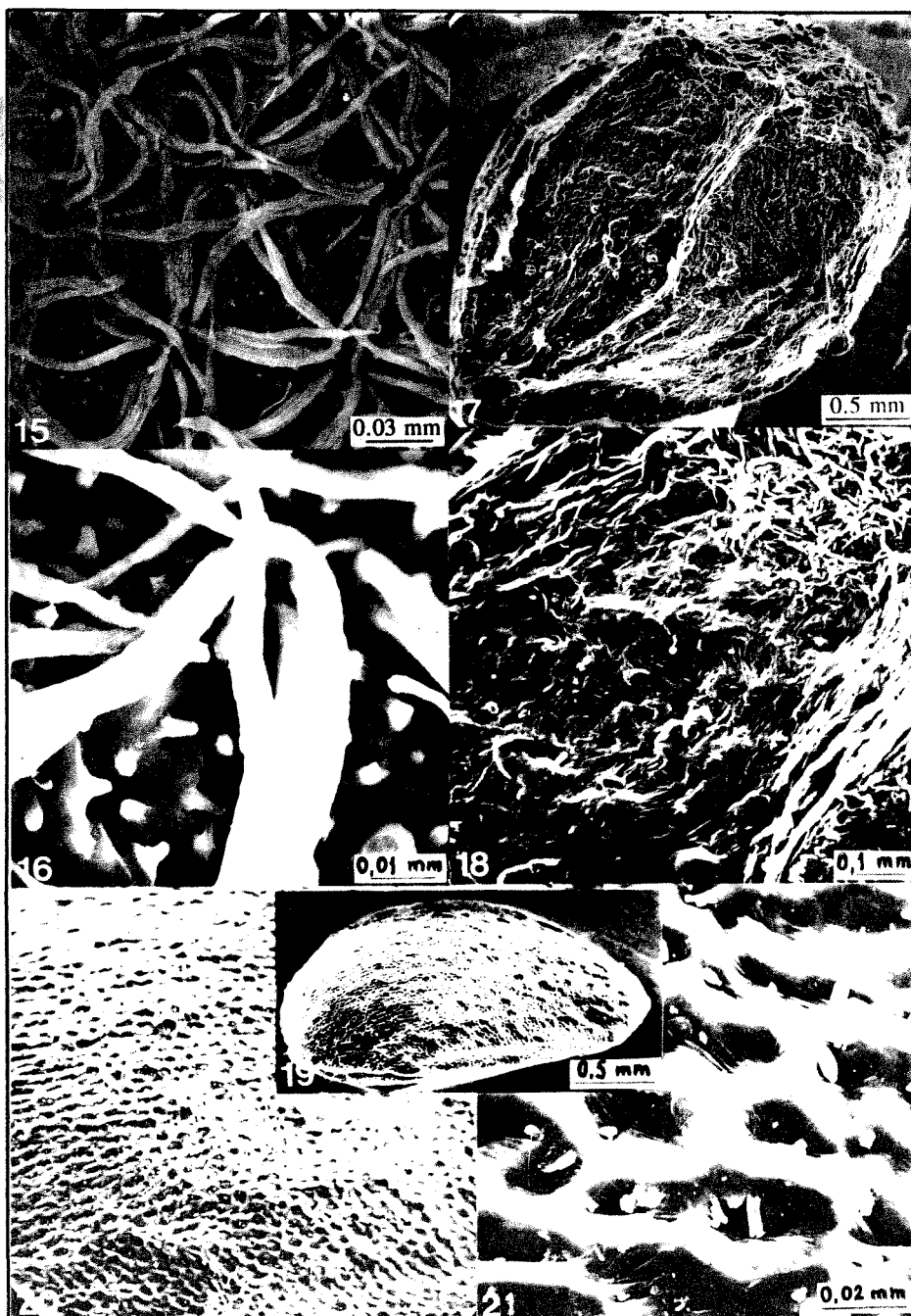


Figs. 1 - 2. - *Ajuga genevensis*; 3. - *A. laxmanii*;  
4 - 6. - *A. reptans*; 7 - 8. - *A. chamaepitys*

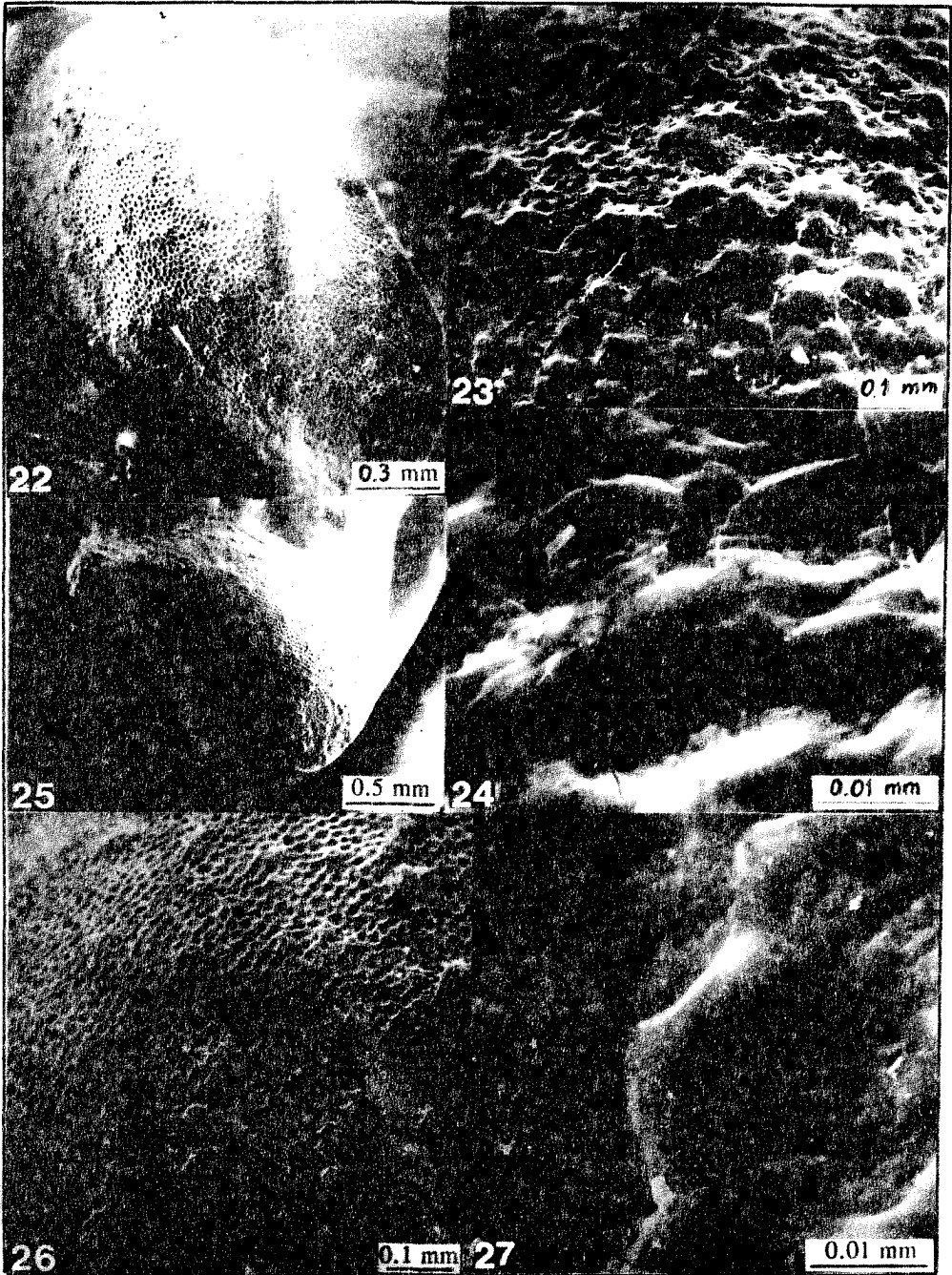
It should be mentioned that *Scutellaria* is subcosmopolitan genus consisting about 360 species, which are heterogenous because of great diversity in their ecological habitats. Probably, it is very old and „well defined” genus, derived from other taxa of *Lamiaceae* (separate subfamily!). The taxonomical significance of nutlet ornamentation, in spite of their great diversity, can not be ignored. The problem is how to explain



Figs 9 - 12. - *Scutellaria altissima*; 13 - 14. - *S. alpina*



Figs. 15 - 16. - *Scutellaria alpina*; 17 - 18. - *Prasium majus*; 19 - 21. - *Sideritis hyssopifolia*



Figs 22 - 24. - *Murrubium incanum*  
Figs 25 - 27. - *M. vulgare*

such a diversity, their evolution and phylogenetical relationship. However, explanation of less variation in calyx morphology and their probable phylogenetic relationship is much easier and more evident.

From the *Stachyoideae* subfamily (sensu Wunderlich) four selected species were analysed. From the tribe *Prasieae*, *Prasium majus* was analysed. Nutlet ornamentation of this species has shown nonspecialized pattern (Figs 17, 18). It could be explained by the fact that *Prasium* species are endozoochorous. The exocarp become fleshy (endocarp becoming sclerotic) and attracting for the animals (Bouman et Meeuse, 1992). The mature nutlets are dried and as result of drying of fleshy exocarp become crowded.

From the tribe *Marrubieae* *Sideritis hyssopifolia* and two *Marrubium* species are presented. Nutlets of the *Sideritis hyssopifolia* are characterised by low protrusions sparsely distributed. The epicarp cells were angular and thick-walled (Figs 19-21). Nutlet ornamentation of *Marrubium incanum* (Figs 22-24) and *M. vulgare* (Figs 25-27) showed very high similarities. Low protrusions formed by one or few cells were sparsely distributed on nutlets. Epicarp cells were angular or ovate, thick - celled. In spite of the facts that these species belongs to the different genera, nutlet ornamentation showed very similar structures. It could be suggested that those genera are very close related.

*Marrubium* and *Sideritis* showed similar pattern in fatty acid composition in nutlets (Marin et al., 1992). In both genera the dominant fatty acid was linoleate. *Sideritis* species consists of slightly higher content of this acid, and lower content of oleate.

Upon the whole the various forms of dispersal in *Lamiaceae* family do not reflect the taxonomic relationships (Bouman & Meeuse, 1992). Those different types of dispersal are connected with different ecological factors. In a single genus it is possible to find different types of primary or secondary dispersal. In most cases the dispersal is result of parallelism and convergence.

However, nutlet ornamentation of species analysed in this survey showed that these parameters have to be included in further taxonomical revisions of any taxa of *Lamiaceae*. It seems that these microcharacters are of the best taxonomical value at generic and tribal level, and in some cases in infrageneric classification of *Lamiaceae*.

In addition, results of recent investigation of fatty acid composition suggest that these parameters can be used as taxonomic markers at different levels within the family.

#### ACKNOWLEDGEMENTS

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Rezi me

PETAR D. MARIN

SKULPTURIRANOST ORAŠICA KOD ODABRANIH VRSTA IZ AJUGOIDEAE,  
SCUTELLARIOIDEAE I STACHYOIDEAE (LAMIACEAE)

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U ovom radu izvršena je mikromorfološka analiza ornamentacije orašica kod 10 odabranih vrsta iz različitih podfamilija (*Ajugoideae*, *Scutellarioideae* i *Stachyoideae*) familije *Lamiaceae*, pomoću skenirajućeg elektronskog mikroskopa (SEM). Dobijeni rezultati su pokazali da ovi parametri treba da budu uključeni i ravnopravno tretirani prilikom taksonomskih revizija različitih taksona kod usnatice. Izgleda da ovi mikrokarakteristi imaju najveću vrednost kao markeri na nivou tribusa i rodova, a u nekim slučajevima i u infrageneričkoj klasifikaciji unutar familije *Lamiaceae*.



UDC 581.4 : 582.751.9 (497.11)  
Original scientific paper

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MARIN

## ANATOMSKA GRAĐA VEGETATIVNIH ORGANA VRSTE *DICTAMNUS ALBUS* L.

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Petković, B., Tatić, B., Ilijin-Jug, M., Marin, P. (1992-1993): *Anatomy of vegetative organs of Dictamnus albus* L. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beograd, Tom XXVI-XXVII, 31 - 39.

This research shows anatomy of vegetative organs of *Dictamnus albus* L. species. The samples of this species are collected at Košutnjak. Root, rhizome, stem and leaves anatomy has been done.

Key words: anatomy, vegetative organs, *Dictamnus albus*.

Ključne reči: anatomska građa, vegetativni organi, *Dictamnus albus*.

### UVOD

Vrsta *Dictamnus albus* L. je u narodu poznata pod imenom jasenak. To je višegodišnja zeljasta biljka koja svojom visinom (60-120 cm), perastim listovima (u gornjem delu stabla) nalik na listove jasena (otuda naziv jasenak) i veoma lepim i opojnim cvetovima privlači pažnju ljudi. Zbog lekovitih svojstava korena, lista, cveta i semena (Jakovljević, 1962; Tucakov, 1984) koristi se u narodnoj medicini. Radi toga se ova vrsta nekontrolisano bere, prenosi u bašte, kida i uništava, te je tako dosta proređena i ozbiljno ugrožena.



Fig. 1. – Areal vrste *Dictamnus albus* L.

Areal of *Dictamnus albus* L.

Areal vrste *D. albus* (Fig. 1) obuhvata južnu i centralnu Evropu, srednju i zapadnu Aziju istočno od severne Kine. Amurai Himalaja (Diklić, 1973; Šilić, 1977).

S obzirom da je anatomski niestražena, ovim smo istraživanjima obuhvatili vegetativne organe (koren, rizom, stablo, list), dok će reproduktivni organi biti predmet naših daljih istraživanja ove vrste.

#### MATERIJAL I METODE

Uzorci vrste *D. albus* L., prikupljeni su sa njenog staništa na Košutnjaku i to u vreme cvetanja. Originalni uzorci analizirane vrste deponovani su u Herbarijumu Botaničke bašte u Beogradu.

Sakupljeni materijal je fiksiran u FAA fiksativu, a za pravljenje serija trajnih anatomskih preseka korišćena je parafinska metoda (Johnsen, 1940). Preparati su sečeni na rotacionom mikrotomu, debljine od 10-20  $\mu\text{m}$ , zatim bojeni kombinacijom boja safranin i svetlo zeleno i snimani na Reichert-ovom mikroskopu Diaster<sup>TM</sup>.

#### REZULTATI I DISKUSIJA

Anatomska istraživanja vegetativnih organa (lista, stabla, korena i rizoma) dala su sledeće rezultate.

List je dorzoventralne građe. Na epidermusu lica i naličja nalazi se sloj kutikule. Debljina kutikularnog sloja na gornjoj strani je nešto veća od istog sloja sa naličja (Fig. 2A). Merenjem je utvrđeno da debljina kutikule na površini lista iznosi u proseku 3,5  $\mu\text{m}$ , dok je na naličju sloj kutikule debljine 3,1  $\mu\text{m}$ .

Epidermis je jednoslojan, ćelije epidermisa su pravougaonog oblika, dužine 33  $\mu\text{m}$ , a visine 20  $\mu\text{m}$  na licu lista, dok su na naličju nešto sitnije, dužine 30  $\mu\text{m}$  i visine 18  $\mu\text{m}$  u proseku. Na samim krajevima lista koji su zaobljeni i blago povijeni naniže uočavaju se krupnije epidermalne ćelije (Fig. 2B). Veoma je interesantno i to da se skoro u svakoj epidermalnoj ćeliji nalaze korpne druze kalcijum oksalata.

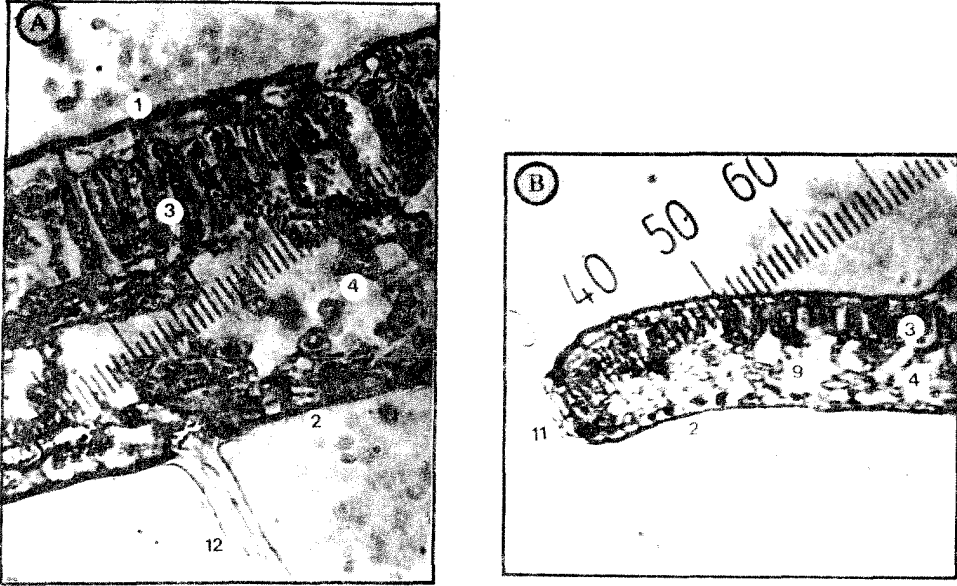


Fig. 2. – Poprečni presek lista (A) i ivice lista (B): 1 – epidermis i kutikula lica lista; 2 – epidermis naličja lista; 3 – palisadno tkivo; 4 – sunderasto tkivo; 5 – ksilem; 6 – floem; 7 – mehaničko tkivo; 8 – bočni provodni snopić; 9 – sekretorni kanal; 10 – kompaktna masa parenhima; 11 – krupnije ćelije epidermisa na ivicama lista; 12 – stome; 13 – jednočelijske dlake.

Cross section of leaf (A) and leaf margin (B): 1-adaxial epidermis; 2-abaxial epidermis; 3-palisade parenchyma cells; 4-spongy parenchyma cells; 5-xylem; 6-phloem; 7-fibres; 8-lateral vascular bundle; 9-secretory canal; 10-compact mass of parenchyma; 11-epidermal cells on leaf margins; 12-stoma; 13-epidermal hair.

Stome se nalaze sa obe strane lista, a više ih ima na naličju. Uvučene su.

Mezofil lista jasno je diferenciran na palisadno i sunderasto tkivo. Palisad je jednoslojan i nalazi se samo na licu lista. Ćelije palisada su izrazito cilindrične, bogate hlorofilom i jako rastresite (Fig. 2A). Visina ovih ćelija iznosi 45  $\mu\text{m}$ , a debljina u proseku 10  $\mu\text{m}$ . Sunderasto tkivo je približno iste širine kao palisad i sastoji se od sitnih parenhimskih ćelija, nepravilnog oblika sa krupnim intercelularima, što ovaj sloj čini mnogo rastresitijim od prethodnog. U mezofilu lista uočavaju se izlučne šupljine (žlezde) ispunjene kapima eteričnog ulja i kristali kalcijum oksalata (Fig. 3A).

U glavnim i bočnim nervima provodni snopić je po tipu kolateralan i zatvoren, okružen parenhimskim i kolenhimskim ćelijama ili grupama likinih vlakana. Centralni provodni snopić glavnog nerva okružen je kompaktnom masom parenhima (Fig. 3B). Ćelije ovog sloja su nešto sitnije od okolnih ćelija osnovnog parenhima. Između osnovnog parenhima i epidermisa, kako na licu, tako i na naličju, nalazi se mehaničko tkivo sa ćelijama jako zadebljanih zidova. Ovaj sloj se uočava samo u području glavnog listnog nerva.

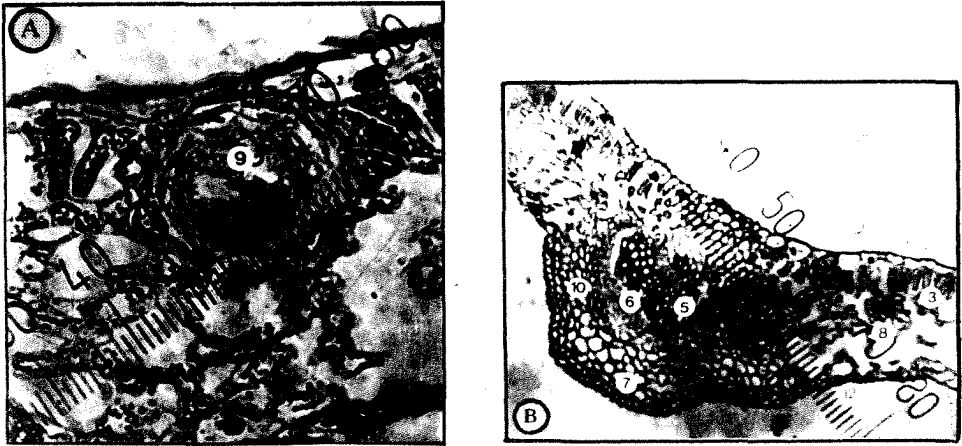


Fig. 3. – Poprečni presek lista sa žlezdama (A) i u nivou glavnog nerva (B): 1-epidermis i kutikula lica lista; 2-epidermis naličja lista; 3-palisadno tkivo; 4-sunderasto tkivo; 5-ksilem; 6-floem; 7-mehaničko tkivo; 8-bočni provodni snopić; 9-sekretorni kanal; 10-kompaktna masa parenhima; 11-krupnije ćelije epidermisa na ivicama lista; 12-stome; 13-jednoćelijske dlake.

Cross section of leaf glands (A) and central bundle (B): 1-axial epidermis; 2-abaxial epidermis; 3-palisade parenchyma cells; 4-spongy parenchyma cells; 5-xylem; 6-phloem; 7-fibres; 8-lateral vascular bundle; 9-secretory canal; 10-compact mass of parenchyma; 11-epidermal cells of leaf margins; 12-stoma; 13-epidermal hair.

**Stablo.** Anatomska građa stabla proučavana je posmatranjem i analizom poprečnih preseka i to u cvetnom regionu (vršni deo stabla, tj. „mlade stablo”) i u regionu stabla bliže osnovi („starije stablo”).

„Mlade stablo” vrste *Dictamnus albus* L. odlikuje se primarnom gradom i na njemu se razlikuju tri osnovne zone: epidermis, primarna kora i centralni cilindar (Fig. 4).

Epidermis stabla je jednoslojan, ćelije su gusto zbijene, a na površini se nalazi deo sloj kutikule. Uočavaju se i jednoćelijske dlake, dužine u proseku 240  $\mu\text{m}$ , a širine 15  $\mu\text{m}$ . Između epidermalnih ćelija zapažaju se nepravilno rasporedene stome.

Ispod epidermisa je višeslojna primarna kora sastavljena od parenhimskih ćelija sa dosta hloroplasta. Ćelije ove zone su loptastog oblika, dosta su krupne, tankih ćelijskih zidova i uzanih intercelulara. U primarnoj kori ispod sloja parenhima pruža se zona sklerenhimskih vlakana, jako zadebljanih i lignifikovanih zidova. Ovakva zona mehaničkog tkiva ne obrazuje kontinuirani prsten, već se javlja u vidu izolovanih traka iznad provodnih elemenata.

Centralni cilindar zauzima unutrašnjost stabla i u njemu se nalaze provodna tkiva raspoređena u vidu dva kontinuirana i koncentrična prstena, pri čemu spoljašnji prsten čini floem, a unutrašnji ksilem. Ovakva građa odgovara tipu stabla bez provodnih snopića. Floemski deo je slabije razvijen i čine ga sitaste cevi, ćelije pratilice i floemski parenhim. Između floema i ksilema pruža se kambijalna zona u obliku prstena. Provodni elementi ksilema su radialno raspoređeni, a kroz ceo centralni cilindar uočavaju se sržni zraci u vidu jednoslojnih traka.

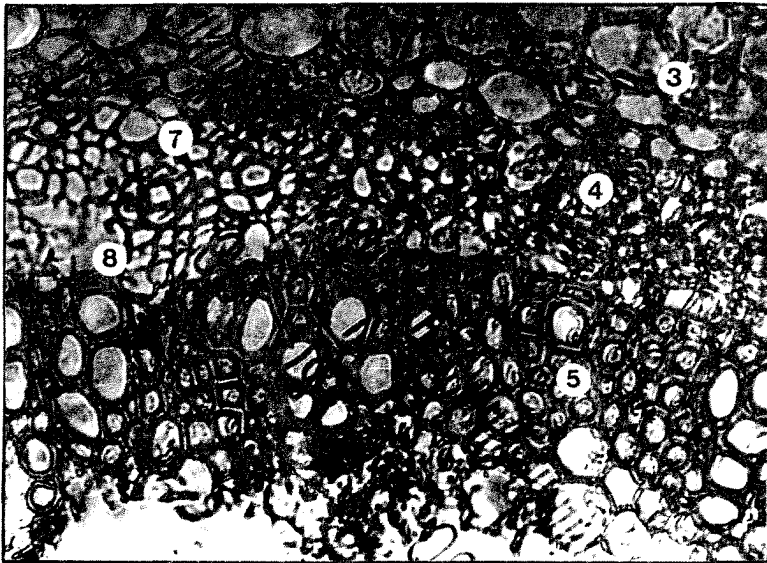
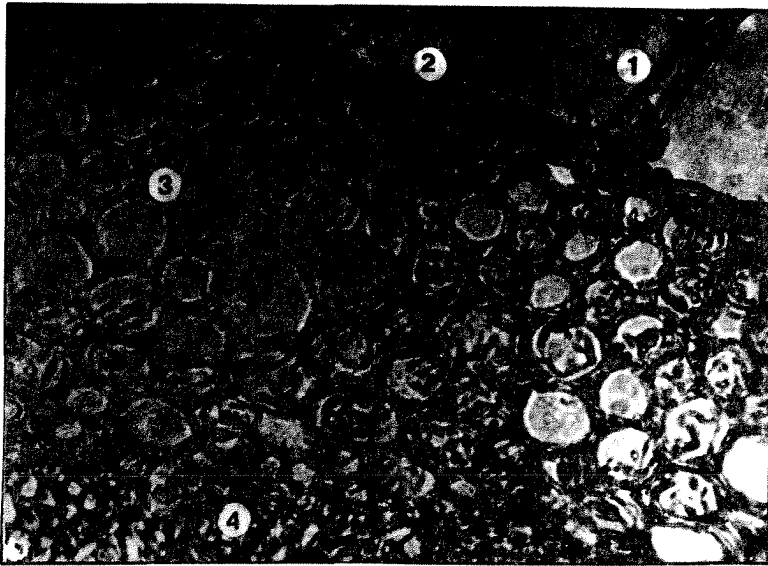


Fig. 4. – Poprečni presek „mladeg stabla”

1-dlaka; 2-epidermis; 3-parenhim primarne kore; 4-floem; 5-ksilem; 6-srž; 7-mehaničko tkivo; 8-kambijum.

Cross section of „younger stem”.

1-hair; 2-epidermis; 3-parenchymatous cortex; 4-phloem; 5-xylem; 6-pith; 7-fibres; 8-cambium

U središnjem delu stabla nalazi se dobro razvijena srž. Čelije srži su krupne, prečnika do 40  $\mu\text{m}$ , tankih su zidova i loptastog oblika. Što su bliže samom centralnom delu, čelije srži su sve krupnije. U poređenju sa „mladim” delom stabla, kod „starijeg” stabla se zapažaju izvesne razlike u anatomske gradbi, koje nastaju kao posledica sekundarnog debljanja. Za razliku od „mladog stabla” primarne grade, kod „starijeg” odsustvuju dakle u epidermisu, kutikula je jače razvijena, a elementi primarne unutrašnjosti stabla sekundarni ksilem postepeno prelazi u primarni. Centralni cilindar ispresecan je mnogobrojnim sekundarnim sržnim zracima (Fig. 5).

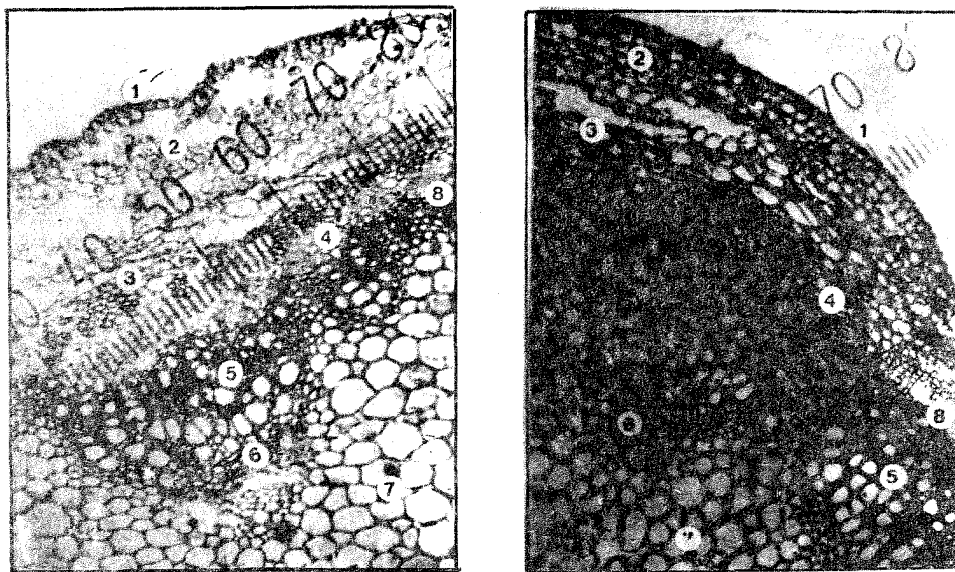


Fig. 5. – Poprečni presek „starijeg stabla”

1-epidermis; 2-parenhim primarne kore; 3-mehaničko tkivo; 4-sekondarni floem; 5-sekondarni ksilem; 6-primarni ksilem; 7-srž; 8-kambijum.

Cross section of „older stem”

1-epidermis; 2-parenchymatous cortex; 3-fibres; 4-secondary phloem; 5-secondary xylem; 6-primary xylem; 7-pith; 8-cambium.

**Koren** (Fig. 6). Na površini korena nalazi se periderm, kojeg čine feloderm, felogen i slojevi plute. Pluta je moćno razvijena i njena debljina iznosi u proseku 35 mm. Čelije feloderma sadrže hloroplaste.

Parenhim primarne kore nastavlja se odmah ispod periderma i odlikuje se dosta krupnim parenhimbim čelijama, koje sadrže puno skroba, drže kalcijum oksalata i kapi ulja.

U zoni centralnog cilindra se uočavaju provodni elementi raspoređeni u radijalnim nizovima. Sekundarni floem je odvojen od sekundarnog ksilema kambijalnim prstenom. Široke traheje i traheidi su izmešani sa mehaničkim elementima, što korenu daje potrebnu čvrstinu. Jednoslojni sržni zraci se jasno uočavaju.

**Rizom.** Anatomska gradba rizoma razlikuje se od nadzemnog dela stabla (Fig. 7).



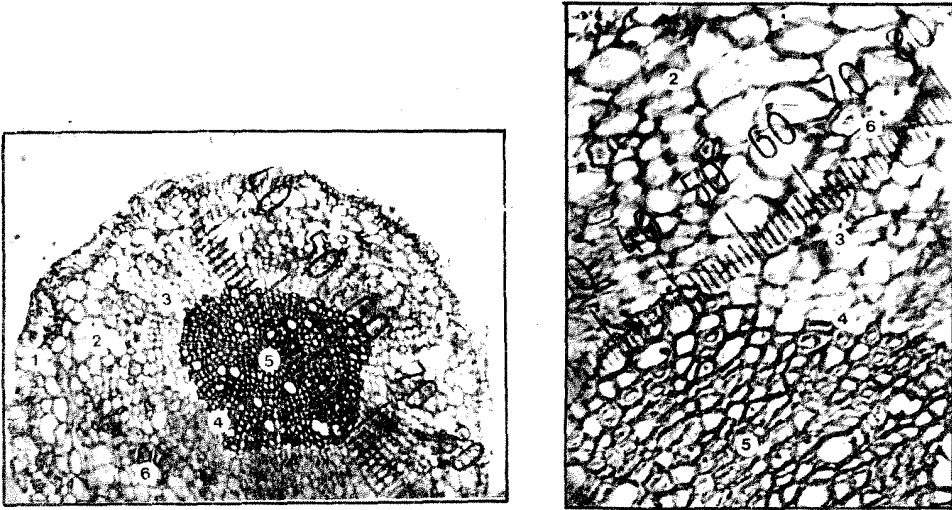


Fig. 6. – Poprečni presek korena  
1-periderm; 2-parenhim kore; 3-floem; 4-kambijum; 5-ksilem; 6-kapi ulja  
Cross section of root  
1-cork; 2-parenchymatous cortex; 3-phloem; 4-cambium; 5-xylem; 6-oil drops.

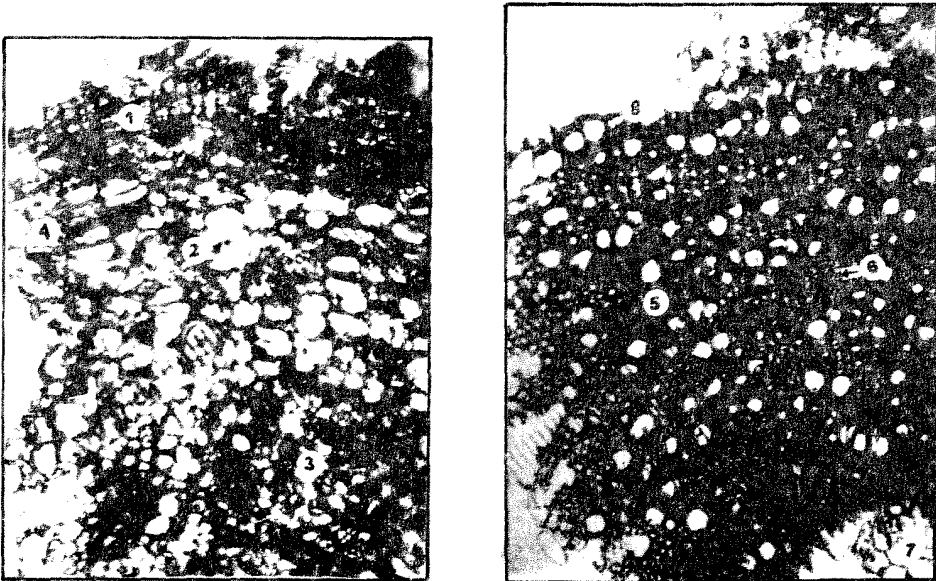


Fig. 7. – Poprečni presek rizoma  
1-kora; 2-parenhim kore; 3-sekundarni floem; 4-kapi ulja; 5-sekundarni ksilem; 6-  
sržni zrak; 7-srž; 8-kambijum.  
Cross section of rhizome  
1-cork; 2-parenchymatous cortex; 3-secondary phloem; 4-oil drops; 5-secondary xylem; 6-medullary ray; 7-  
pith; 8-cambium.

Epidermis je zamenjen dobro razvijenim peridermom, što je posledica sekundarnog debljanja.

Primarna kora je u odnosu na nadzemni deo stabla kompaktnija i zauzima veću površinu. Parenhimske ćelije ovog sloja zbijene su i gusto ispunjene skrobnim zrnima. Ovakva građa kore je u skladu sa njenom specifičnom funkcijom, tj. magacioniranjem hranljivih materija i preživljavanjem nepovoljnih uslova.

Centralni cilindar zauzima daleko manji prostor u odnosu na stablo. Elementi floema i ksilema su raspoređeni u vidu koncentričnih krugova. Sekundarni floem se javlja u tankom sloju, dok je sekundarni ksilem veoma dobro razvijen. Kambijalni prsten je izrazitiji nego u stablu. U sredini rizoma nalazi se srž, svedena na veoma malu zonu u odnosu na srž stabla. Parenhimske ćelije srži ispunjene su sitnozrnim skrobom.

## ZAKLJUČAK

Glavni akcenat istraživanja bio je na proučavanju vegetativnih organa, listova, stabla, korena i rizoma.

List je tipično građen i pokazuje kseromorfni karakter. Pokorično tkivo je predstavljeno jednoslojnim epidermisom, stome su uvučene i javljaju se sa obe strane lisne ploče. Provodni snopići su po tipu kolateralni i zatvoreni, okruženi kompaktnom masom parenhima.

Anatomska građa stabla proučavana je posmatranjem poprečnih preseka u cvetnom regionu i regionu stabla bliže osnovi. Stablo u cvetnom regionu, odn. „mladi deo stabla” se odlikuje primarnom građom, dok donji „stariji region” ima karakteristike sekundarne građe.

Poprečni presek korena pokazuje sekundarnu građu, odn. presek zahvata stariji deo korena. Na površini je moćno razvijen sloj periderma. Anatomska građa rizoma pokazuje sličnu građu sa nadzemnim delom stabla, a u skladu je sa njegovom specifičnom funkcijom.

Ovakva anatomska građa vrste *D. albus* je svakako u korelaciji sa njenim fiziološkim i ekološkim osobinama.

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### Summary

BRANIMIR PETKOVIĆ, BUDISLAV TATIĆ, MIRJANA ILIJIN-JUG, PETAR MARIN

#### ANATOMY OF VEGETATIVE ORGANS OF *DICTAMNUS ALBUS* L.

Institute of Botany and botanical garden „Jevremovac”, Faculty of Biology, Belgrade

The accent of this research was the anatomy of the root, rhizome, stem and leaves.

Leaf is typical build and it has a xeromorphic character. Epidermis is one layer and stomates are situated on the adaxial and abaxial surface. Vascular bundles are colateral surrounded with parenchyma cells.

Stem anatomy is investigated at the clovery region and at the region close to the stem base. First one shows primary built, the other shows secondary built.

Root anatomy shows the secondary built. Developed periderm is at the surface.

Rhizome anatomy is similar to the stem and its anatomy is related to the function.

Anatomy of *D. albus* is related to its physiological and ecological characteristics.



UDC 581.522.5 : 582.635.1 (497.11)  
Original scientific paper

TATJANA GRABELJŠEK

### SOME ECOPHYSIOLOGICAL AND MORPHO-ANATOMICAL CHARACTERISTICS OF THE SPECIES *ZELKOVA CRENATA* SPACH.

Faculty of Veterinary Medicine, University of Belgrade

Grabeljšek, T. (1992-1993): *Some ecophysiological and morpho-anatomical characteristics of the species Zelkova crenata Spach.* – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 41 - 48.

The study concerns the investigations of water regime factors, transpiration, quantity of water in leaves, water saturation deficit and osmotic pressure of a cell sap of the species *Zelkova crenata* Spach. At the same time there have been observed some of the microclimatic factors: light intensity, solar radiation intensity, air and soil temperature, air and soil relative humidity and evaporation. The study also involves the analysis of some morpho-anatomic characteristics of the leaves of this species. The investigations have been carried out in the Botanical Garden in Belgrade.

Key words: *Zelkova crenata* Spach., water relations, leaf anatomy, ecological adaptations, Tertiary relict.

Ključne reči: *Zelkova crenata* Spach., vodni režim, anatomska grada lista, ekološke adaptacije, tercijarni relikat.

## INTRODUCTION

Although *Zelkova crenata* Spach. is considered to be an introduced species, the history of genus and species shows its ancient existence on the territory of Europe. During tertiary the genus *Zelkova* Spach. was spread through the whole Europe. On the basis of Walter's data (Walter & Straka, 1954, 1970), it could be found in upper miocene as eastern-Asia element included in Laurisilvae vegetation, together with 300 other species from *Magnoliopsida* class. During pliocene it was still present in Europe at the territory of today's Germany. It survived during the ice Age in pleistocene, so we can find it in interglacial period Ris-Virm, too. Its present domicile as tertiary thermophile genus is the region of Caucasus, Northern Persia, China nad Japan. Belonging to the family of *Ulmaceae* Mirb., genus *Zelkova* Spach. is presented by 6 species in the world today.

*Zelkova crenata* Spach. is a woody species, 20-25 m high, which was spread through the whole Europe during tearing terciar (Jovanović, 1967). Its today's habitat is in the warm oak forest at Caucasus and surrounding regions, and it could be found also in depression and at alluvium (Jovanović, 1967). According to Turill's data (Turill, 1929) today, in the region of Europe, taxonomically, the most closely related to it is *Zelkova abelicea* Boiss., which is distributed at Cyprus and Crete. Today, people grow this species in the great part of Europe as ornamental plant, and in all botanic gardens, so that it could be found in Belgrade Botanic garden.

Since the matter of our work is terciar species which natural habitat was in Southern Europe, we considered that the investigation of its adaptation to the present habitat conditions was necessary, if we wanted to understand the surviving mechanisms of this species, which vanished from Europe during the Ice Age.

For complete understanding of the ecology of plant species it is necessary to investigate its water regime. A water regime of a plant is influenced by both external factors of the environment and numerous internal morpho-anatomic and physiological factors.

The investigations of water regime obtains the analysis of the transpiration, quantity of water in leaves, water saturation deficit and osmotic pressure of a cell sap. The influence of a external factors such as the temperature and the humidity of the air and soil, the evaporation, the light intensity and solar radiation have been analyzed too. The morpho-anatomical investigations have been carried out on the leaves of the species *Zelkova crenata* Spach.

## MATERIALS AND METHODS

The investigations of water regime and morpho-anatomical structure of the *Zelkova crenata* Spach. leaves, were carried out at the Botanical Garden in Belgrade. The tree wich was used for the experiment is about 70 year old. It grows in a dense structure that is made by the surrounding trees. Since *Zelkova crenata* Spach. is a sciophyte, these conditions are completely suitable to it.

The investigations of water regime were carried out in may, july and september 1989. Daily dynamics was controlled every two hours, from 8 to 18 h. Transpiration was determined by the method of Stocker (1929, 1956), and it is presented in mg of transpired water on g of fresh leaf weight during 1 min. period (mg/g/min). The quantity of water in leaves and water saturation deficit were obtained by the method

of Stocker (1929), Slatyer (1960) and Slavik (1974), and expressed in percentage (%). The osmotic pressure of a cell sap was obtained by the method of Walter (1931, 1970) at semimicroosmoeter made by Knauer, and is expressed in bars.

Simultaneously it has been investigated the influence of some external factors such as: solar radiation intensity ( $J/cm^2/min$ ), light intensity (lux), temperature ( $^{\circ}C$ ) and relative humidity (%) of the air at the height of 100 cm and the soil temperature in the depth of 20 cm. The relative humidity of the soil layer in the depth of 100 cm has been analyzed once a day. These microclimatic investigations were carried out by microclimatic station according to Janković, M. M. (1971).

Water regime dynamics of *Zelkova crenata* Spach. and the dynamics of microclimatic conditions of its habitat, are presented by minimal, maximal and mean daily values. Seasonal dynamics was observed by following the average daily values in may, july and september.

The anatomical analysis of the leaves of the observed species was carried out at preparations by standard paraffin embedding procedure and double stained by safranin and light green (Blaženić, 1979).

## RESULTS AND DISCUSSION

The ability of the plant species to adapt to conditions of the habitat is reflected in its anatomical structure. Namely, the structure of leaf is the most indicative for ecological research.

Our investigations shows that the upper and lower leaf surfaces are covered with short straight hairs which are quite rare. Cell walls of the both the upper and lower epidermis, observed at the leaves surfaces are very recurved, which points to the mesomorphic leaf structure. Stomates are located only at the lower side of the leaf, and their number is approximately 160 per  $1\text{ mm}^2$ .

The upper and lower epidermis are single layered, with thin cuticle, while the cells are equal in size (Tab. 1, Fig. 1). Mesophyl consists of the palisade and spongy tissue which are about the same thickness. Palisade tissue is of one layer, consisting of the extended cells, almost without intercellulars, while the spongy tissue is rather loose with wide intercellulars.

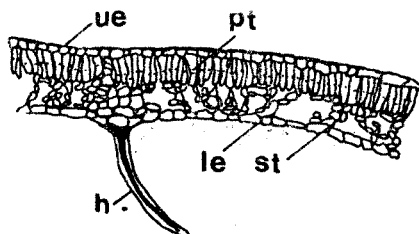


Fig. 1. - Cross section of a leaf of a species *Zelkova crenata* Spach.: ue - upper epidermis, le - lower epidermis, pt - palisad tissue, st - spongy tissue, h - hair.

Tab. 1. – Anatomical features of leaves on cross section in  $\mu\text{m}$ 

Anatomical features	min. - max. ( $\mu\text{m}$ )
Leaf thickness at the level of the central bundle	270 - 291
Leaf thickness	72 - 103
Upper epidermis height	9 - 16
Upper epidermis width	13 - 31
Palisade tissue	25 - 41
Spongy tissue	30 - 41
Lower epidermis height	9 - 16
Lower epidermis width	9 - 28
Hair length	approx. 188

According to the literature (Vukićević, 1987) and on the basis of our own investigations, we have concluded that *Zelkova crenata* Spach. can be considered as a shade species (sciophyte).

The general aspect of the water regime and its dynamics shows a correlation with a present environmental conditions and seasonal changes. The greatest amount of water in leaves (68%) during whole investigation period was found in may. In keeping that, the water saturation deficit was the lowest (16%), while the transpiration of just developed leaves was the most intensive (6,6 mg/g/min). The osmotic pressure of a cell sap was the lowest in this spring month in regard to the summer and autumn months (12 bar).

In July all the water regime parameters were changed comparing to the may values, which is connected with the changes in the environment. In that period the light intensity and solar radiation were the highest, as well as the temperature which values reached the maximum comparing to may and september (22,6°C). The air humidity was considerably lower comparing to may, and because of that the evaporation was the most intensive. The soil layer humidity goes down insignificantly comparing to the previous month. In such a microclimatic environmental conditions, the quantity of water in leaves also goes down comparing to may (62%), water saturation deficit increases (22%), transpiration goes down insignificantly (6 mg/g/min) (because of the high light and temperature intensity), while in keeping with that, the osmotic pressure of a cell sap increases (17 bar).

In september the quantity of water in leaves was unchangeable comparing to July (62%). The water saturation deficit reached the highest value comparing to may and July (27%), and the transpiration intensity went down (3,8 mg/g/min). This fall of transpiration can be connected with the changes that happened in the environment (such as the lowest light intensity, lower air temperature than in July, low evaporation and small quantity of water in the soil). The osmotic pressure of a cell sap were the highest in this month (18 bar), which can be explained by leaves growing old and accumulation of the metabolic products (Fig. 2-3, Tab. 2-3).



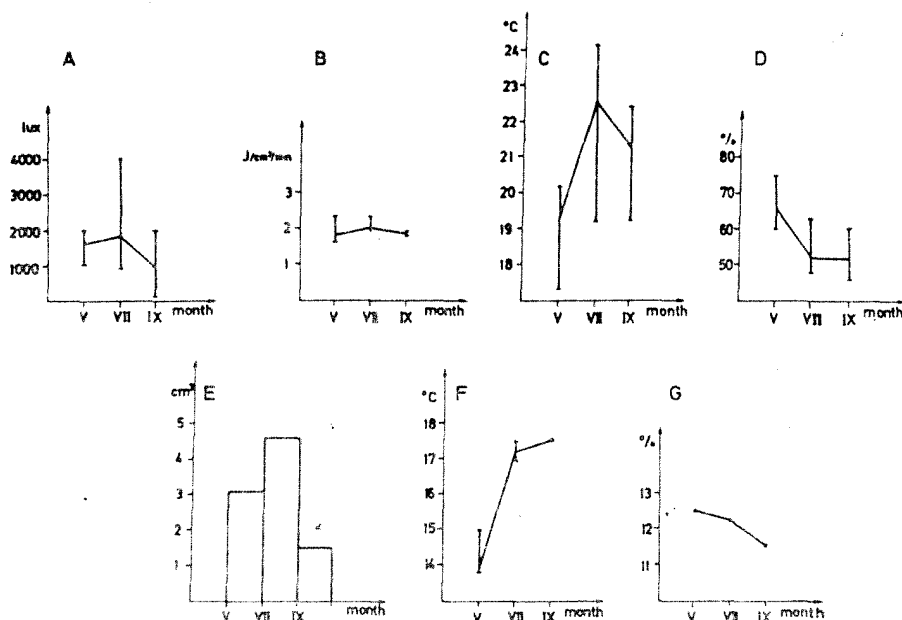


Fig. 2. – Seasonal dynamics of microclimatic conditions of a habitat: (A) light intensity, (B) solar radiation intensity, (C) air temperature, (D) relative air humidity, (E) evaporation, (F) soil temperature, and (G) relative soil humidity.

Tab. 2. – Dynamics of microclimatic factors during the vegetative period

Microclimatic factors	month	minimum	maximum	mean values
A Light intensity (Llux)	V	1000	2000	1680
	VII	900	4000	1836
	IX	100	2000	983
B Solar radiation intensity (J/cm <sup>2</sup> /min)	V	1.68	2.37	1.88
	VII	1.95	2.23	2.04
	IX	1.81	1.95	1.83
C Air temperature (°C)	V	17.30	20.20	19.42
	VII	19.20	24.20	22.60
	IX	19.20	22.40	21.33
D Relative air humidity (%)	V	60	75	66.33
	VII	48	63	52.50
	IX	46	61	52.00
E Evaporation (cm <sup>3</sup> )	V	-	-	3.10
	VII	-	-	4.60
	IX	-	-	1.50
F Soil temperature (°C)	V	13.80	15.00	13.90
	VII	17.00	17.50	17.12
	IX	17.50	17.50	17.50
G Relative soil humidity (%)	V	-	-	12.55
	VII	-	-	12.20
	IX	-	-	11.50

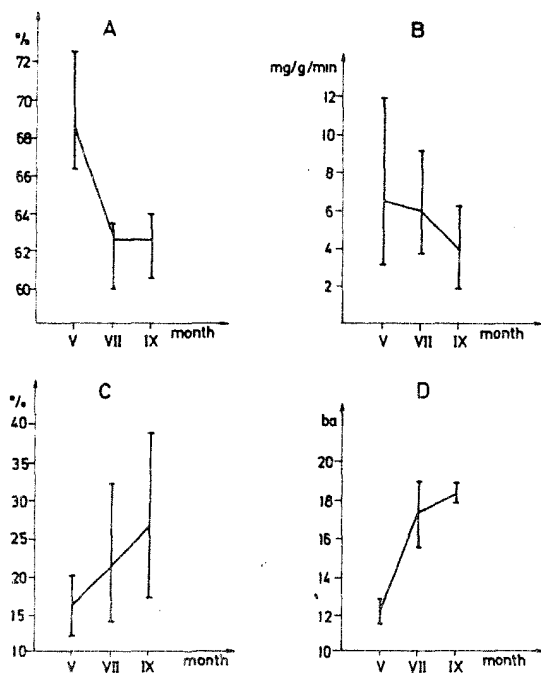


Fig. 3. – Seasonal dynamics of water regime factors: (A) quantity of water in leaves, (B) transpiration, (C) water saturation deficit, and (D) osmotic pressure of a cell sap.

Tab. 3. – Dynamics of water regime during the vegetative period

Water regime factors	month	minimum	maximum	mean values
A. Quantity of water in leaves (%)	V	66.25	72.44	68.23
	VII	59.80	63.36	62.55
	IX	60.43	63.92	62.44
B. Transpiration (mg/g/min)	V	3.16	11.91	6.58
	VII	3.72	9.02	6.04
	IX	1.99	6.32	3.75
C. Water saturation deficit (%)	V	12.31	20.56	16.35
	VII	14.32	32.38	21.80
	IX	17.74	38.95	26.75
D. Osmotic pressure of a cell sap (bar)	V	11.50	12.99	12.17
	VII	15.53	19.09	17.43
	IX	17.92	19.04	18.48

On the basis of the above reported results, it can be seen that during the vegetation period the quantity of water in leaves and the transpiration intensity goes down from spring to autumn months while water saturation deficit and osmotic pressure of a cell sap in this period shows a significant increase.

During whole vegetation period the dynamic changes of water regime of this introduced species are noticed. The investigations of water regime and the anatomic structure showed the physiological stability of the species *Zelkova crenata* Spach. at the Botanical garden in Belgrade.

## CONCLUSIONS

According to morpho-anatomical adaptations and the water regime characteristics, the investigated species is sciophyte with mesomorphic structure, which survives in the areas where are no great variations on the environmental conditions.

The basic characteristics of water regime are relatively weak transpiration during the whole vegetation period as well as the moderately high osmotic pressure of a cell sap.

The species *Zelkova crenata* Spach. has a stable water regime which is in keeping with the seasonal changes in the environment.

## ACKNOWLEDGEMENTS

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### Rezime

TATJANA GRABELJŠEK

#### NEKE EKOFIZIOLOŠKE I MORFO-ANATOMSKE KARAKTERISTIKE VRSTE *ZELKOVA CRENATA* SPACH.

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*Zelkova crenata* Spach. je kao tercijarna vrsta naseljavala južnu Evropu, da bi tokom ledenog doba nestala sa ovih prostora. Danas se sa uspehom gaji u velikom delu Evrope kao ukrasno drvo, dok su termofilne šume Kavkaza njeno prirodno stanište. Ova vrsta ispitivana u Botaničkoj bašti u Beogradu, odlikuje se dinamičkim promenama vodnog režima, što je u skladu sa sezonskim promenama u spoljnoj sredini. Osnovne karakteristike vodnog režima su relativno slaba tanspiracija u toku čitavog vegetacijskog perioda i umereno visok osmotski pritisak. Proučavanje anatomske strukture lista ukazuje na njegovu mezomorfnu građu.

Po morfo-anatomskim adaptacijama i po karakteru vodnog režima, ispitivana vrsta je skiofita sa mezomorfnom građom, koja se održala na mestima gde nema velikog variranja u uslovima spoljne sredine. Istraživanja ukazuju na fiziološku stabilnost vrste *Zelkova crenata* Spach. u uslovima Botaničke bašte u Beogradu.

UDC 582.772.2 : 581.522.5 : 504.3.054 (497.11)  
Original scientific paper

MARINA MAČUKANOVIĆ, GORDANA DRAŽIĆ<sup>1</sup>, BRANKA  
STEVANOVIĆ<sup>2</sup>

**UTICAJ AEROZAGADENJA NA EKOFIZIOLOŠKE I ANATOMSKE  
KARAKTERISTIKE VRSTA *ACER NEGUNDO* I *ACER  
PSEUDOPLATANUS***

Veterinarski fakultet, Beograd, <sup>1</sup>INEP, Zemun,

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Mačukanović, M., Dražić, G., Stevanović, B. (1992-1993): *Effect of air pollution on ecophysiological and anatomical characteristics of the species Acer negundo and Acer pseudoplatanus*. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 49 - 62.

The ecophysiological and coanatomical investigations were carried out on the species *Acer negundo* and *A. pseudoplatanus* in order to establish their vitality and ability to survive in polluted urban areas of Belgrade.

The investigations were concerned with the seasonal dynamics of chlorophyll content as well as the leaves structure injuries of both species analyzed from the downtown localities and from the city neighbourhood respectively.

*A. pseudoplatanus* was established as more resistant species although the pollution damages were observed in both trees studied.

Key words: air pollution, *Acer negundo*, *Acer pseudoplatanus*, anatomical adaptations, chlorophyll content.

Ključne reči: aerozagadenje, *Acer negundo*, *Acer pseudoplatanus*, anatomske adaptacije, sadržaj hlorofila.

## UVOD

Zagađenost vazduha kao posledica intenzivne urbanizacije, industrijalizacije i saobraćaja, jedan je od gorućih problema razvijenih gradova. Na području Beograda, takođe, sve je upadljivije negativno dejstvo zagađivača iz vazduha, vode i zemljišta na biljni i životinjski svet, odnosno na populaciju gradskog stanovništva.

Glavni izvori zagađenja vazduha u razvijenim gradovima, pa i u Beogradu, potiču od ljudske aktivnosti: industrije, komunalnih delatnosti (individualne kotlarnice u kojima sagorevaju čvrsta i tečna goriva za zagrevanje domaćinstava, deponije smeća i drugo) i saobraćaja. U atmosferu se oslobadaju primarni i sekundarni polutanti (sumpor-dioksid, vodonik-halogenidi, oksidi azota, ozon, peroksiacetil-nitrat, ugljovodonici, dim, čađ i prašina), dok se u zemljištu i vodi često povećava koncentracija štetih i otrovnih hemijskih supstanci (fenolnih derivata, jedinjenja teških metala, deterdženata, veštačkih đubriva). Fitotoksične supstance prodiru u biljke iz okolne, vazdušne sredine ili ih one apsorbuju korenovima iz zemljišta i vode.

Pod dejstvom atmosferskih polutanata različite koncentracije, biljke trpe akutna i hronična oštećenja. Ponekad, to su lako vidljivi simptomi, hloroze ili nekroze biljnih tikiva, pre svega listova, sušenje, ubrzano starenje i opadanje lišća, pa čak i umiranje cele biljke. Pri malim koncentracijama fitotoksičnih supstanci, za određeno vreme, nema lako uočljivih promena na biljkama. Dugotrajno dejstvo malih koncentracija gasovitih zagađivača izaziva hronična oštećenja sa kojima se biljka bori i opstaje, zavisno od granica tolerancije. Hronične promene, međutim, dovode do stalnog, postepenog smanjenja produkcije biomase (Kozłowski, 1980; Ormrod, et al., 1981; Crawford, 1989). Zbog toga je značajno ustanoviti rane simptome nepovoljnog uticaja aerozagađivača na biohemijske i fiziološke procese kod biljaka, kao i prve promene anatomske strukture listova biljaka koje rastu na urbanom području.

U strogom centru Beograda, duž gradskih saobraćajnica, drveće je godinama izloženo izduvnim gasovima i drugim polutantima. Koncentracija zagađivača se menja, zavisno od vremenskih uslova, pre svega prisustva vetra (košave). Pod dejstvom aerozagađivača, ali i promenjenih gradskih klimatskih uslova, na lišću drveća se javljaju hlorotične mrlje i požutele ivice, ono ranije stari i prevremeno opada (već krajem avgusta, pa i ranije). Čitava biljka, tokom vremena postaje neotporna, stvara sve manju biomasu, kruna postaje sve prorednija, naročito u vršnom delu koji se sve slabije snabdeva vodom, grane se postepeno suše i otpadaju i drvo najzad umire. Simptomatologija oštećenja od aerozagađivača je veoma različita i nespecifična. Stoga, veliki praktični značaj ima analiza ranih znakova oštećenja, utvrđivanje rezistentnosti i adaptacija biljaka u uslovima urbane sredine.

U tom cilju analizirana je dinamika količine hlorofila, kao parametar fizioloških promena, i anatomska grada listova, kao parametar strukturnih promena, kod drvenastih biljaka *Acer negundo* i *A. pseudoplatanus* pod dejstvom urbanog aerozagađenja. Ispitivanja su obavljena na biljkama iz strogog centra Beograda, koji je, prema merenjima koje svakodnevno obavlja Gradski zavod za zaštitu zdravlja, izložen visokoj koncentraciji različitih atmosferskih polutanata. Radi preciznije interpretacije rezultata obavljena je uporedna analiza istih parametara, na istim vrstama drveća koje raste u parku (bašti) istraživačkog centra INEP, desetak kilometara udaljenog od strogog centra Beograda.

## MATERIJAL I METODIKA

Biljni materijal za morfo-fiziološka ispitivanja uziman je u strogom centru grada, na opštini Stari Grad. Na prvom lokalitetu, u ulici Đure Đakovića, analizirani su listovi *Acer pseudoplatanus*, a na drugom lokalitetu, u ulici Dunavskoj, listovi *Acer negundo*. Ove gradske saobraćajnice, kojima prolazi laki, putnički i teški, transportni saobraćaj, nalaze se u neposrednoj blizini Ulice 29. novembra u kojoj postoji merna stanica Gradskog zavoda za zaštitu zdravlja. Pomenuti lokaliteti označeni su u radu kao zagađena sredina.

Provera analiziranih karakteristika listova javora obavljena je upoređivanjem sa istim, morfo-fiziološkim parametrima, na listovima javora koji su rasli u daljoj okolini grada, na prigradskom lokalitetu, u dvorištu INEP-a. Ovaj lokalitet označen je kao nezagađena sredina.

### Analiza hlorofila

Sadržaj hlorofila i ukupnih pigmenata u biljnim ekstraktima u DMF izmeren je simultanom spektrofotometrijskom metodom na aparatu AMINCO DW 2. Pigmenti su ekstrahovani u DMF u toku 72 sata na 4°C, u mraku. Za određivanje količine pigmenata korišćeni su koeficijenti po Moranu (Moran, 1982). Rezultati predstavljaju srednju vrednost najmanje četiri nezavisna merenja. Dobijeni podaci obrađeni su kompjuterski, standardnim statističkim metodama.

### Anatomska analiza

Listovi biljaka za anatomsku analizu sakupljeni na terenu, radi trajnog čuvanja, stavljeni su u rastvor alkohola i formalina, a u laboratoriji preneti u fiksativ Navašina (Prozina, 1960). Trajni preparati pravljeni su standardnim postupkom koji obuhvata obradu fiksnog materijala parafinskom metodom, sečenje preseka na ručnom mikrotomu (15 µm debljine) i dvojno bojenje preparata svetlo-zelenim i safraninom (Chamberlain, 1921, Prozina, 1960).

### Kontrola zagađenosti vazduha

Kontrola zagađenosti vazduha u Beogradu obavljena je svakodnevno od strane Gradskog zavoda za zaštitu zdravlja. Preuzeti rezultati ovih merenja prikazani su u tabeli 1.

Tab. 1.

Ulica 29. novembra				
Izvor aerozagađivanja				
stacionarni izvori			motorna vozila	
mesec	SO <sub>2</sub>	čadj	CO	olovo
V	86	56	4,7	4,3
VI	79	68	4,6	4,3
VII	80	71	3,7	3,5
VIII	92	75	3,4	3,2

Na tabeli su date srednje mesečne vrednosti izražene u µg/cm<sup>3</sup>, a, prema standardima, maksimalna dozvoljena doza (MDK) za SO<sub>2</sub> je 150 µm/m<sup>3</sup>, a za čadj 50 µm/m<sup>3</sup>.

### Zagađena sredina

Stablo *Acer negundo* sa kojeg su uzeti listovi za analizu, visoko je oko 15 metara, sa razgranatom, proređenom krošnjom, i nalazi se neposredno uz kolovoz u Dunavskoj ulici.

Stablo *Acer pseudoplatanus* čiji su listovi analizirani nalazi se na trotoaru, okruženo asfaltom, u ulici Đure Đakovića i visoko je oko 10 metara, sa relativno dobro razvijenom i u vršnom delu proređenom krošnjom.

### Nezagađena sredina

Stabla *A. negundo* i *A. pseudoplatanus* sa kojih su uzimani listovi radi kontrolnih merenja, visoka su oko 15 m, približne starosti oko 25 godina, odlikuju se veoma razgranatom i gustom krošnjom, kao i neoštećenim, zelenim listovima.

## REZULTATI

Vrsta *Acer negundo* L. pajasen ili jasenolosni javor poreklom je iz Severne Amerike, gde je rasprostranjen od Kanade do Meksika i Floride. Raste u dolinama reka i jezera, uglavnom na dovoljno vlažnim zemljištima. Jasenolisni javor je severnoamerički florni elemenat i značajna adventivna drvenasta biljka, uspešno introdukovana u Evropi i drugim delovima sveta. Izuzeto često se gaji po parkovima, baštama i drvoredima, zbog čega, na širem prostoru, na takvim mestima, raste i samoniklo. Ovaj javor je heliofilno drvo, ali je osetljiv na nedostatak vode u podlozi i jake vetrove. Relativno je kratkotrajno, tako da u drvoredima dolazi do propadanja delova ili čitave krune već posle 25-30 godina. Cveta rano, već u martu, a cvetovi su dvodomi. Rasejava se i razmnožava vetrom.

Veoma je pogodno drvo za urbane uslove jer brzo raste, naročito u mladosti i veoma je dekorativnog izgleda. Na gradskim „zelenim” površinama u Beogradu spada među najčešće gajeno drveće u drvoredima i parkovima (Jovanović, 1950).

Vrsta *Acer pseudoplatanus* L., javor, rasprostranjena je u celoj Evropi, izuzev na Pirinejskom poluostrvu. Najčešće se sreće u brdskom šumskom pojasu, dok u južnoj Evropi raste u regionu bukve ili mešovitim šuma bukve i jele. Pripada srednjeevropskom flornom elementu. Najbolje uspeva na umereno vlažnim i umereno hladnim staništima, sa dobro razvijenim zemljištem. Ovo drvo u prirodi može da dostigne veliku starost (do 300 godina). Počinje da cveta tek kad dostigne oko 20 godina starosti. U isto vreme cveta i lista, i to od aprila do juna. Raste brzo, formira stablo sa velikom i dekorativnom krunom zbog čega se često gaji u parkovima, baštama i gradskim drvoredima. U hortikulturi su cenjene ukrasne forme ovog javora sa lišćem od svetlo do tamno zelene i zeleno-crvene boje. Dostiže visinu od oko 30 m i širinu stabla od oko 2,5 m.

Rezultati merenja količine ukupnog hlorofila kod vrsta *A. negundo* i *A. pseudoplatanus* iz nezagađene (prigradske) i zagađene (gradske) sredine prikazani su na tabeli 2.



Tab. 2. – Količina hlorofila ( $\mu\text{g}/\text{cm}^2$ ) u listovima *Acer negundo* i *A. pseudoplatanus* tokom vegetacijskog perioda, u zagadenoj i nezagadenoj sredini.

Chlorophyll content ( $\mu\text{g}/\text{cm}^2$ ) in leaves of *A. negundo* and *A. pseudoplatanus* during vegetation period in polluted and unpolluted area.

datum (date)	<i>Acer pseudoplatanus</i>		<i>Acer negundo</i>	
	Chl <sub>a+b</sub> " ( $\mu\text{g}/\text{cm}^2$ )	Chl <sub>a+b</sub> ( $\mu\text{g}/\text{cm}^2$ )	Chl <sub>a+b</sub> ( $\mu\text{g}/\text{cm}^2$ )	Chl <sub>a+b</sub> ( $\mu\text{g}/\text{cm}^2$ )
	nezagadeno (unpolluted)	zagadeno (polluted)	nezagadeno (unpolluted)	zagadeno (polluted)
22/5/90	61.36	70.50	42.68	42.06
25/6/90	69.87	56.80	45.44	58.30
23/7/90	82.86	39.03	31.72	57.75
30/8/90	70.05	39.28	38.88	37.15
3/10/90	67.30	15.76	36.73	17.43

*Acer negundo*

Sadržaj hlorofila

Kod mladih listova jasenolisnog javora, u maju, izmerena je skoro podjednaka količina hlorofila kako u nezagadenoj ( $42,7 \mu\text{g}/\text{cm}^2$ ) tako i u zagadenoj sredini ( $42,1 \mu\text{g}/\text{cm}^2$ ). Tokom sezone, međutim, koncentracija hlorofila se neznatno povećavala u listovima biljke iz nezagadene sredine, tako da je u junu postigla vrednost od  $45,4 \mu\text{g}/\text{cm}^2$ , a zatim se, do kasno jesenjeg perioda zadržala na nešto manjim, ali konstantnim vrednostima (prosečno za period juli-oktobar  $35,7 \mu\text{g}/\text{cm}^2$ ). U istom periodu, koncentracija hlorofila u listovima biljke iz zagadene sredine dostigla je visoku vrednost u junu od  $58,3 \mu\text{g}/\text{cm}^2$ , a zatim se naglo smanjivala krajem leta i početkom jeseni. U oktobru je konstatovana vrednost od svega  $17,4 \mu\text{g}/\text{cm}^2$  što je upola manja koncentracija hlorofila od one konstatovane kod listova jasenolisnog javora iz nezagadene sredine (Sl. 1).

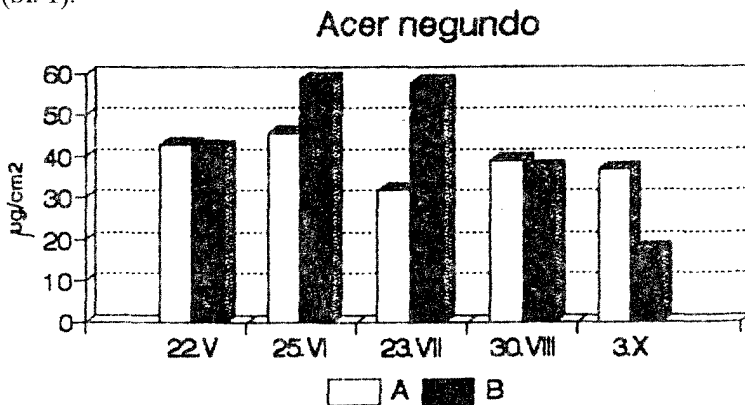


Fig. 1. – Sezonska dinamika količine hlorofila ( $\mu\text{g}/\text{cm}^2$ ) u listovima *Acer negundo* u A – nezagadenoj i B – zagadenoj sredini.

Seasonal variations of chlorophyll content in leaves of *Acer negundo* from A – unpolluted, and B – polluted area.

## Ekoanatomske odlike

Listovi *A. negundo* su neparno perasto složeni, najčešće od 5 listića dugih oko 5-10 cm, na vrhu zašiljenih, a pri osnovi okruglastih, i po obodu fino nazubljenih.

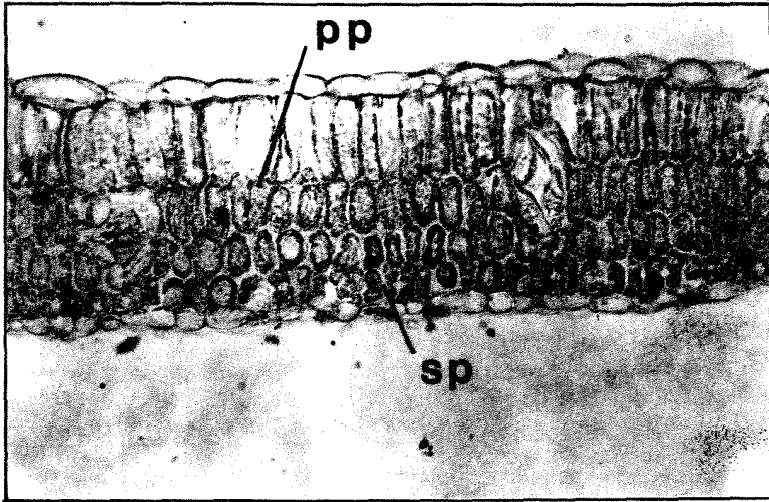


Fig. 2. – Poprečni presek kroz list *Acer negundo* iz nezagađene sredine: pp – palisadni parenhim, sp – sunderasti parenhim.

Cross section of *Acer negundo* leaf from unpolluted area: pp – palisade parenchyma, sp – spongy parenchyma.

List jasenolisnog javora je hipostomatičan, slabo izražene dorziventralne strukture mezofila. Čelije epidermisa su relativno krupne, izrazito većih dimenzija na licu u odnosu na naličje lista, sa slabo razvijenom kutikulom. Mezofil čine četiri jasno izdvojena sloja ćelija čija se veličina pravilno, a postepeno smanjuje od lica ka naličju lista. U sva četiri niza ćelije su raspoređene na način uobičajen za ćelije palisadnog parenhima, tako da se teško zapaža diferencijacija na palisadno i sunderasto tkivo. Ipak, dva niza (samo retko tri sloja) ćelija, ispod epidermisa lica, čine duguljastije i pravilne, gusto naslagane palisadne parenhimske ćelije, dok jedan do dva sloja ćelija ispod njih, odnosno iznad epidermisa naličja, čine nešto okruglastije i manje uredno raspoređene ćelije sunderastog tkiva (Sl. 2). Pored toga, u mezofilu se zapažaju krupne ćelije ispunjene izuzetno velikim pojedinačnim kristalima. Stome su sitne, mnogobrojne na poprečnom preseku, u nivou ćelije epidermisa naličja, čak, sasvim neupadljivo ispučene. Sitni kristali prisutni su u parenhimskim ćelijama oko povodnih snopića, naročito u regionu centralnog nerva lista.

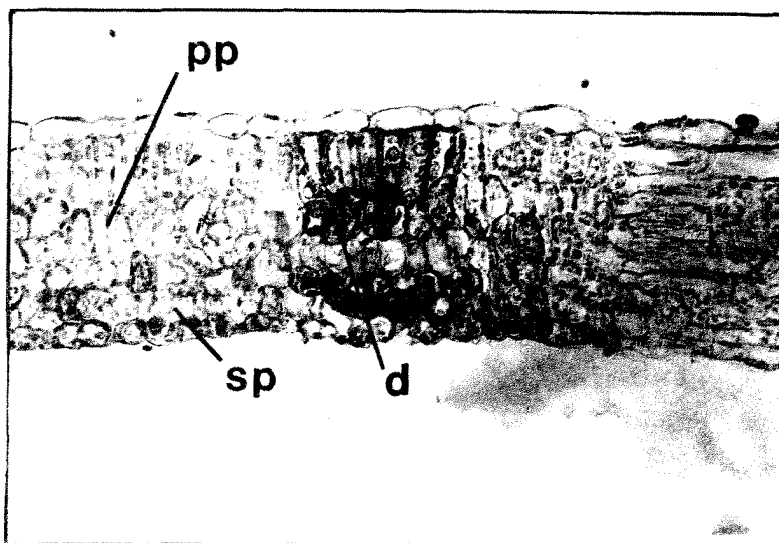


Fig. 3. – Poprečni presek kroz list *Acer negundo* iz zagađene sredine: pp – palisadni parenhim, sp – sunderasti parenhim, d – oštećene i deformisane ćelije.

Cross section of *Acer negundo* leaf from polluted area: pp – palisade parenchyma, sp – spongy parenchyma, d – injured and deformed cells.

List biljke iz zagađene sredine je bledo-zelene do bledo-žute boje. Hlorotične mrlje su upadljive, česte po obodu liske ili razbacane po površini liske, između nerava.

Promene u anatomskoj građi lista iz zagađene sredine zapažaju se u oštećenjima na epidermisu, na stomama, ali i u ćelijama mezofila. Na više mesta oštećen je spoljašnji zid epidermalnih ćelija, ili su potpuno uništene jedna ili više epidermalnih ćelija. Oštećenja su upadljiva na naličju lista (retko na licu lista), polaze od epidermisa i zahvataju manju ili veću zonu tkiva mezofila. Uglavnom, oštećenja stižu do drugog niza palisadnih ćelija, tako da prvi sloj ćelija palisadnog tkiva, odmah ispod epidermisa lica, ostaje neoštećen (Sl. 3). List jasenolisnog javora iz zagađene sredine je manje debljine od onog iz nezagađene sredine. Kod ovog lista ćelije mezofila su kraće i nešto šire. U ćelijama palisadnog tkiva uočavaju se okruglasti, nabrekli hloroplasti, neravnomerno raspoređeni, za razliku od vretenastih hloroplasta u ćelijama mezofila listova iz nezagađene sredine, koji su kao brojanice nanizani duž zidova palisadnih ćelija. Pored toga, u listu su prisutni brojni kristali, rede u ćelijama mezofila, a izuzetno često oko, a naročito ispod, provodnih snopića.

#### *Acer pseudoplatanus*

##### Sadržaj hlorofila

U listovima ovog javora konstatovana je značajna količina hlorofila, pre svega kod biljaka iz nezagađene sredine ( $61,4-82,9 \mu\text{g}/\text{cm}^2$ ), ali i kod biljaka iz zagađene sredine ( $79,5-15,8 \mu\text{g}/\text{cm}^2$ ). Sezonska dinamika količine hlorofila, međutim, ima različiti tok kod listova iz nezagađene (prigradske) i zagađene (urbane) sredine.

Kod biljaka iz nezagadene sredine, u mladim listovima, količina hlorofila iznosila je  $61,4 \mu\text{g}/\text{cm}^2$ . Tokom vegetacijske sezone koncentracija hlorofila se postepeno povećavala i u julu dostigla maksimalne vrednosti od  $82,9 \mu\text{g}/\text{cm}^2$ . Prema jeseni, sa starenjem listova, koncentracija hlorofila postepeno opada do vrednosti od  $67,3 \mu\text{g}/\text{cm}^2$  izmerene u oktobru (Sl. 4)

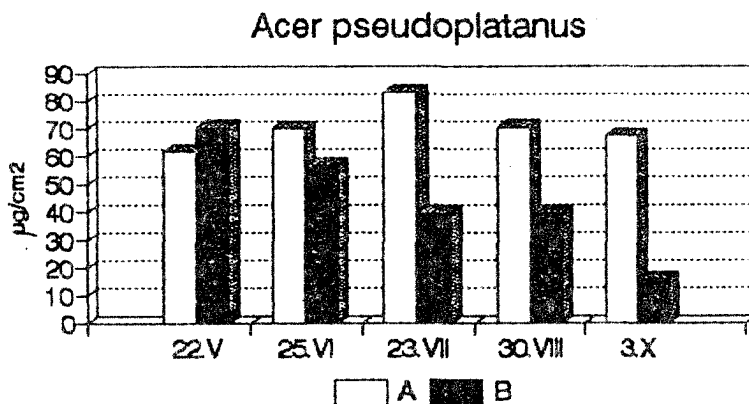


Fig. 4. – Sezonska dinamika količine hlorofila ( $\mu\text{g}/\text{cm}^2$ ) u listovima *Acer pseudoplatanus* u A – nezagadenoj i B – zagadenoj sredini.

Seasonal variations of chlorophyll content in leaves of *Acer pseudoplatanus* from A – unpolluted, and B – polluted area.

Nasuprot tome, mladi listovi *Acer pseudoplatanus* iz zagadene sredine odlikovali su se značajnom količinom hlorofila od  $70,50 \mu\text{g}/\text{cm}^2$  u maju. Kako su listovi sazrevali i starili, tako se sadržaj hlorofila konstantno i naglo smanjivao. Skorog upola manje vrednosti zabeležene su već u julu ( $39,0 \mu\text{g}/\text{cm}^2$ ) i avgustu ( $39,3 \mu\text{g}/\text{cm}^2$ ). U oktobru, sasvim požuteli listovi sadržali su svega  $15,8 \mu\text{g}/\text{cm}^2$  hlorofila (Sl. 4).

#### Ekoanatomske odlike

Vrsta *Acer pseudoplatanus* odlikuje se listom široke osnove i liskom usečenom na pet režnjeva. Listovi su tamnozeleni, na licu glatki, a na naličju slabo dlakavi, ili sa čupercima dlaka uz nerve.

List ovog javora je hipostomatičan i dorziventralne građe. Čelije epidermisa su relativno krupne, na licu lista veće, uglavnom pravougaone, dok su na naličju lista sitnije, okruglaste i papilozne (Sl. 5). Palisadno tkivo čini jedan red izrazito izduženih, stubastih i pravilno raspoređenih ćelija. U dvo- do troslojnom sunderastom tkivu ćelije su nepravilno četvorouglaste do okruglaste, razdvojene intercelularima znatnih dimenzija. Odnos palisadnog i sunderastog tkiva je 1:1. Stome su sitne, brojne, u nivou unutrašnjeg tangencijalnog zida papilozne epidermalne ćelije naličja lista. Dlake su dugačke, višeceljske i nerazgranate.

Znatna oštećenja listova na stablu iz zagadene sredine uočavaju se već krajem juna. Mrke fleke javljaju se na ivicama liske i šire se ka sredini (centralnom delu) liske, kako po površini, tako i u dubini lista zahvatajući tkiva mezofila. Kod ovog javora vidljiva su oštećenja i na licu lista. Ukupna širina lista je manja, naročito su kraće ćelije palisadnog tkiva, tako da je sunderasto tkivo neznatno šire od palisadnog parenhima (Sl. 6). Palisadno tkivo čini jedan sloj ćelija u kojima se zapažaju uvećani, okruglasti

hloroplasti, raštrkano raspoređeni u okviru jedne ćelije. U ćelijama sunderastog tkiva česti su krupni kristali. Oštećenja su vidljiva na pojedinačnim ćelijama, ili obuhvataju grupu ćelija, pre svega na donjoj strani lista, ili su, čak, kompletno nekrotični manji ili veći delovi svih tkiva mezofila. Na mestima oštećenja ćelije sunderastog ili palisadnog parenhima su promenjenog oblika, uvećane i ispunjene specifičnim, u ovom slučaju nedefinisanih sadržajem ili velikim kristalima.

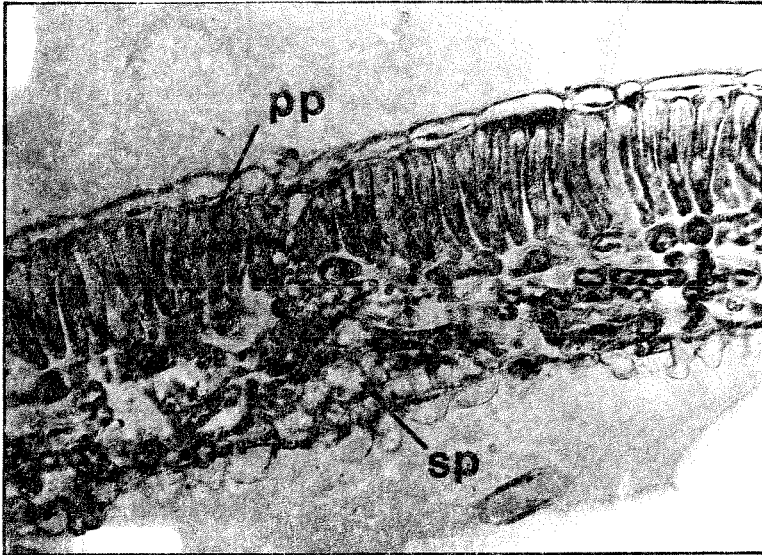


Fig. 5. – Poprečni presek kroz list *Acer pseudoplatanus* iz nezagadene sredine: pp – palisadni parenhim, sp – sunderasti parenhim.

Cross section of *Acer pseudoplatanus* leaf from unpolluted area: pp – palisade parenchyma, sp – spongy parenchyma.

Nekrotična oštećenja različitog porekla zahvataju velike delove starijih listova, počevši od sredine leta.

## DISKUSIJA

Sadržaj ukupnog hlorofila ispitivanih vrsta javora varira shodno ekofiziološkim odlikama i starosti listova ovih drvenastih biljaka, kao i u zavisnosti od sezonskih promena spoljašnje sredine, tokom vegetacijskog perioda. Pored toga, u uslovima zagadene sredine u Beogradu, gde se izmerene vrednosti  $SO_2$  nalaze blizu maksimalno dozvoljenih koncentracija, a količina čadi prevazilazi MDK, ove promene su naglašene, tako da je evidentno smanjenje količine ukupnih pigmenata kod ispitivanih vrsta drveća iz uskog gradskog područja.

U strogom gradskom centru Beograda, vrste *Acer negundo* i *A. pseudoplatanus* izložene su višestrukim stresnim uslovima. Sinergističko dejstvo polutanata, deficita vlažnosti i visokih letnjih temperatura kontinentalne klime ovog područja utiče na brže

starenje listova ovih biljaka, pojavu nekrotičnih mrlja već u rano leto, manji prinos biomase listova i proređivanje krune drveća. Najčešće mala količina zemljišta u uličnim drvodredima oko pojedinačnog drveća i nedovoljno navodnjavanje (zalivanje) pojačava uslove nepovoljnog vodnog režima neizbežno termofilnijih urbanih staništa.

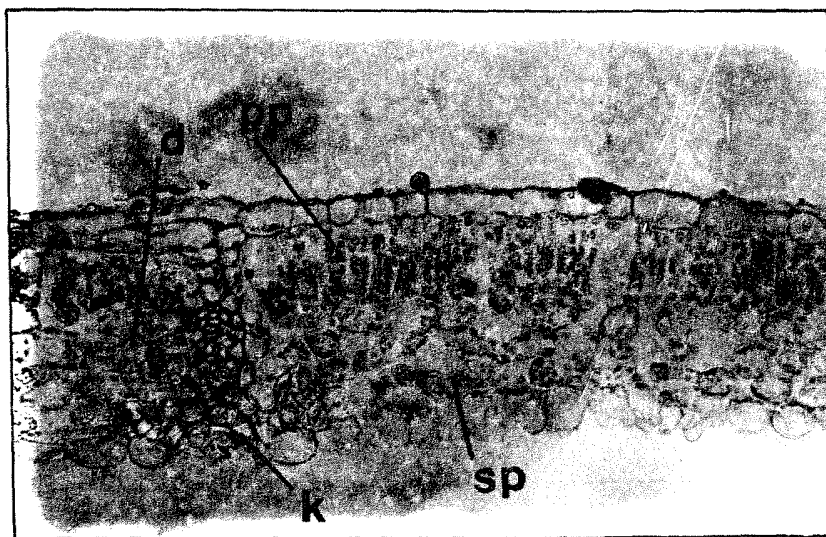


Fig. 6. – Poprečni presek kroz list *Acer pseudoplatanus* iz zagađene sredine: pp – palisadni parenhim, sp – sunderasti parenhim, d – oštećene i deformisane ćelije, k – kristali.

Cross section of *Acer pseudoplatanus* leaf from polluted area: pp – palisade parenchyma, sp – spongy parenchyma, d – injured and deformed cells, k – crystals.

U urbanoj sredini biljke rastu izložene kombinovanom dejstvu više polutanata (Kozłowski, 1980), a pre svega sumpordioksida i nekih oksidacionih supstanci (Black and Black, 1979; Black and Unsworth, 1980; Krizek et al., 1985), pri čemu, najčešće, dolazi do bržeg zatvaranja stoma (Elkiey and Ormrod, 1979; Constantinidou and Kozłowski, 1979), razgradnje hloroplasta, ograničavanja fotosinteze i prinosa biljaka (Dugger and Ting, 1970; Hallgren and Huss, 1975; Phillips et al., 1977). Hlorotične i nekrotične mrlje na listovima, i između nerava, kao i na obodu lista, uočene kod obe vrste javora odgovaraju promenama koje nastaju usled štetnog delovanja kombinacije nekoliko aerozagadivača. Promene na listovima *A. negundo* i *A. pseudoplatanus* iz urbanog područja Beograda konstatovane su, pre svega, u epidermisu naličja lista i donjem delu mezofila (prema naličju lista). Najčešće pod dejstvom gasovitih aerozagadivača oštećenja počinju oko stominih i okolnih epidermalnih ćelija, zalaze u tkiva mezofila i utiču na promenu i smrt ćelija sunderastog tkiva, ispod stominih otvora.

Pored toga, listovi ispitivanih biljaka, naročito vrste *Acer negundo*, bili su spolja pokriveni prljavštinom, prašinom i česticama čadi koje su pristanale za lepljivu površinu liske. Lepljiva prevlaka ili „medna rosa” na listu može se povezati i sa povećanim prisustvom fitofagnih filobionata, koji, sa svoje strane, takode, ubrzavaju negativne promene kod biljaka izloženih fitotoksičnim substancama (Keller and Muller, 1958; Fluckiger et al., 1979).

Opšte nepovoljne urbane uslove za razvoj drvenastih biljaka zbog zagađenja, nespecifičnih konkurentskih odnosa, kidanja mladica i grana, „higijenskih seča” krune drveća i drugog, pogoršavaju i sledeće okolnosti:

- nepovoljan vodni režim uskog prstena zemljišta oko stabala, naročito u drvoredima na asfaltiranim trotoarima ulica. Ova mala površina zemljišta je, pored toga, najčešće gažena i utabana, bez dobre aeracije, zagađena naftom, benzinom, olovom, insekticidima i raznim drugim otpadnim materijama.

- promenjeni gradski klimatski uslovi koji se odlikuju, uopšte uzev, višim temperaturama vazduha i zemljišta i, često, manjom ukupnom količinom padavina.

- često prisustvo fitofagnih filobionata (koji sišu ili bodu list i stvaraju lepljivu prevlaku – „mednu rosu”) ili, čak, patogenih mikroorganizama zbog poremećenih odnosa ekološke ravnoteže.

Nepovoljni uslovi urbane sredine deluju na sličan način kao i uslovi stresa (suša, niske temperature, preterano zračenje, nedostatak mineralnih elemenata) u nenarušenoj, prirodnoj sredini na morfo-fiziološke karakteristike biljaka. Osnovne promene, u ovakvim okolnostima, odgovaraju adaptacijama kserofilnog (kseromorfnog) tipa. Konstatovano je da su listovi ispitivanih javora iz centra Beograda manjih dimenzija, da im je redukovana mezofil, pre svega je smanjen broj palisadnih slojeva, a palisadne ćelije su kraće. Promene manje ili više kseromorfnog tipa su prvi stepen odbrane ovih biljaka u nepovoljnim uslovima urbane sredine. Na taj način se pojačava njihova rezistentnost i odlaže konačni negativni efekat već prisutnih oštećenja.

S obzirom na svoje poreklo, ekološko-evolucijsku prilagodенost i rasprostranjenje, *Acer negundo* i *A. pseudoplatanus* pokazuju različit stepen rezistentnosti, odnosno osetljivost na delovanje aerozagadivača i druge antropogene uticaje u gradskim sredinama

Ekofiziološka i morfo-anatomska istraživanja su pokazala da se *Acer negundo* odlikuje većom osetljivošću na promene u sastavu vazduha u urbanoj sredini grada Beograda u odnosu na *A. pseudoplatanus*.

Vrsta *Acer negundo* u gradskim uslovima odlikuje se manjom ukupnom količinom hlorofila u odnosu na *A. pseudoplatanus*, kao i velikim variranjima u sadržaju hlorofila tokom vegetacijske sezone. Ovakve promene, verovatno, utiču na fotosintezu i mogu se posredno povezati sa smanjenom opštom produkcijom i prirastom biljke. Hloroze, registrovane već na mladim listovima ovog javora, ukazuju na direktne i indirektne negativne efekte urbane spoljašnje sredine. Pojava hloroza i kod listova iz dalje okoline grada, mada manjeg obima i učestalosti, uočena tek sredinom leta, posledica je, najverovatnije, izrazito sušnih uslova klime u godini kada su vršena ispitivanja. Inače, 1990. godina karakterisala se velikom sušom, tako da je prosečna, mesečna količina padavina tokom vegetacijske sezone (u periodu između aprila i oktobra) iznosila 44 mm, srednja mesečna temperatura, u istom periodu, bila je 18°C (prema podacima Republičkog hidrometeorološkog zavoda). Uopšte uzev, jasenolisni javor, koji raste, pre svega, na dovoljno vlažnim mestima, sa manjom ekološkom

plastičnošću podnosi zagađenja vazduha u šumo-stepskim uslovima klime područja Beograda. Povećana količina ukupnog hlorofila tokom prva tri meseca, u proleće i rano leto, u zagađenoj sredini, uprkos vidljivim hlorotičnim promenama na listu, govore o maksimalnim mogućnostima biljke da fiziološki produži rezistentni odgovor na uslove aerozagađenja.

Medutim, kod vrste *Acer pseudoplatanus*, maksimalna vrednost koncentracije hlorofila ( $70,5 \mu\text{g}/\text{cm}^2$ ), izmerena u prolećnim mesecima, naglo opada tokom leta. Istovremeno se uočavaju znatna oštećenja listova kod biljaka sa centralnog gradskog područja. Sve ove promene predstavljaju simptome ranog sušenja i istarenja listova. Oni počinju da opadaju već tokom kasnog leta (krajem avgusta). Prema tome, ovo drveće se odlikuje skraćenim vegetacijskim periodom tokom kojeg obrazuje manju količinu nove biomase. Ovakve ekološke okolnosti, tokom vremena, utiču na smanjenje adaptivnih mogućnosti i kompetitivnosti ove vrste javora u okviru urbanih vegetacijskih celina. Pored toga, drveće sa oštećenim lišćem i smanjenom ukupnom biomasom krune slabo doprinosi popravljaju opštih ekoloških uslova grada. Zbog toga se, u gradskim sredinama mora, s jedne strane, nastojavati na smanjenju aerozagađenja da bi se održale već postojeće vrste biljaka (drveća), a s druge strane, treba negovati i održavati rezistentne vrste drveća. Time se obezbeđuje dugotrajnija i efikasnija uloga vegetacije, kao i njena veća estetska vrednost u gradovima.

Ipak, *A. pseudoplatanus* se odlikuje značajnom vitalnošću. Uprkos negativnim promenama tokom vegetacijske sezone pod uticajem, pre svega aerozagađivača, ovo drvo se, svakog proleća dobro i konstantno obnavlja, razvija snažnu krunu sa relativno neoštećenim listovima sve do sredine leta. Prilagodan na umereno vlažne i umereno tople uslove na svojim prirodnim staništima na brdsko-planinskom području, ovaj javor se trajnije suprotstavlja negativnom dejstvu urbane sredine. Već u bližoj okolini grada *A. pseudoplatanus* se nesmetano normalno razvija i odlikuje dužom vegetacijskom sezonom, s obzirom da je u svojim ekološkim zahtevima dovoljno plastičan i prilagodljiv. Ekološki monitoring ove vrste javora, na različitim mestima u gradu, pomogao bi otkrivanju ranih simptoma oštećenja, kao i u ustanovljavanju adaptivnih osobina rezistentnih formi *A. pseudoplatanus* na uslove povećane koncentracije gasovityh zagađivača.

## ZAKLJUČAK

Introdukovani jasenolisni javor *Acer negundo* i autohtoni javor *A. pseudoplatanus* spadaju u izuzetno često gajene biljke u najužem delu Beograda. Oštećenja ovog drveća, njihovo brže starenje i propadanje, u urbanoj sredini Beograda izazvalo je sinergističkim delovanjem aerozagađivača (sumpordioksid, supstance sa oksidacionim dejstvom, čad, prašina) i ksero-termofilnih uslova gradske klime šumostepskog područja. Akutne i hronične promene kod obe vrste javora konstatovane su na osnovu analize dnevne i sezonske dinamike količine hlorofila i promene anatomske strukture listova.

Količina ukupnog hlorofila listova *Acer negundo* u prvoj polovini vegetacijske sezone fluktuirala, a zatim naglo opada. Na listovima su vidljive brojne hlorotične i nekrotične mrlje i fleke, između nerava i duž oboda liske. U anatomske strukturi lista uočavaju se oštećenja epidermisa (veći broj izumrlih ćelija), gubitak pravilne slojevitosti ćelija mezofila, kao i brojne oštećene ili promenjene, oblikom i sadržajem, ćelije sunderastog, rede palisadnog tkiva.



Kod *Acer pseudoplatanus* količina ukupnog hlorofila konstantno, a u drugoj polovini leta naglo opada, tako da se visok sadržaj hlorofila u maju svodi na upola manju količinu krajem jula. Već početkom leta zapažaju se oštećenja ćelija epidermisa i deformacije ćelija mezofila.

Iako su kod obe vrste javora konstatovana strukturna oštećenja i smanjenje količine hlorofila u listovima, ipak, u uslovima urbane sredine Beograda, *A. pseudoplatanus* pokazuje veću rezistentnost od *A. negundo*. Naime, uprkos konstantnim i ranim oštećenjima već na mladim listovima, *A. pseudoplatanus* zadržava nepromenjenju sposobnost obnavljanja i svakog proleća razvija krunu velikih dimenzija, te, makar i u kraćem periodu, veoma efikasno obavlja proces fotosinteze. S druge strane, aerozagadenje u sprezi sa drugim nepovoljnim faktorima urbane sredine izaziva hronične promene kod *A. negundo*; tokom godine, na ovom drvetu se uočavaju ne samo oštećenja listova, već i vidljivo proredivanje i smanjenje ukupnog obima krune.

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### Summary

MARINA MAČUKANOVIĆ, GORDANA DRAŽIĆ<sup>1</sup>, BRANKA STEVANOVIĆ<sup>2</sup>

#### EFFECTS OF AIR POLLUTION ON ECOPHYSIOLOGICAL AND ANATOMICAL CHARACTERISTICS OF *ACER NEGUNDO* AND A. *PSEUDOPLATANUS*

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The introduced boxelder (*Acer negundo*) and autochthonous sycamore (*A. pseudoplatanus*) are commonly grown trees along the streets, in parks and squares of Belgrade. The injuries of these trees, their faster senescence and perishing, in the urban area, are caused by synergistic effects of air pollutants (sulphur dioxide, nitrogen oxides, ozone and other oxidants, soot and dust) and general climatic conditions (of the forest-steppe region) in and around Belgrade. The acute and chronic damages in both species were established on the basis of diurnal and seasonal variations of chlorophyll content and changes in leaf anatomical structure.

Total chlorophyll content in *Acer negundo* leaves is varying during first half of vegetation season, and then abruptly decreased.

Irregular numerous marginal and interveinal necrotic and chlorotic blotches are evident on boxelder expanded leaves. As for anatomical leaf structure, epidermis shows injury symptoms (dead cells), layers of mesophyll are disordered, and many cells of spongy, more rarely of palisade parenchyma are either injured or their shape and content changed.

In *A. pseudoplatanus* the total chlorophyll content constantly decreases, whereas in the second half of the summer it sharply declines so that its high content in May is reduced to half that toward the end of July.

Already at the beginning of summer injury symptoms on the epidermal cells and deformations of mesophyll tissue cells are evident on sycamore leaves.

It should be noted, however, that *A. pseudoplatanus* is more resistant to air pollution in Belgrade urban environment than *A. negundo*. Namely, in spite of injury symptoms appearing already on the young leaves, sycamore retains the unaltered ability of renewal so that each spring it restores a large crown, which enables very efficient photosynthesis though over a shorter period of time. On the other hand, air pollution, in combination with other unfavourable urban environmental stresses causes chronic changes in boxelder reflected not only in leaf chlorosis or necrosis but also in obvious thinning and decrement of tree crown.

UDC 581.92 : 582.669 (497.1)  
Original scientific paper

MARJAN NIKETIĆ

***CERASTIUM NEOSCARDICUM*, A NEW SPECIES OF *SER. ALPINA*  
FROM MT SHAR-PLANINA, SERBIA**

Natural History Museum, Belgrade

Niketić, M. (1992-1993): *Cerastium neoscardicum*, a new species of *Ser. Alpina* from Mt Shar-planina, Serbia. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 63 - 70.

*Cerastium neoscardicum* Niketić is described and illustrated from serpentine high-mountain region of Shar-planina mountain (S.W. Serbia: Kosovo). This new species is closely related to *C. alpinum* and *C. decalvans* Schlosser & Vuk.

Key words: genus *Cerastium*, Serbian flora, Mt Shar-planina, new species, *Cerastium neoscardicum*.

Ključne reči: rod *Cerastium*, flora Srbije, Šar-planina, nova vrsta, *Cerastium neoscardicum*.

INTRODUCTION

While studying the flora of the Mt Shar-planina mountain (S.W. Serbia: Kosovo), we encountered an isolated population of the genus *Cerastium* L. (*Ser. Alpina* Borza), occurring on the serpentine grounds. The comparison of the collected specimens with the herbarium material from the extant collections resulted in one species new for the science.

## DESCRIPTION

***Cerastium neoscardicum* Niketić, sp. nova**E subsectione *Cerastium* serie *Alpina* Borza.

I c.: Fig. 1a, Fig. 3(a-c), Fig. 4(a-c)

**T y p u s :** Flora serbica: Mt Šar-planina (Kodža Balkan: Ostrovica) apud urbem Prizren; 1400-2090 m; in rupestribus serpentinicis, ad Pinetum heldreichii, leg. M. Niketić & N. Stevanović 7.8.1987. (H o l o t y p u s : BEO; I s o t y p i : B, BD, C, LJU, SO).

**D i a g n o s i s :** Laxe vel subdense caespitosum, caulibus infracto-ascendentibus 10-25(35) cm altis, inferne saepe obscure sublignosis, noduloso incrassatis, parum radicosis, cum surculis sterilibus evolutis. Caules floriferi inferne sparse, superne subdense vel dense villosa-tomentosi. In axillis foliorum infimorum turiones steriles saepe evoluti. Folia caulina elliptico-lanceolata vel lanceolata, etiam obovato-lanceolata, raro inferna lineari-lanceolata, 1-2(3,5) cm longa, plerumque acuta, interdum unilateralia et falcata, viridia, ubique sparse vel subdense villosa ad villosa-tomentosa, superficies folii etiam nitide glanduliformi lepidota. Folia surculorum sterilium latiora, juvenilia cano-viridia, villosa ad villosa-lanata, margine barbulata, pilis mollibus 25-40  $\mu\text{m}$  latis obsita. Inflorescentia (1)3-6(15)-flora; rami inflorescentiae post anthesin saepe basi refracti. Bracteae omnes pellucido-scarioso-marginatae, interdum infimae herbaceae. Pedunculi dense crispato ad tomentoso-hirsuti, pilis mollibus et pilis patentibus, interdum etiam fastigiato-deflexo-subulatis obsiti; post anthesin saepe basi refracti, apicem versus paene recti. Sepala ovato-oblonga vel ovato-lanceolata, 7-8(9) mm longa, acuta, dense hirsuta cum pilis subulatis fastigiato-deflexis; pellucido-scarioso-marginata, post anthesin saepe intense contracta. Petala calyce fero duplo longiora. Capsula paene recta, calyce vix vel duplo longiora. Semina 1,3-1,8 mm longa, scabro-tuberculata. Floret a fine Julii in Augustum.

Differt ab *C. alpino* et *C. lanato*, caulibus basi robustioribus et crassioribus, superne tomentosis, surculis sterilibus longioribus, turionibus sterilibus frequentioribus, foliis angustioribus, inflorescentia ampliore, bracteis infimis plerumque scarioso-marginatis, pedunculis crispato ad tomentoso-hirsutis pilis mollibus obsitis, etiam seminibus majoribus.

A *C. decalvanti* distinctum turionibus sterilibus paucis, bracteis infimis interdum herbaceis, ramulis inflorescentiae sicut pedunculis post anthesin saepe basi refractis, pedunculis crispato ad tomentoso-hirsutis pilis patentibus obsitis, calyce tantum cum pilis hirsutis, ab subspecie *decalvans* etiam foliis juvenilibus barbulatis.

A *C. transilvanico* diversum habitu robustiore, caulibus circiter duplo crassioribus non superne refractis, surculis sterilibus sicut turionibus pluribus, indumento densiore, foliis juvenilibus cano-viridibus, pedunculis crispato ad tomentoso-hirsutis non crispato-hirsutis.

**E t y m o l o g i a :** Šar-planina = Mt Scardus [non *C. scardicum* T. Georgieff ex Soška in Glasn. Skopsk. Naučn. Društva 18: 233 (1938)].

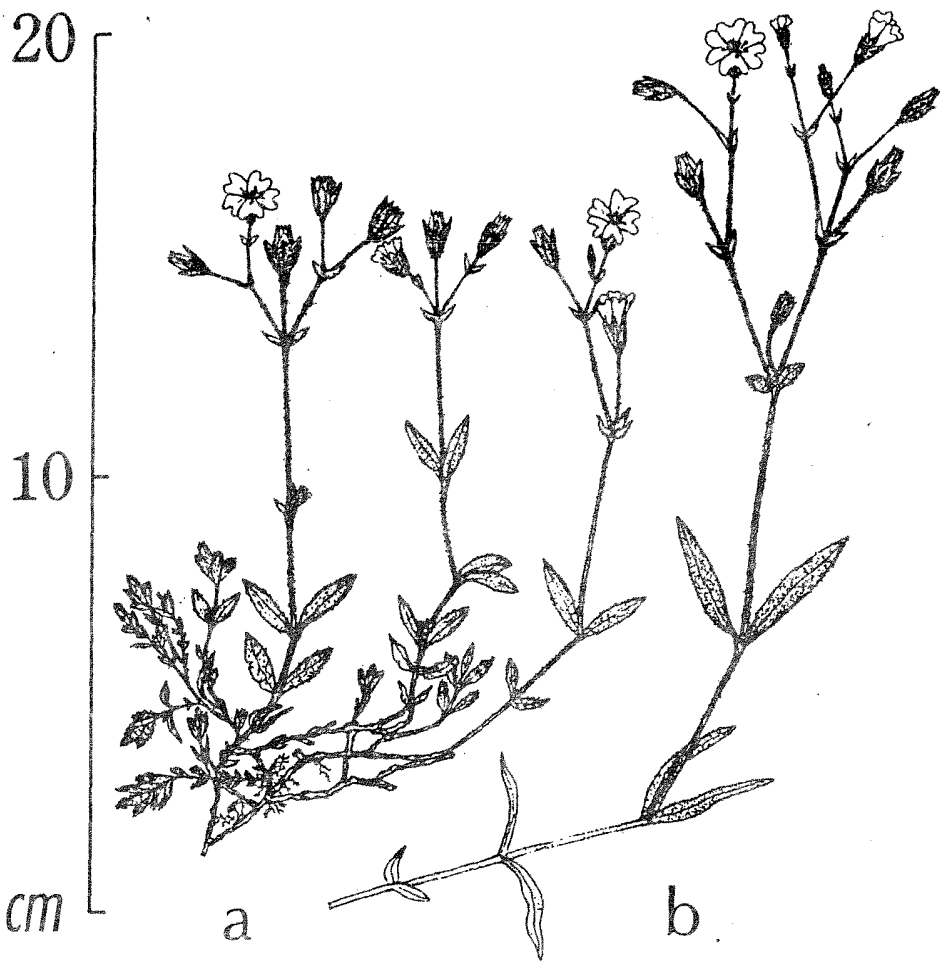


Fig. 1. - Morphological variability of *Cerastium neoscardicum* Niketić: a - habitus, b - flower stem of *Cerastium neoscardicum* Niketić f. *adenotrichum* Niketić

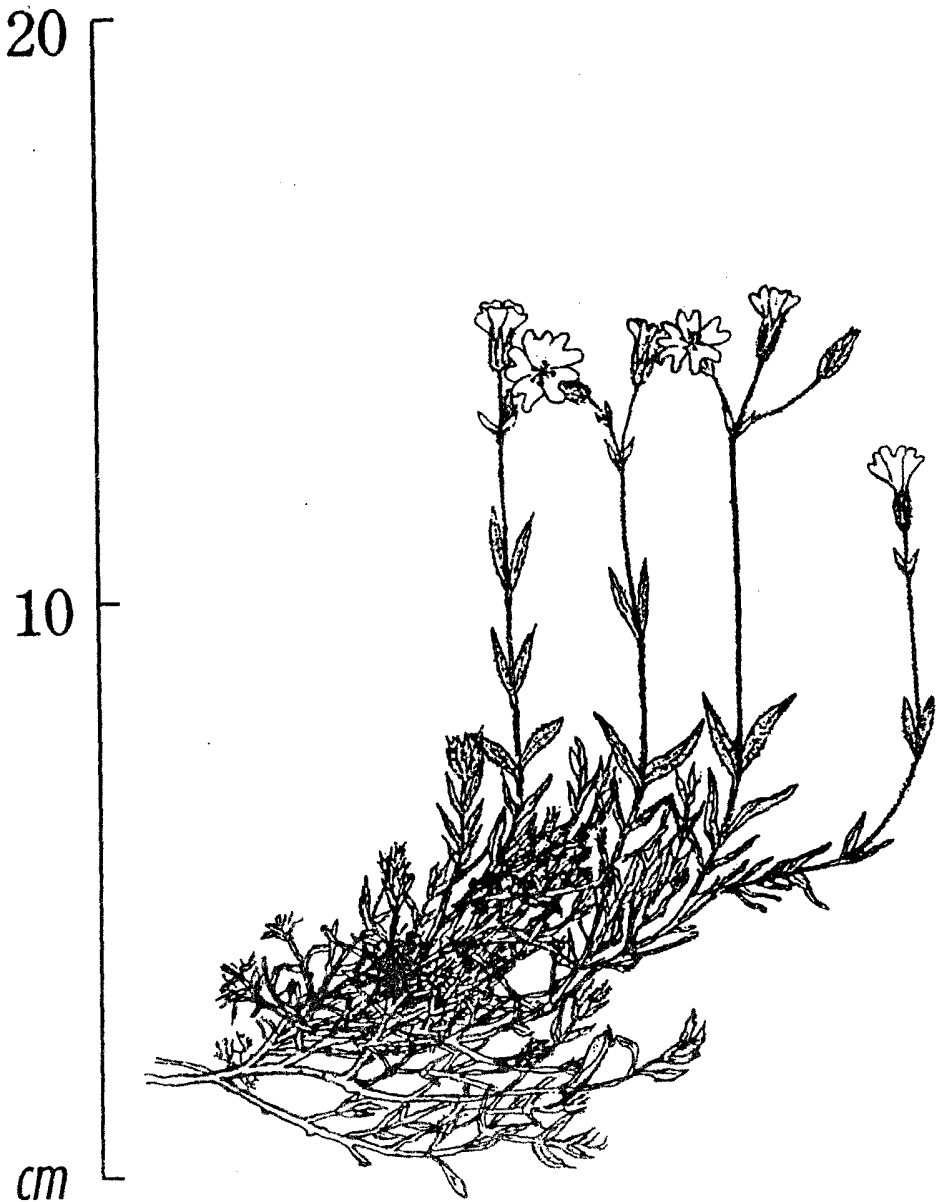


Fig. 2. – Habitus of *Cerastium neoscardicum* Niketić f. *glandulosum* Niketić

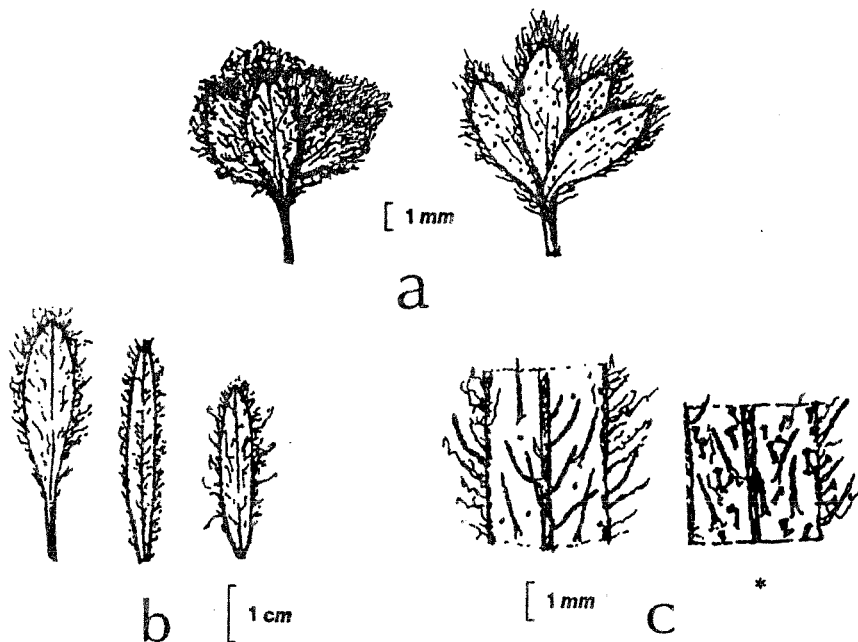


Fig. 3. – Morphological variability of *Cerastium neoscardicum* Niketić: a – young leaves, b – leaves, c – part of leaf – \**Cerastium neoscardicum* Niketić f. *adenotrichum* Niketić

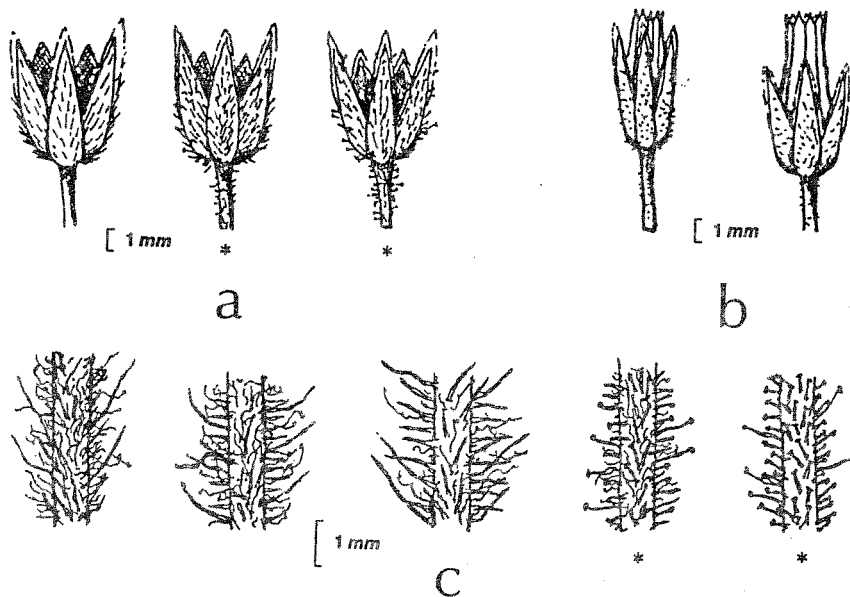


Fig. 4. – Morphological variability of *Cerastium neoscardicum* Niketić: a – sepals, b – capsules, c – part of peduncle – \**Cerastium neoscardicum* Niketić f. *glandulosum* Niketić

## Variabilitas:

f. *adenotrichum* Niketić, f. *nova* - 1 c.: Fig. 3c. - Typus: Flora serbica: Mt Šar-planina (Kodža Balkan: Ostrovica) apud urbem Prizren; 1400-2090 m; in rupestribus serpentinicis, ad Pinetum heldreichii, leg. M. Niketić & N. Stevanović 7.8.1987. (Holotypus: BEO). - Folia glanduloso-villosa ad glanduloso-villoso-tomentosa. Sepala et bracteae, saepe pedunculi et rami inflorescentiae glanduloso-pilosi.

f. *glandulosum* Niketić, f. *nova* - 1 c.: Fig. 1b, Fig. 2, Fig. 4a, Fig. 4c. - Typus: Flora serbica: Mt Šar-planina (Kodža Balkan: Ostrovica) apud urbem Prizren; 1400-2090 m, in rupestribus serpentinicis, ad Pinetum heldreichii, leg. M. Niketić & N. Stevanović 7.8.1987. (Holotypus: BEO). - Sepala et bracteae, saepe pedunculi et rami inflorescentiae glanduloso-pilosi.

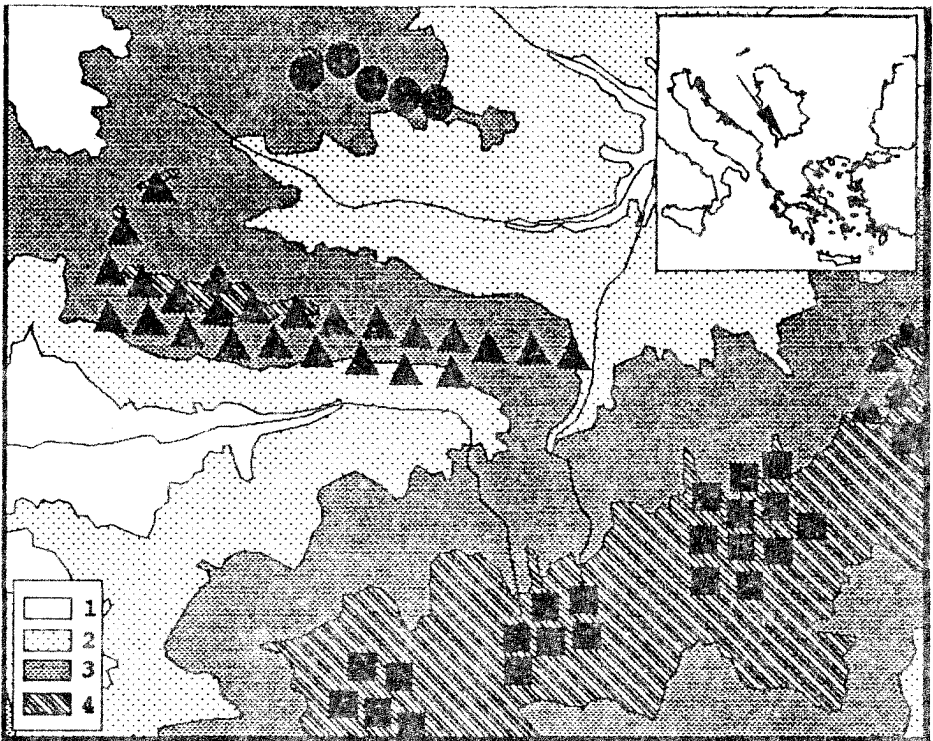
f. *neoscardicum*

Fig. 5. - Distribution of Some Species of Ser. *Alpina* in North-Eastern Part of Shar-planina Mountain: circle - *Cerastium neoscardicum* Niketić, triangle - *Cerastium decalvans* Schlosser & Vuk., square - *Cerastium alpinum* L.; altitude: 1 - < 1000 m 2 - 1000-1400 m, 3 - 1400-2000 m, 4 - > 2000 m

## DISCUSSION

*C. neoscardicum* morphology represents a transition from *C. alpinum* L. to *C. decalvans* Schlosser & Vuk. As both of the mentioned species can be found in the broader Mt Shar-planina region (Fig. 5), *C. neoscardicum* is most probably of a hybrid origin. The specific habitat conditions - the serpentine high-mountain geological



substratum, somewhat lower altitude than that of *C. alpinum*, palebark pine from Heldreich community – have induced have caused a complete independence of the *C. neoscardicum* population. Therefore it would be plausible to give this taxon a hybridogenous species status.

This standpoint is supported by the existence of an independent form with glandular-pubescent leaves, *C. neoscardicum* f. *adenotrichum*. In *C. alpinum*,<sup>1</sup> such a form has not been recorded so far, while some glandular-pubescent forms of *C. decalvans* have been found in an area limited to the central Serbia, Former Yugoslav Republic of Macedonia and Greece, and then only at lower altitudes.

It is interesting to note that a record of the considerable variability of the *C. alpinum* species (*C. alpinum* var. *nudipes* Fenzl ex Griseb.), together with its three forms – „Jusus 1 pumilus, uniflorus (alpinus)”, „Jusus 2 cyma pauciflora, bracteis latius scariosus”, „Jusus 3 cyma multiflora, bracteis herbaceis” – growing on the limestone ground of the Kobilica peak, exists in the first paper ever published on the flora of the Shar-planina mountain (Grisebach, 1843). On basis of these diagnoses and the materials collected by Doerfler on the limestone grounds of the Ljuboten peak, Wettstein (1882) concludes that the forementioned locality hosts the transitory forms from *C. alpinum* to *C. arvense* L. Since, in his paper, Griesebach wrongly identifies *C. decalvans* from the same locality as *C. arvense* var. *alpicotum* Fenzl (Grisebach, 1843), Wettstein's idea of *C. arvense* remains unclear. Essentially, Wettstein (1892) records *C. decalvans* (under the name of *C. lanigerum* Clem.) on the Kobilica peak only, disregarding Griesebach's record of it on the Ljuboten peak; in his paper there is also no recorded of *C. arvense* on any locality of the Shar-planina mountain.

Unfortunately, it was impossible for us to see Doerfler's materials, but having examined the extensive herbarium materials, we came to the following conclusion: The region of the Shar-planina mountain is one of the most important foci of the Ser. *Alpina*, represented by the greatest number of its species: *C. alpinum*, *C. decalvans*, *C. lanatum* Lam. and *C. neoscardicum*. Furthermore, it is the only place of introgression between the *C. alpinum* and *C. decalvans* species. Generally speaking, the morphological differences between these two species, when growing on limestone and silicate grounds, are much less marked than in the same species originating from the any other localities. Within each species, there are forms characteristic of their respective species, slightly reminiscent of the other species. The spontaneous hybrids have not been found. The serpentine are home to *C. neoscardicum* only, which is most probably of a hybrid origin. As for the Ser. *Cerastium* (*Arvensia*), its only species recorded with any certainty on the Shar-planina mountain is *C. banaticum* (Rochel) Heuffel.

#### ACKNOWLEDGEMENTS

I am specially grateful to Desa Djordjević for her illustrations and A. Petrović who made possible their adaptation for computer use.

<sup>1</sup> According to the current classifications (Greuter et al., 1984; Jalas, 1993) this form could be brought to connection with the glandular-pubescent *C. hekuravense* Jáv. of Albania. However, this taxon has been returned from Ser. *Alpina* to Ser. *Latifoliae* as an infraspecific form of *C. dinaricum* G. Beck & Szysz. (Niketić, 1995).

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## Re z i m e

MARJAN NIKETIĆ

***CERASTIUM NEOSCARDICUM*, NOVA VRSTA IZ SER. *ALPINA* SA PODRUČJA ŠAR-PLANINE, SRBIJA**

Prirodnjački muzej u Beogradu

Tokom florističkih istraživanja flore Šar-planine (Kodža Balkan: Ostrovica) naišli smo na jednu izolovanu populaciju iz roda *Cerastium* L. (Ser. *Alpina* Borza), koja se javlja na serpentinitskoj geološkoj podlozi. Nakon komparacije sakupljenih primeraka sa postojećim materijalom iz herbarskih zbirki zaključili smo da se radi o novoj vrsti za nauku.

*C. neoscardicum* po svojoj morfoloiji čini prelaz od *C. alpinum* L. ka *C. decalvans* Schlosser & Vuk. Kako na širem području Šar-planine rastu obe pomenute vrste, *C. neoscardicum* je najverovatnije hibridnog porekla. Specifični stanišni uslovi (serpentinitski visokoplaninski geološki supstrat, nešto manja nadmorska visina od one na kojoj se javlja *C. alpinum*, zajednica munike) usloveli su potpunu samostalnost populacije *C. neoscardicum*. Zbog toga je ovom taksonu ispravno dodeliti status hibridogene vrste.

U ranijim florističkim radovima sa područja Šar-planine (Grisebach, 1843; Wettstein, 1892) pominje se znatna varijabilnost vrste *C. alpinum*, kao i prelazni oblici ka drugim vrstama. Naša istraživanja populacija *C. alpinum* i *C. decalvans*, na silikatnim i krečnjačkim terenima Šar-planine, pokazala su da se kod obe vrste javljaju oblici koji striktno pripadaju jednoj, a samo po nekim karakteristikama podsećaju na drugu vrstu. Spontani hibridi nisu primećeni. Na serpentinitskim terenima raste isključivo *C. neoscardicum*, koji je najverovatnije hibridnog porekla.

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Original scientific paper

VLADIMIR RANDJELOVIĆ, NOVICA RANDJELOVIĆ, BOJAN  
ZLATKOVIĆ<sup>1</sup>

***ELEUSINE INDICA* (L.) GAERTN. - AN ADVENTIVE PLANT SPECIES IN  
THE FLORA OF THE SOUTHEASTERN YUGOSLAVIA**

Faculty of Technology, Leskovac  
<sup>1</sup>SISSB „Josif Pančić”, Faculty of Science, Novi Sad

Randjelović, V., Randjelović, N. and Zlatković, B. (1992-1993):  
*Eleusine indica* (L.) Gaertn. – an adventive plant species in the flora of the  
southeastern Yugoslavia. – Glasnik Instituta za botaniku i botaničke  
bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 71 - 76.

On the basis of floristic investigations, review of herbarium material  
(BEOU, Herb. Moesiacum Doljevac) and data from literature, new dis-  
tribution maps for Balkan Peninsula and Yugoslavia are presented for the  
adventive plant species *Eleusine indica*.

Key words: *Eleusine indica*, adventive plant species, locality

Ključne reči: *Eleusine indica*, adventivna biljna vrsta, lokalitet

INTRODUCTION

*Eleusine indica* is autochtonic weed species of the paleotropical floristical district  
(Weber, 1961), which arrived on the european ground at the end of the last century  
with seeds of american oil-flowers (Thellung, 1912) and wool (Weber, 1961). For  
the first time this species was noted for Europe in Belgium and Germany at the

beginning of this century (Ascherson & Graebner, 1899), and five years later it was found at the Balkan peninsula (Halaszy, 1904). The first finding of this species for Yugoslavia is coming from 1955. in Kotor, Piran and Umag (Hodak, 1959/60), and in Serbia three years later in the streets of Novi Sad (Slavnić, 1961). From that time during next thirty years this species was found in 25 localities on the territory of the Yugoslav countries. Except for mentioned localities this species was also recorded in the surrounding of Novi Sad (Obradović, 1966, Ivković, 1975, Čapaković & Ivković, 1978), Belgrade (Ivković, 1982, Jovanović, 1992) and Vukovar (Topić & Šegulja, 1978), spreading throughout Vojvodina, Mačva (Ivković, 1982), Slavonija and Baranja (Topić & Kusulja, 1989), from the district of Bačka and Srem to the other bank of the Danube and the Sava. It is also recorded in Turopolje region (Hulina, 1971, 1989), in surrounding of Zagreb (Marković & Hulina, 1970) and Split (Ilijanić, 1989), in the streets of Dubrovnik and Rijeka, and on some localities in Montenegro (Ivković, 1982). At the district of southeastern Yugoslavia this species was not found up to now.

## RESULTS AND DISCUSSION

During investigation of ruderal vegetation in southeastern Serbia we have found species *Eleusine indica* in some areas of plant communities *Lolio-Plantaginietum majoris* Bg. 1930. and *Polygono-Bidentetum* (Koch, 1926) Lohm. 1950. In first community this species is presented in the villages Kočane (near the road Niš-Priština), Doljevac, Pukovac (on the cattle market-place) and Koritnica and on the railway station in the city of Leskovac. Single turfs of this species we have found in the yards of mentioned villages. In second community this species has been met on the bank of the Jablanica river in village Živkovo, near by Leskovac. Also, it is found on sandy bank of the Toplica river in vegetation of alliance *Nanocyperion flavescens* W. Koch 1926 in the village Šarlinac.

### Distribution of *Eleusine indica* at the Balkan Peninsula (Fig. 1)

Croatia: Piran, Umag (Hodak, 1959/60), in the surrounding of Zagreb (Marković & Hulina, 1970), Turopolje region (Hulina, 1971, 1989), Vukovar (Topić & Šegulja, 1978), Rijeka, Dubrovnik (Ivković, 1982), Split (Ilijanić, 1989).

Bulgaria: Čustendil (Ganchev, 1963), Petrič, Čustendil (Stojanović et al., 1966), Znepole region, Struma valley, Thracian plain (Andreev et al., 1992).

Greece: (Hayek, 1933), Thessaloniki (leg. Heldreich, Herb. Norm. N. 1294 in Halaszy, 1904).

### Distribution of *Eleusine indica* in Yugoslavia (Fig. 2)

Montenegro: Kotor (Hodak, 1959/60), Donja Plavnica, Titograd, Sutomore, Stari Bar (Ivković, 1982), Budva (7.8.1994, Lakušić, D., BEOU).

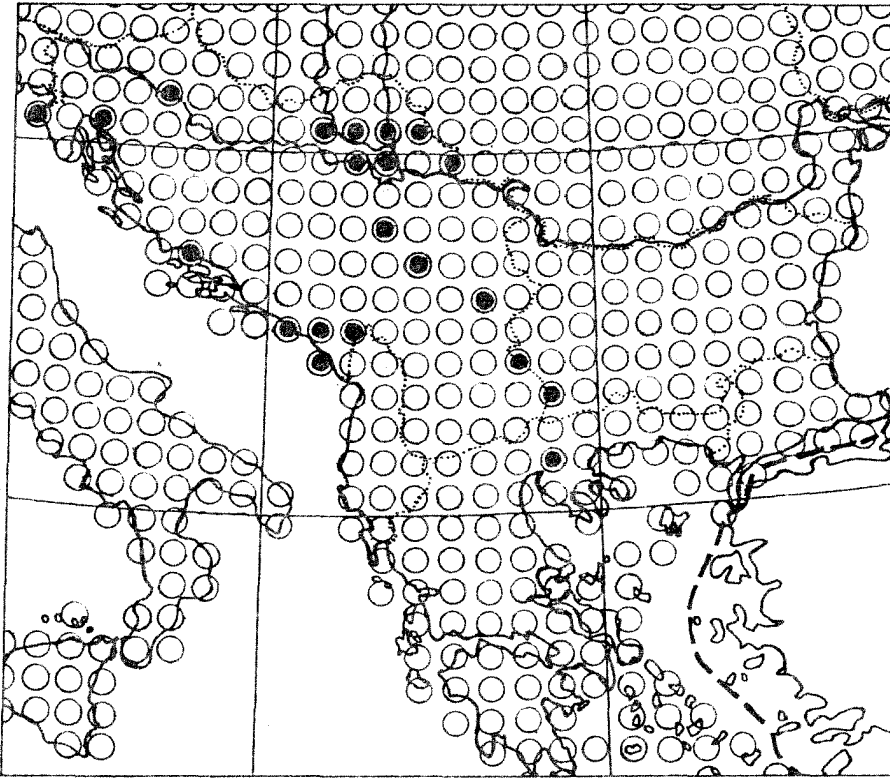


Fig. 1. – Distribution of adventive plant *Eleusine indica* at the Balkan Peninsula

North Serbia: Novi Sad (Slavnić, 1961, Ivković, 1975, Čapaković & Ivković, 1978), Petrovaradin (Obradović, 1966), Kač, Titel (Čapaković & Ivković, 1978), Stara Pazova, Nova Pazova, Batajnica, Beograd, Drenovac, Šabac (Ivković, 1982), Bela Crkva (10.8.1993, Lakušić, D., BEOU br. 331/93), Ruma (15.9.1993, Randjelović, V., Herb. Moesiacum Doljevac), Zrenjanin (28.9.1994, Randjelović, N., Herb. Moesiacum Doljevac), Mataruška banja (15.8.1994., Lakušić, D., BEOU), Pepeljevac near by Lajkovac (9.1994, Lakušić, D., BEOU).

Southeastern Serbia: Kočane, Pukovac (1977, Randjelović, N., Herb. Moesiacum Doljevac), Kočane (19.10.1994, Randjelović, V., Herb. Moesiacum Doljevac), Živkovo (9.10.1990, Randjelović, V. & N., Herb. Moesiacum Doljevac; Randjelović, 1992), Doljevac (27.8.1991, Randjelović, N. & V., Zlatković, B., Herb. Moesiacum Doljevac), Leskovac (22.9.1991., Randjelović, N. & V., Zlatković, B., Herb. Moesiacum Doljevac), Koritnica near by Bela Palanka (31.8.1983., Randjelović, N., Herb. Moesiacum Doljevac).

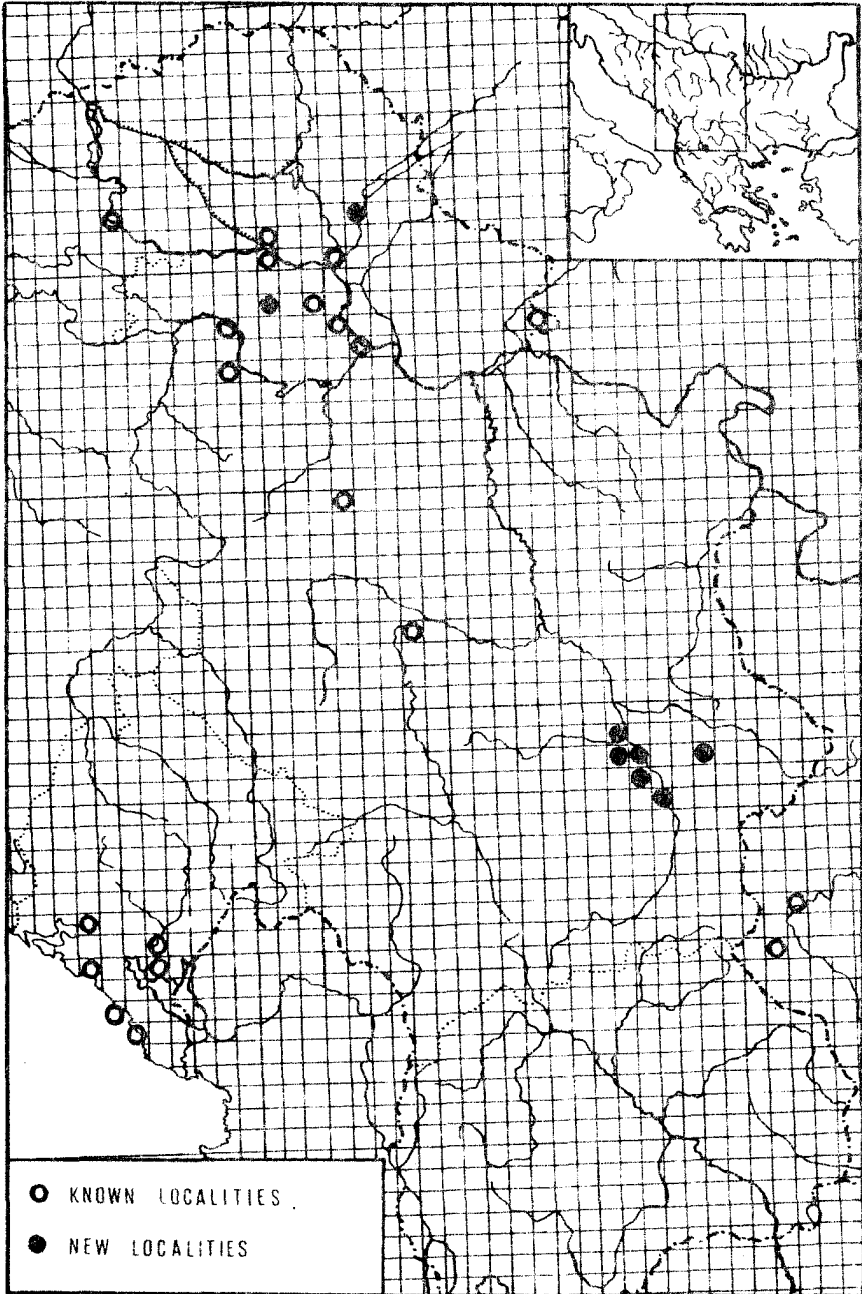


Fig. 2. – Distribution of adventive plant *Eleusine indica* in Yugoslavia

## CONCLUSION

Adventive plant species *Eleusine indica* was noted for Croatia, Montenegro and northern Serbia (Fig. 1) up to now. Species finding in the city of Leskovac and in villages Šarlinac, Doljevac, Kočane, Pukovac, Živkovo and Koritnica are new localities on the territory of Yugoslavia. In the southeastern Serbia as well as in the other parts of Yugoslavia this species is an inhabitant of ruderal plant communities.

## ACKNOWLEDGEMENTS

The author's gratitude belongs to Dr Ljerka Marković, professor at University of Zagreb, and mr DMITAR Lakušić, assistant at University of Belgrade.

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## Rezime

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### *ELEUSINE INDICA* (L.) GAERTN. - ADVENTIVNA BILJNA VRSTA JUGOISTOČNE JUGOSLAVIJE

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Adventivna biljna vrsta *Eleusine indica* je do sada nalažena u Hrvatskoj, Crnoj Gori i severnoj Srbiji, tako da nalazišta ove vrste u gradu Leskovcu i selima Šarlincu, Doljevcu, Kočanu, Pukovcu, Živkovu i Koritnici u jugoistočnoj Srbiji predstavljaju nove lokalitete i proširenje njenog areala na teritoriji jugoslovenskih zemalja. Kao i u ostalim delovima Jugoslavije i u jugoistočnoj Srbiji je ova vrsta stanovnik ruderalnih biljnih zajednica.



UDC 581.526.3 : 581.92 (497.16)  
Original scientific paper

JELENA BLAŽENČIĆ, ŽIVOJIN BLAŽENČIĆ<sup>1</sup>

## MACROPHYTES OF LAKE CRNO JEZERO ON DURMITOR MOUNTAIN (MONTENEGRO)

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<sup>1</sup>Faculty of Veterinary Medicine, University of Belgrade

Blaženčić, J., Blaženčić, Ž. (1992-1993): *Macrophytes of lake Crno Jezero on Durmitor Mountain (Montenegro)*. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XXVI-XXVII, 77 - 86.

In the frame of complex hydrobiological researches on the territory of National Park Durmitor, performed in the period od 1978-1984, botanical investigation of macrophytes has been undertaken in order to establish floristical composition and distribution of plant species and their populations found to live in the lakes. This article contains investigation results related only to lake Crno Jezero.

In lake Crno Jezero (Black Lake) 18 macrophytic species are recorded, of which 11 are vascular plants, 1 belongs to moss and 6 to charophytes. Besides, species distribution and analysis of ecological conditions are also presented in the paper.

Key words: Freshwater plants, *Charophyta*, flora, distribution, Yugoslavia.

Ključne reči: Slatkovodne biljke, *Charophyta*, flora, distribucija, Jugoslavija.

## INTRODUCTION

Mt Durmitor, named „Soa nebeska” (Pillar of the sky) by its inhabitants, inspires by splendid beauty of its dense forests, blooming pastures, deep canyons and crystal clear rivers, diverse paysages and numerous lakes. Ten lakes and plenty of smaller hydrographic objects make this mountain peculiar, what first have been noticed by Cvijić (1899) and, later on, confirmed by other researchers (Stanković, S., 1975, 1985, 1992; Bešić, Z., 1963). The lakes are situated at the altitude between 1409 and 1788 m above sea level and differ mutually by its aspect, largeness, hydrological characteristics and genesis (Stanković, S., 1975, 1992). This diversity of biotops resulted in diversity of plant world living there, with regard to flora and space distribution of species and their populations.

Among Mt Durmitor lakes, lake Crno Jezero (Black Lake) distinguishes from the others by its largeness, genesis, hydrographical characteristics as well as by its touristic and economic importance. In consideration to diverse life conditions existing in the lake, floristical diversity and distribution of macrophytes were supposed to be significantly different from the other biotops of Mt Durmitor. In attempt to analyse this phenomenon, the authors carried out comprehensive investigation during summers 1982 and 1983.

## MATERIAL AND METHODS

Sampling for floristical analysis and examination of space distribution is performed by transverse profiles and transects methods, using hook- and rake-type devices constructed by the authors and described in previously published papers (Blaženčić, J. & Blaženčić, Ž., 1991).

Charophytes determination is achieved according to Corillion, R., 1957, 1975, Gollerbah, M.M. & Krasavina, L.K., 1983 and by consulting Monograph and Iconograph by Wood, R.D. & Imahori, K., 1964, 1965. Vascular plants are defined using keys from Flora SR Srbije 1-8 (Josifović, M., 1970-1977), Flora SSSR 1 (Komarov i Il'in, 1934), Illustrierte Flora von Mittel-Europa (Hegi, D.G., 1965).

Bathymetric chart of lake Crno Jezero is taken over from Stanković (1975).

## RESULTS AND DISCUSSION

*Principal characteristics of biotop.* Lake Crno Jezero lies near by village Žabljak, on Mt Durmitor, at the altitude of 1418 m above sea level. The lake's water level varies seasonally to a great extent, from 8 to 10 metres. The lake basin is divided in larger part, trending NE and named Big Lake, and smaller part towards SW, named Little Lake (Fig. 1).

Big Lake's littoral is almost entirely slightly sloped up to the bottom what is very favourable for macrophytic vegetation development. On the contrary, Little Lake's littoral along lakeshore is steep and rocky. Anyway, lake Crno Jezero lies on the limestone bedrock, with maximal depth of 24.5 m in Big Lake and 49.1 m in Little Lake.

Very transparent water and low organic production make this lake belong to oligotrophic and alpine-type. According to Purić (1983) the lake's water appears to be calcium-bicarbonate-type of freshwaters, with slightly alkaline reaction and pH-va-

lues range between 7.7 and 8.06. Water temperature fluctuates from 2.3°C in December, to 18°C in summer months. Being characterized by high oxygen content, low BOD<sub>5</sub> - values, absence of ammonium and nitrate ionic species, lake Crno Jezero seems to have a clean water.

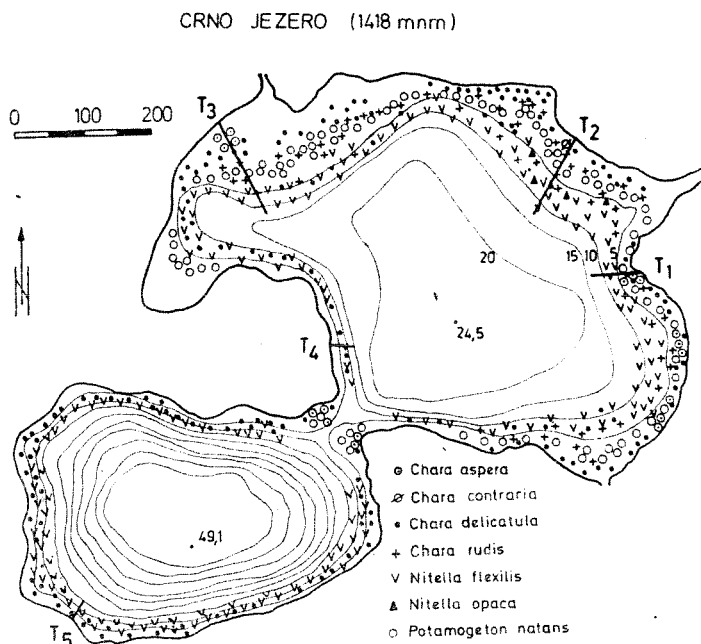


Fig. 1. - Distribution of dominant species in lake Crno Jezero

*Floristical composition and macrophytes distribution in lake Crno Jezero.* By floristical analysis of samples collected in the lake have been evidenced 18 species of which 6 species of algae belonging to division of *Charophyta*, 11 species of vascular plants and 1 species of moss.

According to available literature data, species recorded by the authors to live in lake Crno Jezero have not been evidenced so far. Most frequently mentioned genera are *Chara*, *Potamogeton*, *Myriophyllum* and *Ranunculus*. (Ivanović et al., 1968; Petković, Sm. & Petković, St., 1972; Petković, Sm., 1981). Rohlena (1942) reported *Nuphar luteum* L., *Potamogeton mucronatus* Schrad., *P. pectinatus* L., which have not been found by the authors to grow in the lake. Mentioned data point out to the fact that macrophytic flora in lake Crno Jezero has not been well known

as well as that some species have been disappeared with time from this locality. Only two species (*P. filiformis* and *P. natans*) from floristic list, established on the base of the author's investigation results, have been known in the lake's flora before. All others are new for the lake's flora, even for Mt Durmitor's flora, while *Nitella flexilis* is new species for the flora of Montenegro (Blaženčić, J., Blaženčić, Ž., Cvijan, M., 1991).

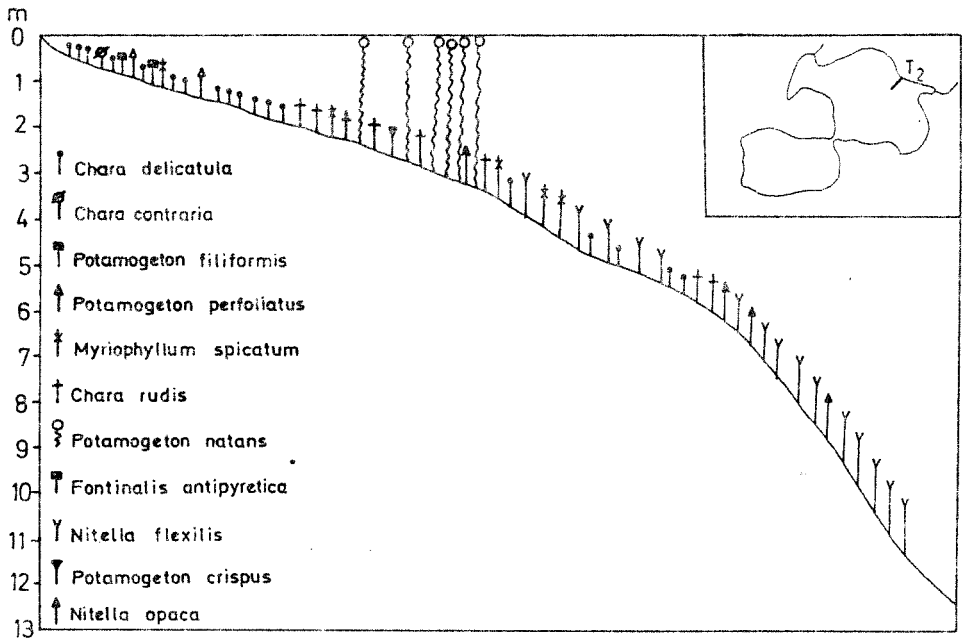


Fig. 2. – Distribution of aquatic plants along transect T2

In shallow water of the Big Lake's littoral, up to 2 m of depth, in detritus and shallow mud, between rocks, populaitons of *Chara aspera* Deth. ex Willd., *Chara contraria* A. Br. and *Chara delicatula* Ag. occur mosaically dispersed, making pure populations or mixed populations associated with *Chara rudis* A.Br., *Potamogeton pusillus* L., *P. filiformis* Pers., *P. perfoliatus* L., *P. natans* L. and *Ranunculus paucistamineus* Tsch. (Figs 1, 2, 3, 4).

Almost around the entire Big Lake (Northeastern part of lake Crno Jezero), at the depths between 2 and 4 m, extends the zone of floating *Potamogeton natans*. Before and behind and partly inside this zone, groupings of *Chara rudis* and *Ch. delicatula* are recorded (Figs 1, 2, 3, 4).

Mixed populations, developed between 2 and 6 m of the depths, beside mentioned species, include also *Myriophyllum spicatum* L., *P. perfoliatus*, *P. crispus* L. and *Nitella flexilis* Ag.

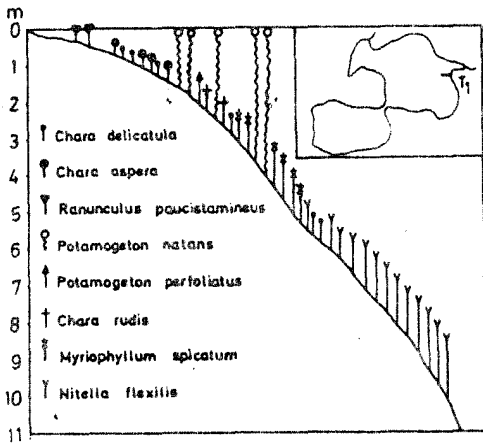


Fig. 3. – Distribution of aquatic plants along transect T<sub>1</sub>

Further vegetation zone in Big Lake lies at depth of 6 to 10 m, building high and dense populations of *Nitella flexilis* and *N. opaca* Ag. (Figs 1, 2, 3, 4). In adjacent parts of this zone, toward the previous zone, occur *Nitella opaca* associated with *Ch. rudis* and *Myriophyllum spicatum*, and *Nitella flexilis* in association with *Ch. rudis*, *Ch. delicatula*, *P. natans*, *P. perfoliatus* and *Miyriophyllum spicatum*. Deeper of 6 m, only populations of *N. flexilis* and *N. opaca* are found to grow. The lower distribution limit of these species (11 m) is, at the same time, the lower distribution limit of macrophytic vegetation at all in Big Lake.

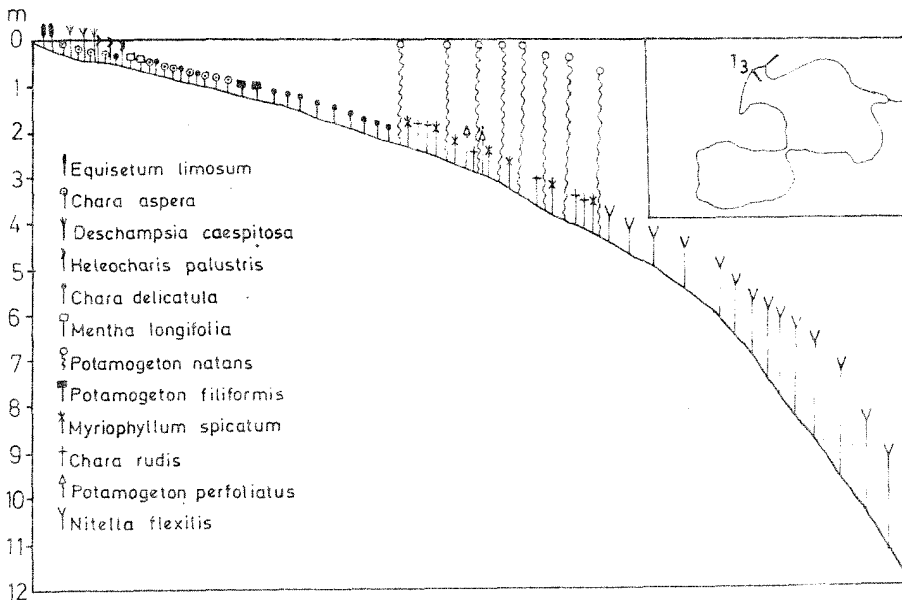


Fig. 4. – Distribution of aquatic plants along transect T<sub>3</sub>

Macrophytic flora in Little Lake (Southwestern part of lake Crno Jezero) is represented with populations of *Chara delicatula* and *Nitella flexilis* (Figs 1, 6) which appear at the depths more than 4 m, caused by steep and rocky lake shorelines. Between 4 and 6 m, *Ch. delicatula* is more abundant than *Nitella flexilis*. With depth *Nitella flexilis* becomes more dominant, so that pure populations of this species appear somewhere at 5 m yet, but always at depths more than 8 m. Maximal depth of its extending recorded so far is 14.5 m. Identical floristic composition and similar vertical distribution exist also in Big Lake, in habitats with similar characteristics such as steep and rocky lake bottom (Figs 1, 4).

Northwestern part of the lake, where the Mlinški Potok enters in, is very interesting from floristical and phytocoenological viewpoint (Figs 1 - T<sub>3</sub>, 4). This lakeshore is under the water in the case of high water level or covered with swampy meadows under the conditions of low water level.

In the investigation period the lake bottom was covered with dense carpet of *Chara aspera* in combination with following species: *Equisetum limosum* (L.) Roth., *Heleocharis palustris* R.Br., *Mentha longifolia* Huds., *Deschampsia coespitosa* Beauv., *Potamogeton filiformis* Pers. and *P. perfoliatus*.

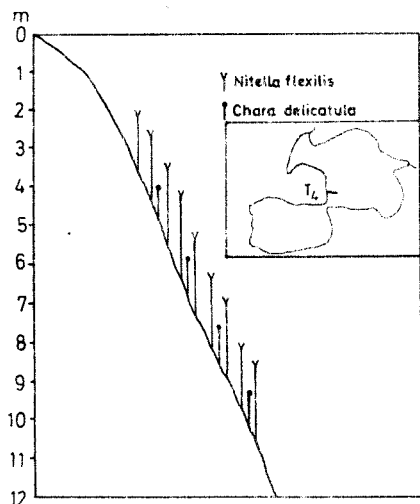


Fig. 5. -- Distribution of aquatic plants along transect T<sub>4</sub>

Being subjected to significant seasonal water level oscillations, some parts of lakeshore are occasionally under the water or overgrown by swampy meadows after the drainage. The soil remains wet for some time, protected from fast drying up by vegetation that lives on after the water drains away. Aquatic plants withstand out of the water developing different adaptations to these conditions. Development of terrestrial forms of aquatic plants is the most frequent adaptation to dry conditions, found by the authors, during earlier investigations on the other places, in following species: *Nymphaea alba* L., *Nuphar luteum* Sm., *Nymphoides flava* Hill., *Potamogeton natans*, *P. gramineus* L., *Myriophyllum spicatum*, *M. verticillatum* L., species belong to genus *Trapa* L., *Hippuris vulgaris* L., *Ranunculus aquatilis* L. (Janković, M. & Blaženčić, J., 1968; Janković, M. et al., 1980).

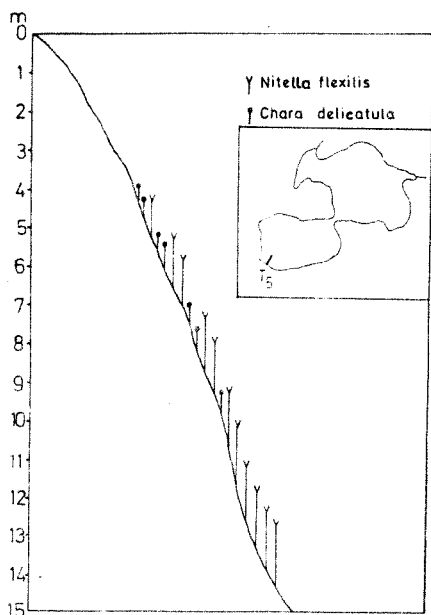


Fig. 6. – Distribution of aquatic plants along transect T<sub>5</sub>

In the identical or similar habitats, charophytes *Chara aspera* and *Ch. vulgaris* L. develop occupying lower layer of marshy meadow vegetation. These species produce oospore thus surviving in wet mud disadvantageous life conditions. Oospores are highly resistant, staying viable during the winter period. Some charophytes produce bulbils in attempt to adapt themselves to unfavourable life conditions, like *Chara aspera* and *Ch. delicatula* in lake Crno Jezero. In re-established and suitable life conditions, charophytes grow vegetatively, most rapidly by developing branches, slowly from bulbils and quite slowly from spores.

### CONCLUSION

Botanical investigation including analysis of floristical composition and space distribution of macrophytic vegetation in lake Crno Jezero on Mt Durmitor (Montenegro) is performed during summers 1982 and 1983.

Lake Crno Jezero (Black Lake) is situated at the altitude of 1418 m above sea level, on the limestone bedrock.

The water enters into the lake from permanent tributary and sublacustrine springs, and leaves the lake through periodical outlet, renewing the water mass, i.e. providing water flow through the lake basin. Great transparency (15 m) and low organic production include this lake in deep (24.5 m in Big Lake and 49.1 m in Little Lake) and oligotrophic, alpine-type of the lakes.

The lake water is calcium-bicarbonate-type with slightly alkaline reaction (pH 7.7-8.06).

Floristical analysis of samples collected along the transects shows the presence of 18 macrophytic species, of which 6 species of algae belonging to division of *Charophyta*, 11 species of vascular plants and 1 species of moss. Following species are recorded: *Chara aspera*, *Ch. contraria*, *Ch. delicatula*, *Ch. rudis*, *Nitella opaca*, *N. flexilis*, *Ranunculus paucistamineus*, *Heleocharis palustris*, *Equisetum limosum*, *Potamogeton natans*, *P. pusillus*, *P. perfoliatus*, *P. filiformis*, *P. crispus*, *Myriophyllum spicatum*. In some places along shoreline (Fig. 4), covered with dense populations of *Chara aspera*, marshy meadow's species *Mentha longifolia* and *Deschampsia coespitosa* are also found. Moss species *Fontinalis antipiretica* appears in few places in the lake. Only two among species mentioned above, have been accounted in flora of the lake before (*Potamogeton natans* and *P. filiformis*). Some species like *Nuphar luteum*, *P. pectinatus* and *P. mucronatus*, recorded by Rohlena (1942), have not been found during the author's investigations, on the basis of what could be concluded that they disappeared in the mean time.

Emergent plant zone does not exist caused by significant fluctuation of lake water level, between 8 and 10 m annually.

Floating plants represented by species *Potamogeton natans* develop only in the part of lake named Big Lake, at the depth of 2 to 4 m.

Submersed plants zone extends from coastal region up to the depth of 11 m in Big Lake or up to 14.5 m in Little Lake (Fig. 1) and is build of charophytes, which grow at all depths up to lower limit of macrophytes distribution, and vascular plants, which grow up to 5-6 m of depth (Figs 1-6).

In the lake shallows, up to 2 m, on shallow mud or detritus, in the zone of significant daily and seasonal fluctuation of ecological conditions, low tufts of charophytes develop scarcely (*Ch. contraria*, *Ch. delicatula*) or in a form of low carpet (*Ch. aspera*). With depth, number of species that participate in macrophytes vegetation decreases, plant carpet becomes more compact and individual plants more elongated. In the zone between 2 and 6 m are found *P. natans*, *P. perfoliatus*, *M. spicatum*, *Ch. rudis* and others.

Particular zone and lower limit of macrophytes distribution in lake Crno Jezero, is represented by dense, high (up to 1 m) submersed meadows built of species *Nitella opaca* and *Nitella flexilis* (Figs 1, 3, 4).

Vertical distribution of macrophytes, described in the paper, is characteristic of the lakes with clean and highly transparent water what represents another evidence which supports the state of Blindow (1992).

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## Re z i m e

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### MAKROFITE CRNOG JEZERA NA PLANINI DURMITOR (CRNA GORA)

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Floristički sastav i distribuciju makrofita u Crnom jezeru na Durmitoru proučavali smo u leto 1982. i 1983.

Crno jezero nalazi se na nadmorskoj visini od 1418 metara. Leži na terenu krečnjačkog sastava. Ima stalnu pritoku, povremenu otoku i ponore na dnu pa pripada protočnom tipu jezera. Velika providnost vode (15 m) i niska organska produkcija uvršćuju Crno jezero u duboka (24.5 m Veliko i 49.1 m Malo jezero), oligotrofna jezera alpskog tipa. Voda Crnog jezera je kalcijum-bikarbonatna, slabo bazna reakcije (pH 7.7-8.06).

Florističkom analizom uzraka konstatovali smo prisustvo 18 vrsta makrofita, od kojih 6 vrsta pripada algama razdela Charophyta, 11 vrsta vaskularnim biljkama i jedna mahovinama. Konstatovane su sledeće vrste: *Chara aspera*, *Ch. contraria*, *Ch. delicatula*, *Ch. nudis*, *Nitella opaca*, *N. flexilis*, *Ranunculus paucistamineus*, *Heleocharis palustris*, *Equisetum limosum*, *Potamogeton natans*, *P. pusillus*, *P. perfoliatus*, *P. filiformis*, *P. crispus*, *Myriophyllum spicatum*. Na pojedinim mestima uz obalu (Sl. 4) gde dno prekriva gusta populacija *Ch. aspera*, zabeležene su i biljke močvarnih livada *Mentha longifolia* i *Deschampsia coespitosa*. Na više mesta u jezeru nalazi se i mahovina *Fontinalis antipiretica* L.

Od navedenih vrsta samo dve (*Potamogeton natans* i *P. filiformis*) su bile poznate za Crno jezero. Neke vrste kao npr. *Najas luteum*, *P. pectinatus* i *P. mucronatus*, koje je zabeležio R o h l e n a (1942) nisu konstatovane u toku naših istraživanja na osnovu čega bi se moglo zaključiti da su tokom vremena iščezle.

Zona emerznih biljaka u Crnom jezeru se ne formira što je pored ostalog posledica veoma izražene astatičnosti nivoa jezerske vode. Godišnja amplituda kolebanja iznosi 8-10 metara.

Flotantne biljke predstavljene su vrstom *P. natans* koja se razvija samo u delu jezera koje se naziva Veliko, na dubini od 2-4 metra.

Zona submerznih biljaka prostire se od oblaskog regiona do dubine od 11 metara u Velikom, odnosno do 14,5 metara u Malom jezeru (Fig. 1). Zonu submerznih biljaka grade harofite i vaskularne biljke. Harofite se nalaze na svim dubinama do donje granice rasprostranjenja makrofita a vaskularne biljke samo do dubine od 5-6 metara (Figs 1-6).

U plićim delovima jezera (do 2 m), na plitkom mulju ili detritisu između kamenja, u zoni izrazitog dnevno-noćnog i sezonskog kolebanja intenziteta ekoloških faktora, razvijaju se niske forme harofita u vidu razbacanih žbunova (*Ch. contraria*, *Ch. delicatula*) ili niskih tepiha (*Ch. aspera*).

Sa povećanjem dubine broj vrsta u vegetaciji makrofita se smanjuje, biljni pokrivač dna je sve kompaktniji, a individue sve izduženije. U zoni između 2 i 6 m dubine konstatovane su vrste *P. natans*, *P. perfoliatus*, *M. spicatum*, *Ch. rudis* i dr.

Posebnu zonu, donju granicu rasprostranjenja makrofita u Crnom jezeru, čine guste, visoke (do 1 m) podvodne livade koje grade vrste *Nitella opaca* i *Nitella flexilis* (Figs 1, 3, 4).

Opisana vertikalna distribucija makrofita karakteristična je za jezera sa čistom i visoko transparentnom vodom i još je jedan dokaz više u prilog mišljenju koje iznosi B l i n d o w (1992).

Ovim istraživanjima stvorena je baza podataka o florističkoj raznovrsnosti i prostornom rasporedu populacija konstatovanih vrsta. Na taj način stvorena je dobra osnova za dopunu florističke liste i praćenje dinamike promena u sastavu flore, a to, uz korišćenje drugih botaničkih i ekoloških kriterijuma može da bude dobar indikator stanja i promena u ekosistemu Crnog jezera.

UDC 553.7.031.2 : 582.2 (497.11)  
Original scientific paper

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## ALGAE IN THERMOMINERAL WATERS OF NIŠKA BANJA SPA (SERBIA)

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Cvijan, M. (1992-1993): *Algae in thermomineral waters of Niška Banja spa (Serbia)*. – Glasnik Inistuta za botaniku i botaničke bašte Unierziteta u Beogradu, Tom XXVI-XXVII, 87 - 96.

During September 1982, July 1983, April 1985 and November 1992 algological samples from thermomineral waters of spring Suva Banja and artificial, concrete canal with the water originated from Main spring were collected.

The qualitative and relatively quantitative composition of algal communities were analysed, as well as ecological characteristics of some presented taxons. On the basis of algological analyses, the algological types of thermomineral waters in Niška Banja spa are given.

Key words: algae, thermomineral waters, Niška Banja spa.

Ključne reči: alge, termomineralna voda, Niška Banja.

### INTRODUCTION

Algological investigations of thermomineral waters in Serbia are of the great scientific interest in consideration of the great number of thermomineral springs in our country (Marković, 1980), which differ mutually by its origin as well as by its physical and chemical characteristics (Godić & Radić, 1963).

Because of their practical importance (medical treatments, tourism and recreation, heating etc.) some of those springs are more or less well investigated from different point of view (amount of water, chemical composition of water, its temperature and radioactivity etc.). Biological investigation has not been performed and reviewing literature we have found only two scientific papers dealing with algae from thermomineral waters of Niška Banja spa (Petrovska, 1969; Cvijan & Blaženčić, 1986).

## MATERIAL AND METHODS

Samples of algae were collected from thermomineral water of Suva Banja spring and artificial, concrete canal with the water originated from Main spring.

Samples were collected in September 1982, July 1983, April 1985 and November 1992 from different places, mostly from concrete and stones which were under water flow, or from concrete or stones which were sprayed by thermomineral water, as well as from surfaces in the area of warm steam.

Collected material was preserved immediately in 3-4% formaldehyde.

Algological material was examined by Diastar<sup>TM</sup> photomicroscope. The most of algae were investigated directly from collected material except the diatoms which were prepared by standard method with sulphuric acid (Hustedt, 1930; Patrick & Reimer, 1966).

Financial support for the laboratory investigation was provided by the Contract No. 0321 Serbian Ministry of science.

## RESULTS AND DISCUSSION

### **The general features of Niška Banja spa and general and particular features of thermomineral springs**

Niška Banja spa is situated in central part of Serbia, 10 km south-east from the town Niš.

The surrounding of Niška Banja is built of limestone, sediments (clay and conglomerate) and river sands (Marković, 1980).

The climate of Niška Banja is moderate-continental with more features which are conditioned by great surfaces under the forests, orchards and vineyards. Good connections with Niš, as well as the presence of thermomineral springs, improved the development of tourism.

The existence of thermomineral water is caused by specific geological factors and by positions of limestone toward the sediments of clay and conglomerates (Đurović, 1963).

Exterior factors (the amount of fails, the time and intensity of snow melting), specially caused by presence of limestone, influence a lot at temperature and chemical composition of thermomineral water.

In Niška Banja spa there are two thermomineral springs: Main spring and Suva Banja spring.

**Main spring** givs 35-120 l/sec of water, tempeurture of 38.2-38.5°C. This water is specially characterized by ions of  $\text{Ca}^{++}$  and  $\text{HCO}_3^-$  and belongs to group of homoeothermal water with moderate radioactivity (Marković, 1980).

**Suva Banja** givs 14-42 l/sec of water, temperature of 12-37°C. Great variability of temperature and amount of water is consequenced by considerable influence of external factors (Marković, 1980). The water is specially characterized by ions of  $\text{Ca}^{++}$  and  $\text{HCO}_3^-$  and belongs to group of homoeothermal water with low radioactivity (Marković, 1980).

The great amount of water of the springs (specially of the Main spring) is captured. This water is using for balneotherapy and only one part of water (surplus that are not utilized) is under the influence of natural factors. But, the influence of man is very expressive all over the year, specially during the summer when there are a great number of visitors.

The samples were collected from different places which we can put in two wholes;

A. Concrete canal. The artificial canal with water from Main spring. The temperature of thermomineral water along the concrete canal are given in Tab. 1.

*Tab. 1. – Temperature of water (C<sup>0</sup>) in concrete canal in different periods of investigations*

Year of collecting	"spring"	central part	end of canal
1982.	38.0	35.5	31.4
1983.	38.2	36.7	33.5
1985.	37.9	34.6	30.0
1992.	34.6	27.8	23.2

The pH reaction of thermomineral water at any time of sampling was between 7.0 and 7.2.

B. Suva Banja. Typical representative of cave-therms (Martinović & Kostić, 1977). Thermomineral water is flowing out of the cave but the entrance of the cave was closed for visitors with metal bars and in 1992 by metal plates. Because of that, the samples were collected only from narrow zone around the entrance of the cave and they were characterised by the temperature of thermomineral water between 32.3 and 37.0°C and pH reaction between 7.1 and 7.2. However, in algological analyzed we distinguished two different groups of samples:

a. The samples from the surfaces inside the cave with constant tempeurture and pH, constant flow of thermomineral water and, specially in 1992, with strong shaded surface of concrete.

b. The samples from the surface out of the cave with some changeable temperature, pH and water flow, and not strong shaded surface of concrete.

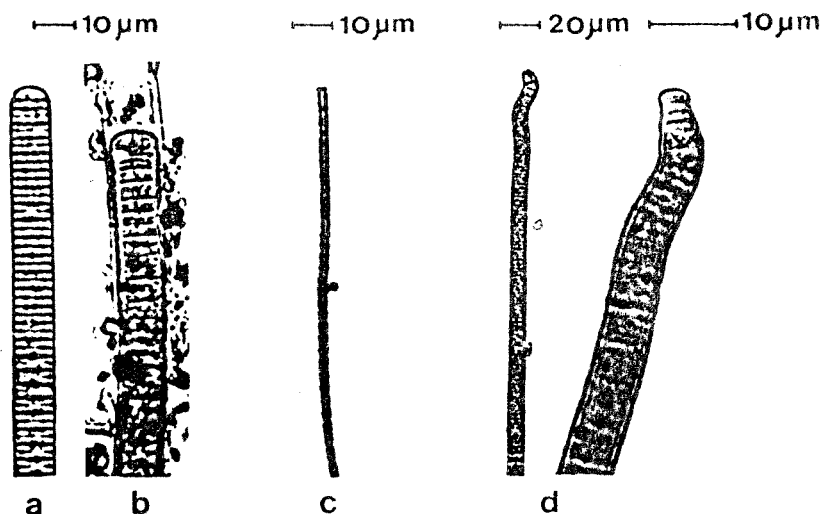


Fig. 1. – Some dominant representatives of algae in thermomineral water of Niška Banja spa

- a – *Oscillatoria limosa*  
 b – *Oscillatoria limosa* f. *phormidioides*  
 c – *Phormidium foveolarum*  
 d – *Oscillatoria cortiana*

#### Algae in thermomineral waters of Niška Banja spa

**Concrete canal.** By the examination of the samples from concrete canal in Niška Banja spa, the presence of algae from 3 divisions was established: *Cyanophyta*, *Bacillariophyta* and *Chlorophyta* (Tab. 2 and Tab. 3).

In the first three years of our investigation some differences in qualitative and quantitative compositions of algae exist, but they were insignificant. In November 1992 we established qualitative and quantitative poverty of algae. On the other hand, *Oscillatoria cortiana* and *Phormidium foveolarum* were dominant. Beside that, *O. cortiana* was not found in any samples collected before November 1992.

All differences are the consequence of specific, unusual conditions for developing of algae in November 1992. Namely, in the period of low temperature (near 0°C), the flowing of thermomineral water through the canal does not exist. Because of that, in November 1992 thermomineral water was not flowing through the canal over the night and the conditions for development of some stable algal communities were not present. Some taxons (*Phormidium foveolarum*, partly *Oscillatoria limosa*, *O. limosa* f. *phormidioides* etc.) are more or less well accomodated to changeable conditions. But, some taxons were eliminated. On the contrary, some new taxons were developed for the first time (*Gloeocapsa montana*, *Oscillatoria anguina*, *O. cortiana*, *O. formosa* and *O. limosa* f. *disperso-granulata*). In that period *O. limosa* as a dominant species in the earlier periods, was substituted by *O. cortiana*.

Tab. 2. - Qualitative and relatively quantitative composition of algae in thermomineral water of concrete canal of Niška Banja spa

Taxons	23-30	30-35	35-38
	In C <sup>0</sup>		
<i>Aphanocapsa thermalis</i> (Kütz.) Brügg.	-	1	r
<i>Calothrix elenkinii</i> Kossinsk.	-	-	r
<i>Galothrix thermalis</i> (Schwabe) Hansg.	-	1	r
<i>Gloeocapsa minuta</i> (Kütz.) Hollerb.	1	2	-
<i>Gloeocapsa crepidium</i> (Rabh.) Thur.	-	-	r
<i>Gloeocapsa montana</i> Kütz.	1	-	-
<i>Gloeocapsa punctata</i> Näg.	1	r	-
<i>Gloeotehece rupestris</i> (Lyngeb.) Born.	-	-	r
<i>Merismopedia punctata</i> Meyen	-	2	-
<i>Oscillatoria anguina</i> (Bory) Gom.	1	-	-
<i>Oscillatoria amphibia</i> Kütz.	-	r	r
<i>Oscillatoria brevis</i> Kütz.	r	1	-
<i>Oscillatoria chalybea</i> (Mert.) Gom.	-	-	1
<i>Oscillatoria cortiana</i> (Menegh.) Gom.	3	-	-
<i>Oscillatoria formosa</i> Bory	1	-	-
<i>Oscillatoria limosa</i> Ag.	1	3	3
<i>Oscillatoria limosa</i> f. <i>phormidioides</i> (Rabh.) Elenk.	1	2	2
<i>Oscillatoria limosa</i> f. <i>disperso-granulata</i> (Schkorb.) Elenk.	1	-	-
<i>Oscillatoria okenii</i> Ag.	-	r	1
<i>Oscillatoria princeps</i> Vauch.	-	-	1
<i>Oscillatoria tenuis</i> Ag.	1	-	1
<i>Oscillatoria tenuis</i> var. <i>symplociformis</i> (Hansg.) Elenk.	-	-	1
<i>Oscillatoria terebriformis</i> (Ag.) Gom.	-	r	1
<i>Phormidium ambiguum</i> Gom.	-	-	r
<i>Phormidium ambiguum</i> f. <i>maius</i> (Lemm.) Elenk.	-	-	2
<i>Phormidium favosum</i> (Bory) Gom.	-	1	r
<i>Phormidium fovoelatum</i> (Mont.) Gom.	5	2	3
<i>Phormidium mole</i> (Kütz.) Gom. f. <i>tenuis</i> (Woronich.) Elenk.	-	-	r
<i>Phormidium tenue</i> (Menegh.) Gom.	r	1	2
<i>Phormidium valderiae</i> (Delip.) Geitl. f. <i>maius</i> (Ag.) Gom. (Lemm.) Elenk.	1	-	2
<i>Phormidium valderiae</i> (Delip.) Geitl. f. <i>tenuis</i> (Woronich.) Elenk.	-	1	1
<i>Achnanthes coarctata</i> Bréb.	-	r	-
<i>Achnanthes lanceolata</i> (Bréb.) Grun.	1	1	1

## Continue

<i>Amphora ovalis</i> (Kütz.) Kütz.	r	r	r
<i>Caloneis silicula</i> (Ehr.) Cleve	-	r	r
<i>Cyclotella radiosa</i> (Grun.) Lemm.	-	r	-
<i>Cymbella affinis</i> Kütz.	1	1	1
<i>Diatoma vulgare</i> Bory	1	1	-
<i>Fragilaria brevistriata</i> Grun.	1	2	2
<i>Fragilaria dilatata</i> (Bréb.) Lange-Bertalot	-	r	r
<i>Fragilaria ulna</i> (Nitzsch.) Lange-Bertalot	r	r	r
<i>Gomphonema acuminatum</i> Ehr.	r	r	r
<i>Gomphonema clavatum</i> Ehr.	r	r	r
<i>Gomphonema angustum</i> Kütz.	-	r	r
<i>Hantzschia amphioxys</i> (Ehr.) Grun.	-	r	r
<i>Navicula cuspidata</i> (Kütz.) Kütz.	1	1	1
<i>Navicula nivalis</i> Ehr.	-	-	r
<i>Navicula pupula</i> Kütz.	r	r	1
<i>Navicula radiosa</i> Kütz.	1	1	1
<i>Navicula rhynchocephala</i> Kütz.	1	r	r
<i>Nitzschia palea</i> (Kütz.) W. Smith	-	-	1
<i>Pinnularia borealis</i> Ehr.	-	1	r
<i>Pinnularia braunii</i> (Grun.) Cleve	1	2	2
<i>Pinnularia viridis</i> (Nitzsch.) Ehr.	1	1	1
<i>Rhoicosphaenia abbreviata</i> (A. Ag.) Lange-Bertalot	r	1	1
<i>Surirella ovalis</i> Bréb.	-	r	r
<i>Cosmarium laeve</i> Rabh	r	r	-
<i>Protococcus viridis</i> Ag.	1	-	r
<i>Scenedesmus quadricauda</i> (Turp.) Bréb.	r	-	-

Tab. 3. – Number of taxons on different temperature in thermomineral water of concrete canal of Niška Banja spa

Divisions	23.2-30.0	30.0-35.0	35.0-38.2
	°C	°C	°C
<i>Cyanophyta</i>	14	15	22
<i>Bacillariophyta</i>	15	24	23
<i>Chlorophyta</i>	3	1	1
TOTAL	36	40	46



In consideration to its temperature in November 1992, water in concrete canal was not thermal from biological point of view (Vouk, 1936).

On the basis of qualitative and relatively quantitative composition of algae, the vegetation of this thermomineral water may be specified as **cyano-diatomaceous** and as *Oscillatoria-Phormidium* - type.

Spring Suva Banja. By the examination of the sampels from the Suva Banja spring, the presence of algae from 3 divisions was established: *Cyanophyta*, *Rhodophyta* and *Bacillariophyta* (Tab. 4 and Tab. 5).

Tab. 4. - Qualitative and relatively quantitative composition of algae in the-rrnomineral water of Suva Banja spring of Niša Banja spa

Taxons	surfaces in the cave	surfaces out of the cave
<i>Aphanocapsa thermalis</i> Brü g.	1	1
<i>Gloeocapsa minor</i> (K ü t z.) H o l l e r b.	1	r
<i>Gloeocapsa punctata</i> N ä g.	1	r
<i>Lyngbya martensiana</i> R a b h.	-	r
<i>Oscillatoria numidica</i> G o m.	-	r
<i>Oscillatoria tenuis</i> A g.	1	r
<i>Oscillatoria tenuis</i> var. <i>symplociformis</i> (H a n s g.) E l e n k.	2	1
<i>Phormidium foveolarum</i> (M o n t.) G o m.	1	1
<i>Phormidium ambiguum</i> G o m.	1	1
<i>Pleurocapsa minor</i> H a n s g. e m e n d G e i t l.	-	1
* <i>Chantransia chalybea</i> (L y n g b.) F r i e s	7	1
<i>Achnathes lanceolata</i> (B r é b.) G r u n.	-	r
<i>Amphora ovalis</i> (K ü t z.) K ü t z.	-	1
<i>Caloneis silicula</i> (E h r.) C l e v e	-	r
<i>Cymbella affinis</i> K ü t z.	1	1
<i>Fragilaria brevistriata</i> G r u n.	1	2
<i>Fragilaria ulna</i> (N i t z s c h.) L a n g e - B e r t a l o t	-	1
<i>Gomphonema clavatum</i> E h r.	-	1
<i>Navicula pupula</i> K ü t z.	-	1
<i>Navicula nivalis</i> E h r.	-	r
<i>Navicula radiosa</i> K ü t z.	r	r
<i>Navicula rhynchocephala</i> K ü t z.	-	r
<i>Pinnularia braunii</i> (G r u n.) C l e v e	1	2
<i>Rhoicosphaenia abbreviatum</i> (C. A g.) L a n g e - B e r t a l o t	-	1
<i>Surirella ovalis</i> B r é b.	-	1

\**Chantransia chalybea* (L y n g b.) F r i e s is not found in any samples collected in Novembar 1992.

Tab. 5. – Number of taxons on different surfaces in thermomineral water of spring Suva Banja of Niška Banja spa

Divisions	surfaces in the cave	surfaces out of the cave
<i>Cyanophyta</i>	7	10
<i>Bacillariophyta</i>	4	14
<i>Rhodophyta</i>	1	1
TOTAL	12	25

The qualitative diversity of algae was not great. The dominant species was *Chantransia chalybea*, specially at the beginning of the cave. Beside *Ch. chalybea* some greater abundance have *Oscillatoria tenuis* var. *symplociformis*, *Fragilaria brevisstriata* and *Pinnularia braunii*.

On the basis of qualitative and relative quantitative composition of algae, the vegetation of thermomineral water of Suva Banja spring may be specified as **rhodo-cyano-diatomaceous** and as *Chantransia* – type.

In November 1992 *Chantransia chalybea* was not found in any of collected samples. As the entrance into the cave was closed by metal plates, this alga was absent inside the cave. That fact means that in 1980, 1983 and 1985, *Ch. chalybea* out of the cave, was originated from the population of *Ch. chalybea* from the cave.

#### Some characteristics of determined taxons of algae from thermomineral water of Niška Banja spa

As we know from literature data, *Oscillatoria cortiana* (M e n e g h.) G o m. is noticed for the first time for territory of Serbia.

With exception of thermomineral waters of Niša Banja spa (C v i j a n, 1986), the next taxons are noticed for the first time in Serbia too: *Calothrix thermalis*, *Gloeocapsa crepidium*, *Gl. punctata*, *Oscillatoria tenuis* var. *symplociformis*, *O. numidica*, *Phormidium mole* f. *tenuis*, *Achnanthes coarctata*.

The next taxons are common inhabitants of therms: *Aphanocapsa thermalis*, *Calothrix thermalis*, *Gloeocapsa minor*, *Gl. punctata*, *Merismopedia punctata*, *Oscillatoria amphibia*, *O. numidica*, *O. okenii*, *O. terebriformis*, *Phormidium ambiguum*, *Ph. ambiguum* f. *maius*, *Ph. mole* f. *tenuis*, *Ph. tenue*, *Ph. valederiae*. But the most of them are represented by small number of individuals. The dominant algae are common inhabitants of water of different types or are living out of water too.

#### CONCLUSION

In the scope of algal studies of thermomineral water of Serbia the respective material from thermomineral water of Niška Banja spa was collected in September 1982, July 1983, April 1985 and November 1992.

Niška Banja spa is situated in central part of Serbia, 10 km south-east of the town Niš.

In Niška Banja there are two thermomineral springs: Main spring (with 35-120 l/sec. of water, temperature of 38.2-38.5°C) and spring Suva Banja (with 14-42 l/sec of water, temperature of 12-37°C). The both of them belong to the group of homeotherm water with moderate high (Main spring) or low (Suva Banja spring) radioactivity. The water of both springs is characterized by ions of  $\text{Ca}^{++}$  and  $\text{HCO}_3$  and pH near 7.

We investigated the thermomineral water of Suva Banja spring and thermomineral water flowing through the artificial, concrete canal originated from Main spring.

By the examination of collected material from concrete canal, the presence of algae from three divisions was established: *Cyanophyta* (31 taxons), *Bacillariophyta* (26) and *Chlorophyta* (3 taxons).

The dominant taxons are *Oscillatoria limosa* A g. and *O. limosa* f. *phormidioides* (R a b h .) E l e n k ., as well as *Phormidium foveolarum* (M o n t .) G o m ., *Fragilaria brevistriata* G r u n . and *Pinnularia braunii* (G r u n .) C l e v e .

The vegetation of thermomineral water of concrete canal in Niška Banja spa may be specified as **cyano-diatomaceous** and as *Oscillatoria-Phormidium* - type.

Only in November 1992 *O. limosa*, as a dominant species in the earlier periods, was substituted by *O. limosa*, (M e n e g h .) G o m . But samples from November 1992 was not typical for this thermomineral water from many reasons.

By the examination of collected material from thermomineral water of cave-spring Suva Banja, the presence of algae from three divisions was established: *Cyanophyta* (10 taxons), *Rhodophyta* (1) and *Bacillariophyta* (14 taxons).

The dominant species is *Chantransia chalybea* (L y n g b .) F r i e s ., specially at the beginning of the cave. Beside *Ch. chalybea* great abundance show *Oscillatoria tenuis* A g. var. *symplociformis* (H a n s g .) E l e n k ., *Fragilaria brevistriata* G r u n . and *Pinnularia braunii* (G r u n .) C l e v e . too.

The vegetation of thermomineral water of Suva Banja spring in Niša Banja spa is specified as **rhodo-cyano-diatomaceous** and as *Chantransia*-type.

Exception is November of 1992 when the entrance of the cave was closed by metal plates and algae disappeared from the cave because of low level of the light intensity.

As we know from literature data (C v i j a n and B l a ž e n č i ć, 1988), *Oscillatoria cortiana* (M e n e g h .) G o m . is noticed for the first time for territory of Serbia.

Some of presented taxons are common inhabitants of the thermns. But the most of them are represented by small number of individuals. Practically, algological base of thermomineral water of Niška Banja spa are presented by algae which are not characteristic for thermomineral water. The most of them are common inhabitants of the waters of different types or are living out of the water too.

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## Rezi me

MIRKO CVIJAN

## ALGE U TERMOMINERALNIM VODAMA NIŠKE BANJE (SRBLJA)

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U okviru istraživanja termomineralnih izvora Srbije u septembru 1982, julu 1983, aprilu 1985. i novembru 1992. sakupljen je algološki materijal iz termomineralne vode izvora Glavni izvor i izvora Suva Banja u Niškoj Banji.

Izvor Suva Banja karakteriše 14-42 l/sec. vode čija je temperatura 12-37°C, a Glavni izvor 35-120 l/sec. vode čija je temperatura 38.2-38.5°C. Oba izvora pripadaju grupi homeotermnih voda sa umereno visokom (Glavni izvor), odnosno niskom (Suva Banja) radioaktivnošću. Takođe ih odlikuje prisustvo jona  $\text{Ca}^{++}$  i  $\text{HCO}_3^-$ , kao i pH oko 7.

U termomineralnoj vodi Glavnog izvora utvrđeno je prisustvo algi iz tri razdela: *Cyanophyta* (31 taksona), *Bacillariophyta* (26) i *Chlorophyta* (3 taksona).

Utvrđena je kvantitativna dominacija *Oscillatoria limosa* A g. i *O. limnosa* f. *phormidioides* (R a b h.) E l e n k., kao i uvećana brojnost *Phormidium foveolarum* (M o n t.) G o m., *Fragilaria brevistriata* G r u n. i *Pinnularia braunii* (G r u n.) C l e v e. Na osnovu toga termomineralna voda Glavnog izvora označena je kao **cyano-dijatomejska**, a bliže kao *Oscillatoria-Phormidium*-tip.

Izuzetak je novembar 1992. kada je *Oscillatoria limosa* (dominantna u ranijim periodima istraživanja), zamenjena vrstom *O. cortiana* (M e n e g h.) G o m. Ali uzorci iz ovog perioda nisu reprezentativni iz više razloga.

U termomineralnoj vodi izvora Suva Banja utvrđeno je prisustvo algi iz tri razdela: *Cyanophyta* (10 taksona), *Rhodophyta* (1) i *Bacillariophyta* (14 taksona).

Utvrđena je kvantitativna dominacija *Chantransia chalybea* (L y n g b.) F r i e s, posebno na ulazu u pećinu. Osim toga visokom brojnošću odlikovale su se i *Oscillatoria tenuis* A g. var. *symplociformis* (H a n s g.) E l e n k., *Fragilaria brevistriata* G r u n. i *Pinnularia braunii* (G r u n.) C l e v e. Na osnovu toga termomineralna voda izvora Suva Banja označena je kao **rodo-cijano-dijatomejska**, a bliže kao *Chantransia*-tip.

Izuzetak predstavlja novembar 1992. kada je ulaz u pećinu bio zatvoren metalnim pločama te su alge u unutrašnjosti pećine, usled prevelike zaseve, bile odsutne.

Na osnovu dostupnih literaturnih podataka, *Oscillatoria cortiana* (M e n e g h.) G o m. po prvi put je zabeležena za teritoriju Srbije.

Neke od determinisanih algi široko su rasprostranjene u termomineralnim vodama. Međutim većina takvih predstavnika u termomineralnoj vodi istažnih izvora u Niškoj Banji zastupljena je malim ili veoma malim brojem jedinki. Praktično, algološku osnovu termomineralne vode oba istražena izvora čine alge koje za takve vode nisu karakteristične, koje su uobičajeni stanovnici voda različitog tipa ili, pak, žive i van vode.

UDC 951.6 : 588.145.1 (497.11)

MILORAD M. JANKOVIĆ

**PRILOG POZNAVANJU TERATOLOŠKIH POJAVA KOD VRSTE  
*DIGITALIS FERRUGINEA* L.  
(prethodno saopštenje)**

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Janković, M.M. (1992-1993): *Contribution to the knowledge of teratologic phenomena of Digitalis ferruginea*. – Glasnik Instituta za botaniku i botaničke bašte Univerziteta u Beogradu, Tom XVI-XVII, 97 - 111.

Teratologic malformations on stems and flowers of *Digitalis ferruginea*, appearing as tissue proliferation in apical flower regions are described in the paper.

Key words: *Digitalis ferruginea*, teratology, phylogeny, ecological factors, malformation, proliferation, plant morphology.

Ključne reči: *Digitalis ferruginea*, teratologija, filogenija, ekološki faktori, deformacije, proliferacija, morfologija cveta.

Pod *teratološkim pojavama* kod biljaka podrazumeva se stvaranje različitih ne normalnih oblika na pojedinim njenim delovima, naročito kod cveta. U stvari, radi se o „nakaznosti” tih delova, ili uopšte sa promenama koje više ili manje odstupaju od normalnog oblika. Postoje različiti teratološki oblici pri čemu su najznačajnije proliferacije (proliferacije), mahrovost cveta, fascijacija, hloroze, različite deformacije, pretvaranje prašnika u plodne listiće, pretvaranje gineceuma u prašnike ili različite nenormalne listove, srastanje listića krunice, povećanje ili smanjivanje njihovog broja, fascijacija, pretvaranje prašnika i delova plodnika, i samog plodnika u veoma čudne i

neobične listolike forme, i dr. Kod proliferacije iz cveta na vrhu grančice, ali i na bočnim stabljikama i njihovim pršljenovima, iz gineceuma (plodnika), izbija, umesto normalnih cvetnih delova, odnosno same cvetne osnove, niz abnormalnih tvorevina, napred navedenih, dakle grančica umesto cveta ali sa njegovim već započetim tvorevinama. Kako rekosmo, pod teratologijom podrazumevamo nakaze i abnormalne pojave kod biljaka (mada se, uzgred budi rečeno, teratologija može odnositi na čitav živi svet – životinje i samog čoveka), ali i nauku koja te nakaznosti proučava. Međutim, u novije vreme mnogi naučnici smatraju da su teratološke pojave veoma korisne za evoluciju, posebno za filogenetski sektor, a manje za ontogenetski, koji je više značajan za ontogenetski aspekt (T u t j a k , 1969).

Uzorci teratoloških pojava mogu biti različiti, mada u tom pogledu nauka još uvek nije dala konačnu reč: a – preterana vlaga i trofičnost podloge (zemljišta); b – preterana sušnost, toplota podloge (velika kserotermija podloge); c – jakost i specifičnost Sunčevog zračenja; d – osobeni virusni uticaji. Pri tome, verovatno, svaki od ovih uzroka može, pretpostavlja se, možda da stavi u pogon nekontrolisani razvoj genetički izuzetno bogato gineceumsko, meristematičko (ali i andreceumsko) i tkivo čitavog cveta. To dovodi do različitih oblika nakaznosti, pri čemu sama proliferacija omogućuje da se cvetni delovi pretvaraju u raznolike oblike, čitavi cvetovi ili pojedini njihovi delovi. Tako na primer, najčešće, plodnik se pretvara u prašnike, ili, rede, prašnici u plodnik, ili i jedni i drugi u raznovrsne neobične listove; tu, često, dolazi i do raznovrsnog srasanja čašice ili krunice, smanjivanje njihovog broja (ili pak povećanje), ili kod srasle krunice sve do smanjivanja njihovog broja u pršljenu proliferovane stabljike samo na jedan jedini. Ali, može se desiti i da se listovi na stabljici ili u čašici i krunici povećavaju brojem. Konkretno kod vrste *Digitalis ferruginea* čak do 7 listića, umesto 5, što je normalan broj. I kod delova plodnika (na primer kod semenih zametaka), pretvaraju se oni u najrazličitije listolike oblike, i to često u velikom broju. Svi delovi cveta pretvaraju se u najrazličitije oblike u odnosu na normalan cvet, često potpuno neprepoznatljivih formi. Prašnici su najčešće održani, ali takođe više ili manje izmenjeni, često samo u jednostavan stubić ili čak samo u malu kvržicu.

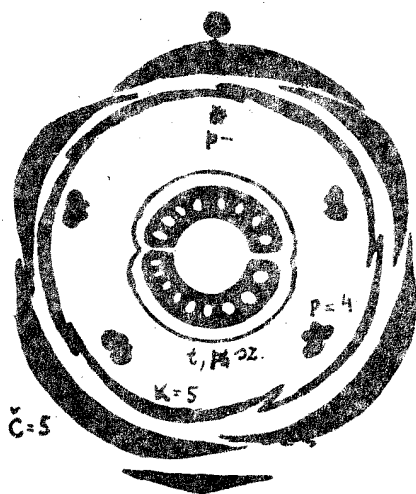


Fig. 1. – Cvetni dijagram vrste *Digitalis purpurea*: č – čašični listići, k – krunični listići, p – prašnici, t – tučak, pl – plodnik, sz – semeni zametak

Floral diagram of *Digitalis purpurea*: č – sepals, k – petals, p – stamens, t – pistil, pl – ovary, sz – ovule

Najznačajnija makropromena jeste sama proliferacija jednog cveta na vrhu vegetativne grančice, pri čemu ta proliferacija grančica-cvet može biti dugačka čak i do 30 cm. U drugom slučaju proliferuju se cvetovi na više bliskih vršnih grančica (izbilih iz glavne stabljike), i tada se sve te proliferovane grančice približavaju jedna drugoj i dodiruju međusobno, stvarajući tako jedan rastresit snop, te čine neku vrstu proliferacijske cvasti (Fig. 2). Najzad, kako je već rečeno, svi delovi teratološki izmenjenih cvetova mogu biti patološki preobraženi: listići, krunični i čašični, sami prašnici, tučak, plodnik i njegovi delovi, delovi plodnika (plodni listići, semeni zameci, placenta, i dr.).

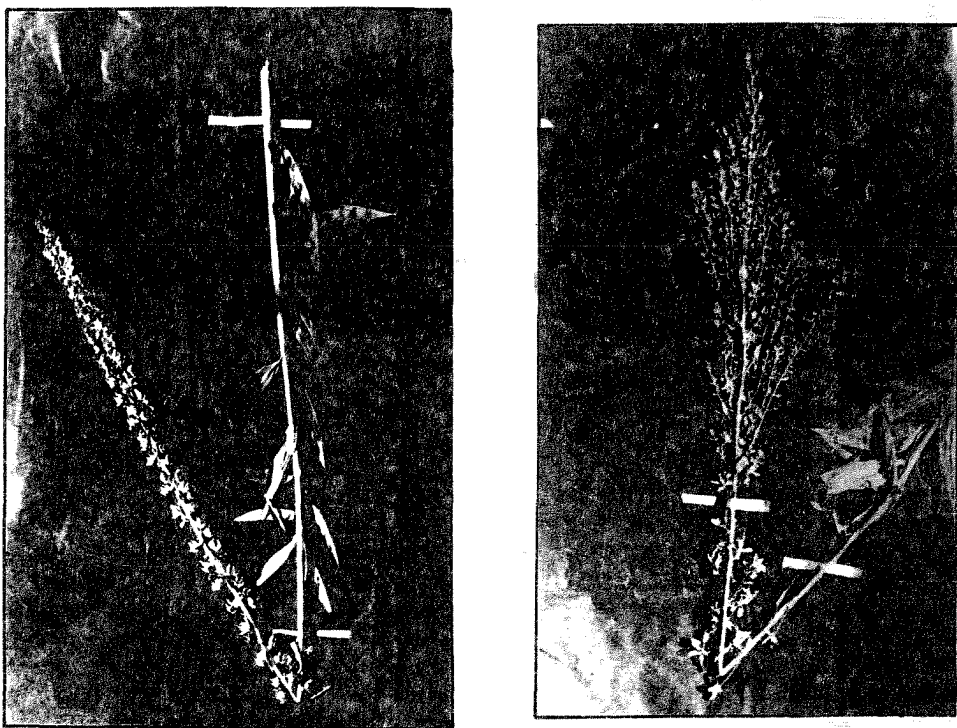


Fig. 2. – Prolifkovane grančice (iz cvetova) vrste *Digitalis ferruginea*, levo sa jednom grančicom, desno sa više grančica u formi „proliferacijske cvasti”. (Foto M.M. Janković)

Proliferous branchlets from the flowers of *Digitalis ferruginea*, left – with one branchlet, right – with few branchlets in the form of proliferous inflorescence. (Photo M.M. Janković)

Naravno, svi ovi delovi teratološki izmenjenih sektora cveta vrste *Digitalis ferruginea* (proliferovane stabljike, delovi cveta i sam cvet u celini), zelene su boje te, posebno kada je reč o cvetovima, radi se u ovom slučaju o tzv. teratološkoj manifestaciji ozeljenjavanja.

Autor ovog rada nije se posebno bavio teratološkim pojavama kod biljaka, kao ni kod samog digitalisa. Ali, s obzirom na izuzetnu zanimljivost ovih teratološki izmenjenih primeraka digitalisa koje sam našao u jednoj populaciji na Fruškoj gori, smatram da ovaj prikaz može da bude inspiracija za dalji rad (uključujući čak i

laboratorijska istraživanja), i to ne samo na rodu *Digitalis* (koji je ovoj manifestaciji teratološkog karaktera naklonjen), već i na mnogim drugim.

Takođe, na teratološke pojave treba ukazati i u vezi sa evolucijom biljaka. Naime, danas, prema mnogim shvatanjima, kao uzroke evolucionih kretanja (ka stvaranju novih vrsta, ali i podvrsta i varijeteta), navode se sledeći faktori:

1. **Mutacije.**

2. **Rekombinacije** (kombinacije, ukrštanje).

3. **Virusno prenošenje naslednih činilaca** (DNK, celih ili samo delimičnih), u ćelije određenih vrsta, i

4. **Teratologija**, kao faktor stvaranja izmene starih i stvaranja novih karaktera, i od kojih će se neki nasledivati i prirodnim odabiranjem učvršćivati u novu, ovim izmenjenu DNK; time se formiraju i novi oblici u okviru stare vrste, čije određene jedinice (populacije) postaju jedinice i populacije nove vrste.

25.IX 1954. godine našao sam na Fruškoj gori nekoliko prolifikovanih biljaka vrste *Digitalis ferruginea* L. Zbog čega o ovim interesantnim nalazima nisam ranije pisao stvar je određenih okolnosti, ali mislim da je bolje „ikad nego nikad“.

Kako do sada nije, koliko je meni poznato, u literaturi zabeležena prolifkacija kod ove vrste, a i za čitav rod *Digitalis* o ovoj pojavi ima malo podataka, smatram da će biti korisno detaljnije iznošenje činjenica u vezi sa prolifikacijom *Digitalis ferruginea* na Fruškoj gori. Istina, Džonson (J o n s o n , 1936) već je ranije opisao teratološke pojave kod cvetova *Digitalis purpurea* L. var. *gloxnaeflora* Vilm., ali se u njegovom slučaju ne radi uvek o tipičnoj prolifikaciji, jer je samo određen broj cvetova prorastao i ozeleneo, dok je čitav niz drugih cvetova imao abnormalno razvijene delove (čashični i krunični listići, andiceum i gineceum, placenta), na najrazličitije načine. Džonson podvlači da ni dva cveta nisu bila istovetna u pogledu manifestovane abnormalnosti. Pored toga u ovom slučaju radi se o (bonitetnom) varijetetu koji se abnormalno razvio u uslovima gajenja u Botaničkoj bašti Kalifornijskog univerziteta u Los Andjelosu. Prema tome, slučaj prolifikacije kod *Digitalis ferruginea* sa Fruške gore interesantan je ne samo zbog toga što se javlja u oblasti prirodnih uslova i na vrsti za koju do sada nije bilo nikakvih podataka (u pogledu teratologije), već i zato što se prolifkacija *Digitalis ferruginea* javila u potpuno prirodnim uslovima i na biljkama koje nisu bile gajene. Osim toga prolifkacija i sveukupna teratologija *Digitalis ferruginea* može se smatrati u potpunosti potpuno tipičnim. Nije bez značaja da je naš slučaj vezan za područje Balkanskog poluostrva, a Džonsonov za sasvim drugi kraj sveta, za Kaliforniju. Najzad, radi se i o dve potpuno različite vrste, mada istog roda: *Digitalis ferruginea* i *Digitalis purpurea*.

Kako je već rečeno prolifkacija *Digitalis ferruginea* konstatovana je 25.IX 1954. godine, i to kod jedne manje grupe (populacije) biljaka od oko 10 do 20 individua, ove vrste. Ova populacija nalazila se na Irškom Vencu, na visini od oko 1.000 m n.v., odmah ispod vrha, na južnoj strani Fruške gore. Stanište ovih prolifikovanih biljaka odlikovalo se znatnom vlažnošću zemljišta i velikom zasenom, s obzirom da su biljke *Digitalis*-a izrasle duž jedne šumske staze koja je bila gotovo potpuno zasvođena i obrasla visokim žbunovima, koje je stvaralo gustu zasenu u donjim slojevima šume. Visina biljaka digitalisa iznosila je prosečno 150 cm. Od preko 10-20 biljaka samo je jedna imala



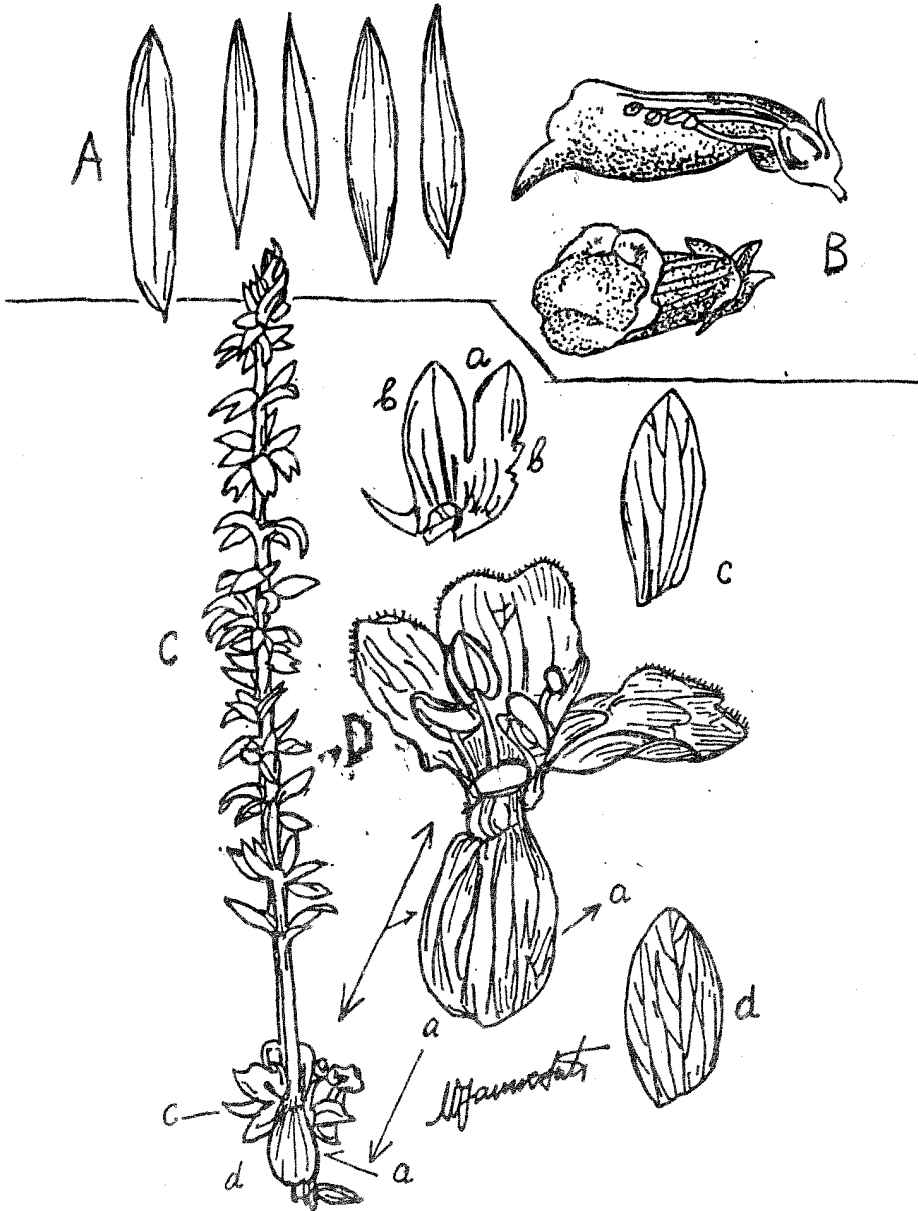


Fig. 3. - A - normalni listovi, B - normalni cvetovi *Digitalis purpurea*, C - proliferovana grančica *Digitalis ferruginea*, D - proliferovan cvet u odnosu na normalan, a - delić degradovanog cveta, c, d - skraćeni listići cveta

A - normal leaves, B - normal flowers of *Digitalis purpurea*, C - proliferous branchlet of *Digitalis ferruginea*, D - proliferous flower compared to normal one, a - a part of degrading flower, c, d - shortened petals

normalno razvijene stabljike, cvetove i plodove, dok su kod ostalih svi cvetovi bili proliferativni i patološki izmenjeni.

Pod proliferacijom podrazumevamo, bliže rečeno, aksialno (osovinsko) izrastanje cveta ili cvasti (i njihovo ozeljenjavanje). Konovalov (1948) o tome kaže sledeće: „U prirodnim uslovima, kod proliferacije, pod uticajem nekih, još neproučenih faktora, razvoj izdanka ne završava se kao obično, obrazovanjem normalnog razvijenog cveta. Razvije poslednjeg na ovom ili onom stupnju je specifično: iz njega ponovo niče listonosni izdanak, koji se sa svoje strane završava cvetom ili cvasti. Proliferacija se ostvaruje kroz morfološke promene cvetnih elemenata”.

Uzroci koji dovode do pojave proliferacije raznovrsni su. Oni se, izgleda, mogu svesti na sledeće osnovne grupe (nešto je o tome već rečeno):

1. Obilje ili nedostatak hrane (hranljivih materija u podlozi).
2. Preterana vlaga u podlozi.
3. Preterana sušnost i toplota podloge; velika kserotermija podloge.
4. Jakost i specifičnost Sunčevog zračenja.
5. Mehaničko delovanje na biljke.
6. Delovanje fitopatogenih virusa, ali i drugih.
7. Virusno delovanje kao prenosioca genetičkih informacija.

Prema svemu sudeći, iz proučavanja teratoloških pojava i proliferacije cvetova *Digitalis ferruginea*, mogu se detaljnije izraziti sledeći zaključci i opisati sledeće pojave.

Pre svega, sve počiva na proliferaciji cveta, iz čije osnove (placente i tučka – plodnika), izbija umesto normalnih delova normalnog cveta, ozelnela stabljičica koja može biti dugačka i do 30 cm (ponekad i više). To se dešava iz osnovnog cveta na vrhu vegetativne stabljike, ali i iz cvetova na samoj proliferativnoj stabljici (naravno, na njoj su svi cvetovi manje ili više nenormalni), koja u sebi već nosi određene teratogene (nenormalne i degenerativne) vegetativne ili genetičke osobine.

U nekim slučajevima proliferativne grančice su usamljene, dok su u drugim, naravno na jednoj istoj biljci, one mnogobrojne te približno jedna drugoj kao da čine „cvast” proliferativnih grančica.

Bez obzira na veličinu teratoloških promena cveta *Digitalisa*, veću ili manju, oni nikada ne liče, gotovo ni malo, na tipičan normalni cvet ove biljke; najčešće, oni su u tolikoj meri različiti od normalnih, da ih sa njima nikako ne možemo povezati; neki su pak prema monstruoznih tvorevina izuzetno izmenjeni i često u velikoj meri više ili manje degradovani i svedeni samo na jedan cvetni deo (npr. tučak ili prašnik uz samo jedan list), monstruoznog oblika.

Kada je reč o samim listovima, onda se može kazati da su oni na proliferativnoj stabljici u odnosu na normalne (sl. 3), uglavnom skraćeni, više ili manje deformisani (sl. 4), često i srasli među sobom; u ovom poslednjem slučaju često su u pitanju mogući čašični ili krunični listići.

U nekim slučajevima deformisani proliferativni cvet (uglavnom sa više/manje skraćenom krunicom), ima iz osnove tučka izraslu (proliferativnu) grančicu, koja na svom vrhu takođe prelazi u novi, takođe proliferativan abnormalan cvet, defektan i degradovan (sl. 5 A, B); ali su neki sasvim patološki deformisani, bez ikakve sličnosti

sa normalnim C, D (sl. 5, sl. 6 E), a u nekim slučajevima cvet je samo krajnje deformisan i uprošćen tučak, inseriran u neobične okruglaste stabljike sa čudnim šupljinama, pršljenasto ili bočno izrasle jedna iz druge (sl. 6 D). Na sl. 9 prikazano je nekoliko krajnje degradovanih cvetova (B, C), sa jednim slučajem (A) cveta svedenog samo na unutrašnju plodnu osovinu gusto obrasloj mnogobrojnim semenim zametcima, neobičnih oblika (1-17); tu su i neobični oblici na već sponenutim krajnje redukovanim cvetovima (B, C), nastalih verovanto na osnovu različitih devijacija semenih zametaka, tučkova, prašnika i kruničnih listića (18-28).

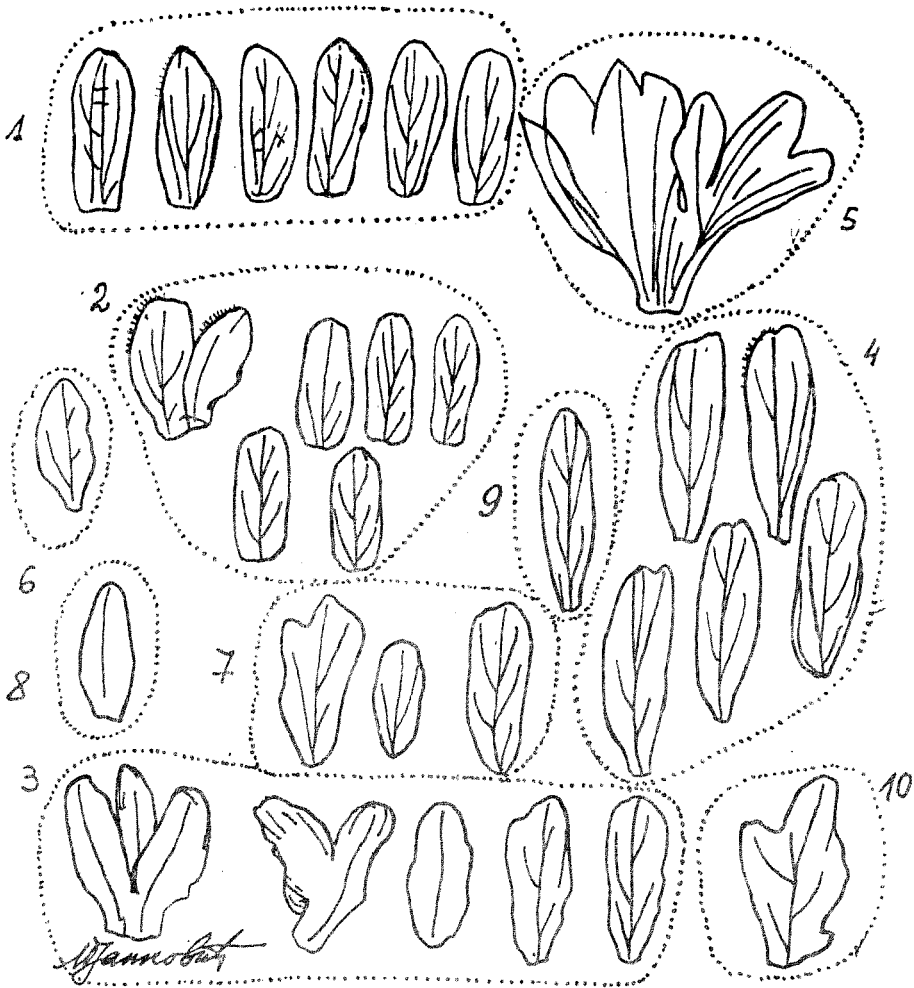


Fig. 4. – Oblik listova na jednoj prolifikujućoj stabljiki, iznad dva plodnikova listića, po redosledu pršljenova duž nje – od 1. do 10. pršljena  
Shape of petals along proliferous branchlets from whorls 1 to 10

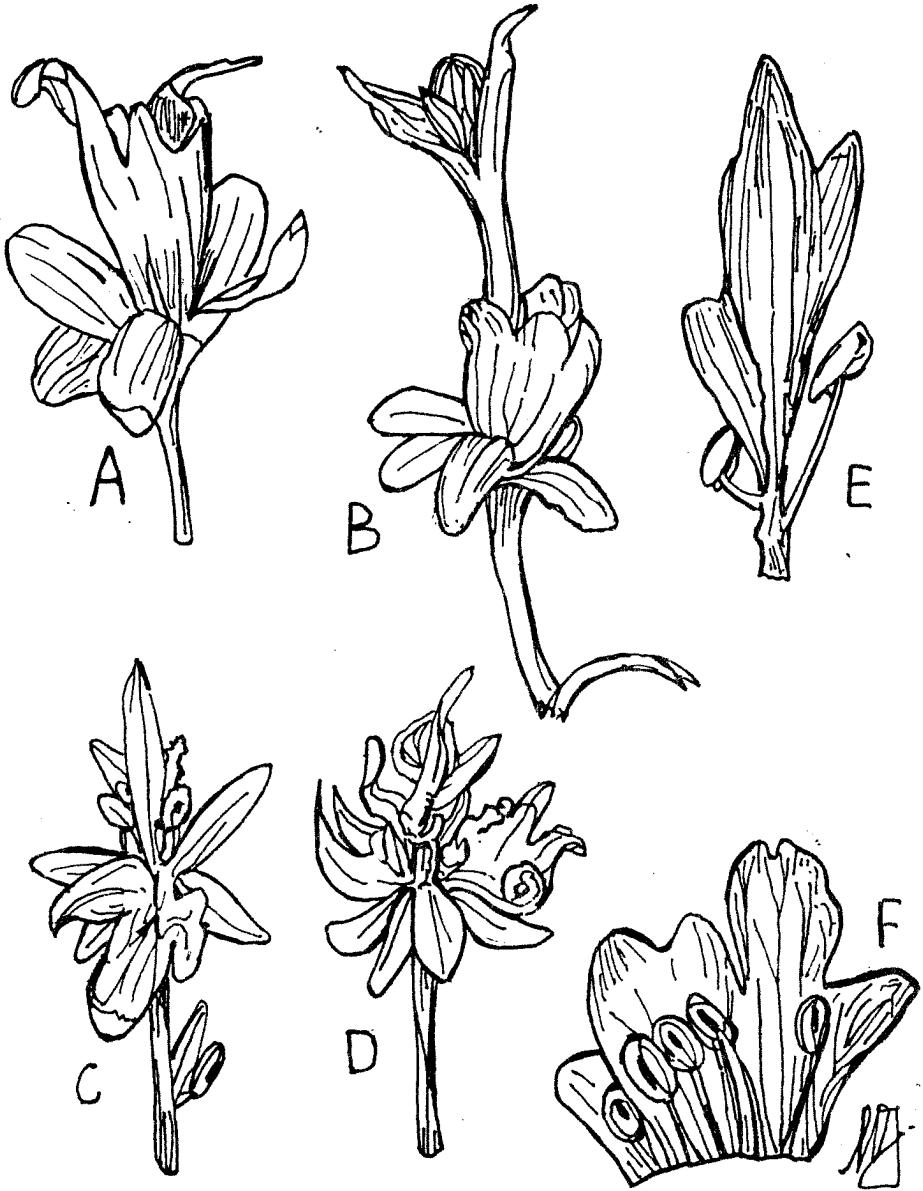


Fig. 5. - A - Jedan cvet pri dnu proliferovane ose, B - proliferovani cvet iznad njega, u sredini stabljice, C, D - proliferovani i jako izmenjeni cvetovi, E - jako degradovan cvet, samo sa sraslim listićem i samo sa dva prašnika, F - otvorena i jako deformisana krunica sa jednim prekobrojnim prašnikom.

A - flower at the bottom of proliferous axis, B - proliferous flower in the middle of the axis, C, D - proliferous and transformed flowers, E - very transformed and degrading sympetalous flower with only two stamens, F - open and very deformed petals with one overplus stamen.

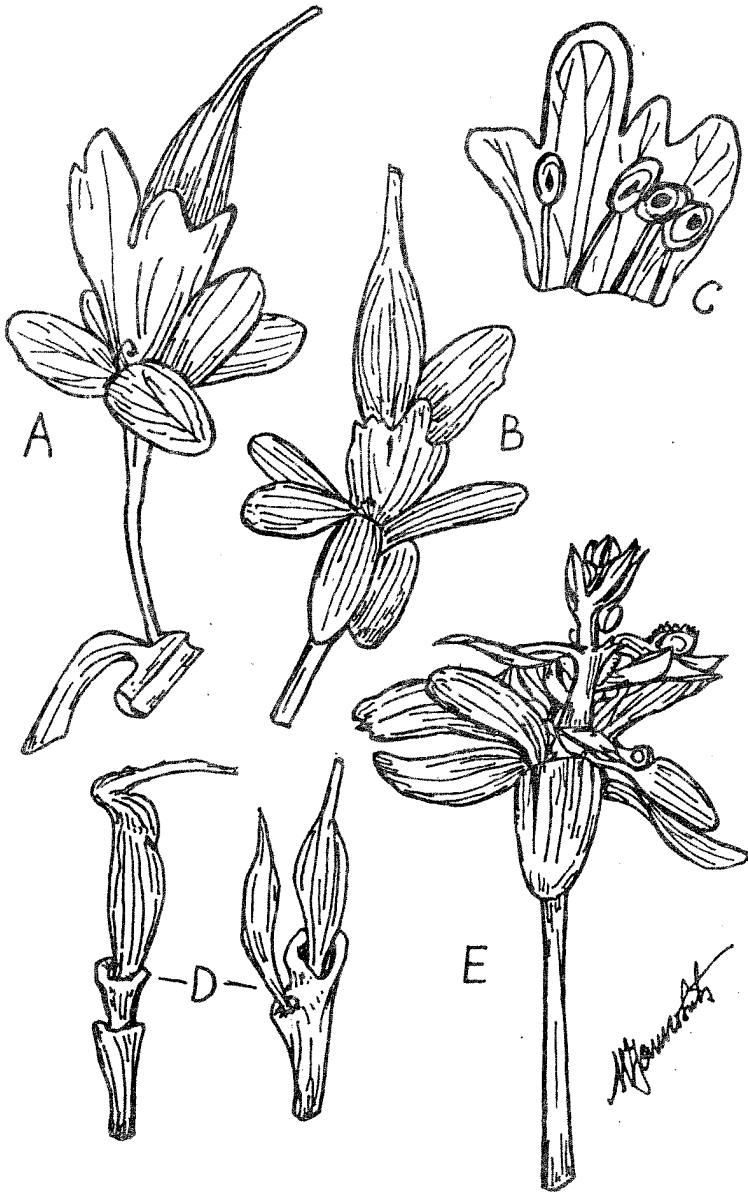


Fig. 6. - A & B - Prolifkovani cvetovi sa jako povećanim plodnikom i skraćenom krunicom, C - deformisana i skraćena krunica, D - neobično deformisani ginecejalni delovi potpuno deformisanog cveta, E - jako deformisan i složen prolifkovani cvet sa haotičnim rasporedom listova i plodnika i naznačenim prašnicima kao i sa tek izbilim prolifkovanim stubićem iz gornjeg dela već prolifkovanog cveta.  
A & B - Proliferous flowers with very enlarged ovary and shortened petals, C - deformed and shortened petals, D - extremely deformed gynoecium of completely deformed flower, E - very deformed and complex proliferous flower with chaotic arrangement of petals, ovary and stamens

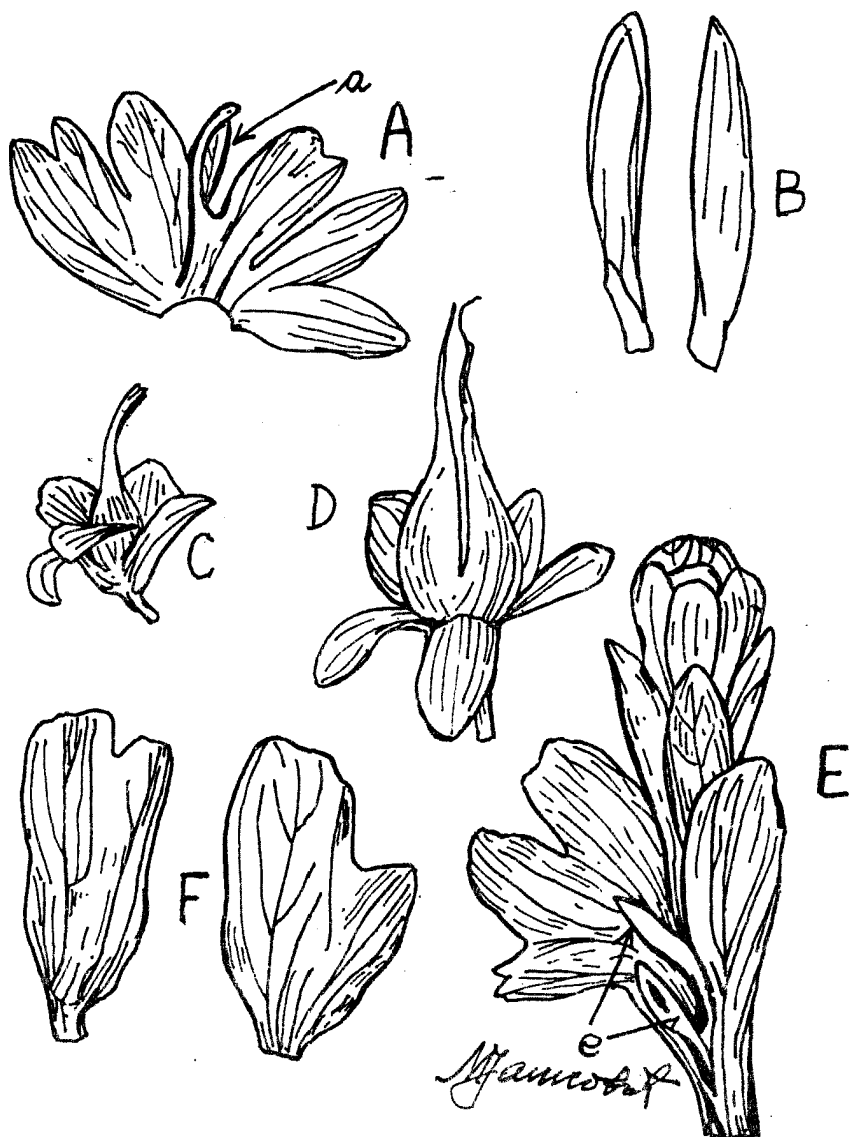


Fig. 7. - A - Na jednoj prolifikujućoj stabljici svi srasli listovi krunice sa jednim deformisanim prašnikom (a), C & D - prolifikujući cvetovi sa predimenzioniranim i više manje izmenjenim gineceumom, kao i slobodnim listićima nesrasle krunice, E - prolifikujući vrh prolifikujuće stabljike sa nenormalnim prašnicima (e), F - karakteristični listići ovog vrha

A - Proliferous axis with one deformed stamen (a) and all sympetalous flowers, C & D - proliferous flowers with oversized more or less transformed gynoecium and free petals, E - proliferous apical part of proliferous axis with abnormal stamens (e), F - characteristic petals of this apex

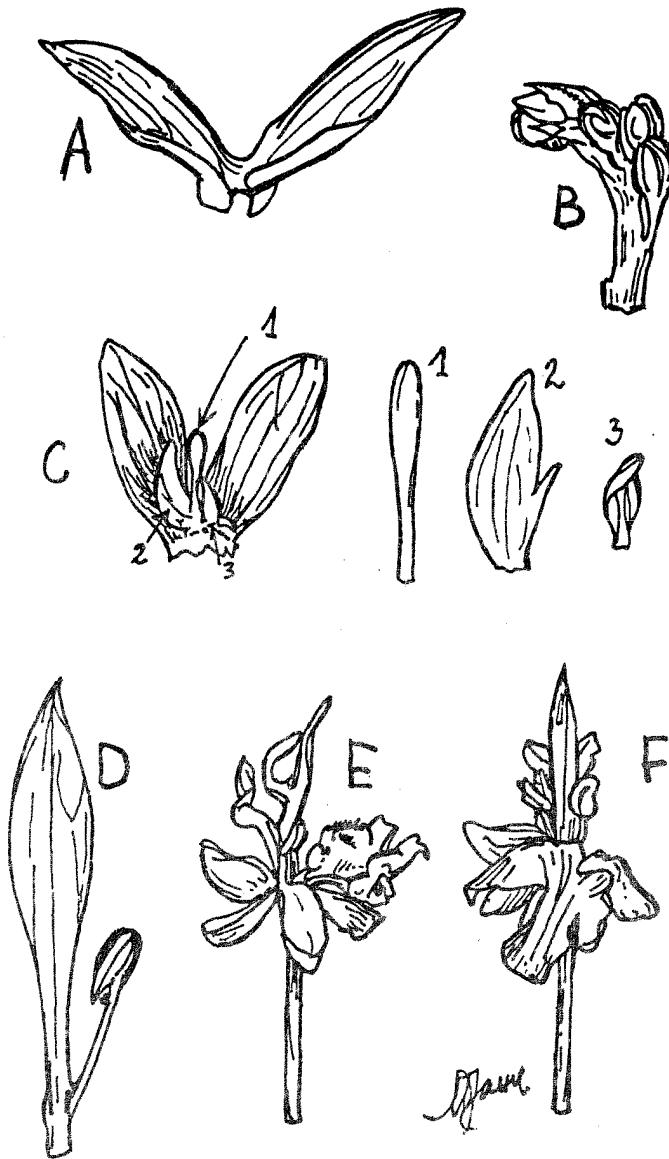


Fig. 8. - A - Čudni srasli listići sa jednog pršljena proliferovane stabljice, B - deformisani vrh jedne proliferacije stabljice, C - deo jednog vršnog proliferovanog lista (1 - veliki netipičan prašnik, 2 - listić plodnika, 3 - prašnik mali), D - jedan list sa prašnikom, E & F - jako deformisani i potpuno izmenjeni proliferovani cvetovi pri dnu proliferovane stabljike

A - Uncommon sympetalous floral parts of proliferous axis, B - deformed apical part of proliferous axis, C - a part of apical proliferous flower (1 - big atypical stamen, 2 - carpel, 3 - small stamen), D - petal with stamen, E & F - very deformed and completely transformed flowers at the bottom of axis.

Posebno treba istaći da neki oblici predstavljaju neobične hibride između kruničnih listića i prašnika, odnosno prašnika i delova tučka (sl. 9 C, B, 23, 25, 26).

Asimetrična segmentacija krunice vrlo je karakteristična i često na nekim umećeno proliferovanim cvetovima, pri čemu su segmenti varirali u pogledu oblika, broja i veličine; pri tome i čašični listići su bili različitog oblika, a javljali su se i nejasno definisani lobusi.

Prašnici, ukoliko ih ima prepoznatljivih, često su redukovani u pogledu broja, (ponekad ih ima i više, npr. 5 do 6), i oni su srasli ali sa kruničnim listićima; retko su sa potpuno razvijenim anterama, ali ih ima i takvih.

Gineceum, sa svojim promenama, nesumnjivo je najznačajniji deo cveta u pojavi proliferacije i različitim često monstruoznim njegovim promenama (deformitetima cvetnih delova, itd.). On je, više/manje produžen, uvek u različitom stepenu izmenjenim ili nepotpuno formiranim stubićem, tamo gde postoji samo jedan stubić; ali i ako je i više stubića, promene su slične. Neki stubići se pomaljavu (razrastaju) i produžavaju se iz placente, a drugi iz ovarijuma; i u jednom i u drugom slučaju reč je o istom gineceumu. U nekim slučajevima gineceum je promenjen u posebno carpeolid lišće, i to u nekoliko sukcesivnih pršljenova (što je upravo specifična potencijalna proliferacija).

Placenta je debela i mesnata; najčešće je bezoblična, a često je i V – strukture; ona je ili izdvojena, ili pak spojena u čvrst centralni stubić, koji je na vrhu razgranat u stubičaste grančice.

Proliferacija gineceuma manifestuje se često u listolike dugačke drške; ne tako retko nosi terminalno karpeloidno lišće, koje je spiralno produženo u dugačke visoke vrhove.

Kako je već rečeno, krunični listići su ili odsutni, ili pak odvojeni ili pak srasli u kratku cevastu tvorevinu. Čašica takođe može biti odsutna, ali ponekad sa jednim ili više petaloidnih segmenata. Karakteristična je i pojava pršljenastih listova sličnih čašici, i to između pravih čašičnih listića i krunice; sve ovo je ponekad toliko izmešano i deformisano, da je teško uočiti ovu pršljenastu segmentaciju cveta.

Jednom rečju, u proliferovanim biljkama vrste *Digitalis ferruginea* sa Fruške gore, toliko je mnogo tipičnih proliferovanih cvetova, sa velikim brojem različitih teratoloških oblika pojedinih delova cveta, toliko neobičnih i čudesnih formi, koja nas prosto zapanjuju, potpuno iščezlih cvetova koji ni po izgledu ni po građi, ni ne podsećaju na tipične normalne cvetove roda *Digitalis*, da, zaista, ovaj autorov materijal i nalaz moramo smatrati veoma zanimljivim i značajnim u vezi sa proučavanjem same teratologije (kao nauke), i određenih teratoloških pojava kao što je, na primer, proliferacija.

Na kraju se može reći da se autor nada da će ovaj prilog podstaći i inspirisati naše botaničare da se u većoj meri posvete pojavi kod nas biljne fitopatologije, uopšte, i proliferacije posebno.



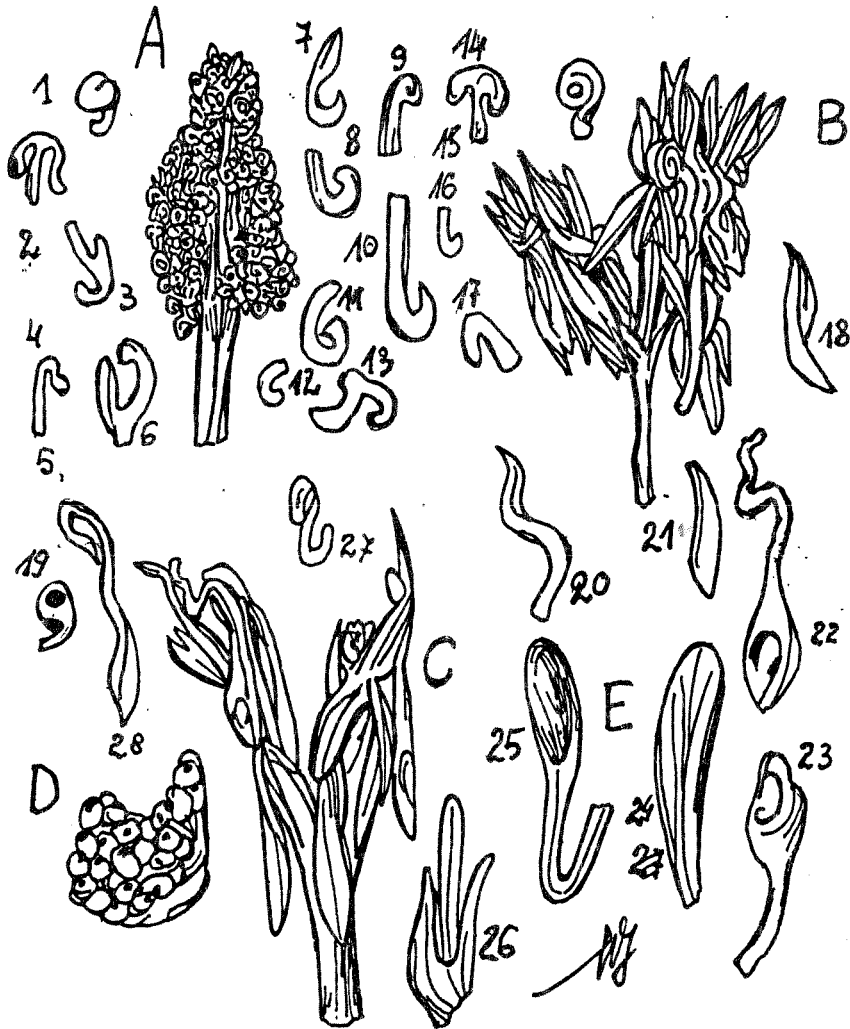


Fig. 9. – A – vrh cvetne ose (deformisani plodnik) sa gustom pokrivenim semenim zametcima koji su u velikom broju oko gineceuma prikazani pojedinačno, B & C – deformisani plodnici, plodnici sa haotično raspoređenim plodnim listićima, semenim zametcima, ponekim prašnikom (od 1 do 28), različitim neobičnim oblicima iz ovih vršnih ginecijalnih delova, a možda i andreceumijskih; ovo je sve iz unutrašnjosti nekih nenormalnih cvetova, u sredini proliferovane stabljike, možda i deformisani prašnici, D – deo placente sa semenim zametcima plodnika jednog cveta, više-manje normalnog na vrhu jedne više-manje normalne stabljice

A – flower apex (deformed ovary) densely covered with ovules which are shown individually, in great number, around gynoecium, B & C – deformed ovaries with chaotic arrangement of carpels, ovules, stamens (from 1 to 28), different shapes of terminal parts of gynoecium and probably of androecium, in the middle of proliferous axis, D – a part of placenta with ovules of more or less normal flower at apical part of more or less normal axis



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Radovi za Glasnik treba da budu napisani na srpskom ili engleskom jeziku uobičajene dužine do 15 kucanih stranica sa svim prilogima. Uredništvu treba poslati original i dve kopije rukopisa sa kompletnim prilogima ili rad na disketi u formatu Word for Windows 2.0, Word Perfect 5.1 ili ASCII.

Rukopisi se recenziraju i moraju se prilagoditi predloženim primedbama.

**Autori dobijaju 30 separata gratis.**

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**Latinska imena** biljaka treba pisati kompletno, sa imenom autora, pri prvom pominjanju u tekstu, a zatim se može skraćivati ime roda, izuzev kada se u radu pominju dva roda sa istim početnim slovom. Za biljne zajednice, takođe, treba pisati pun latinski naziv i ime autora. Latinska imena se podvlače ili kucaju *italikom*.

**Tabele** treba da budu urađene na posebnim listovima i označene, prema redosledu, arapskim brojevima i slovima (A, B, C, itd.) ukoliko su iz delova. Svaka tabela treba da ima naslov i legendu u zaglavlju i treba da bude referisana u tekstu. Autori treba da označe mesto tabele u tekstu. Tabele se citiraju kao Tab. 1, Tab. 1A, Tabs. 1-3. (Tab. 1), itd.

**Prilozi** mogu biti crno-bele fotografije, dijagrami, originalni crteži, mape, grafikoni. Na svakoj ilustraciji grafitnom olovkom označiti broj priloga, orijentaciju i ime autora. Autor treba da odredi mesto ilustracije u tekstu strelicom na levoj margini. Prilozi treba da budu u veličini koja odgovara konačnoj reprodukciji ili dvostruko većih dimenzija (maksimalna širina teksta je 12.5 cm, a dužina 18.5 cm). Prilozi se citiraju u tekstu kao Fig. 1, Fig. 1A, Figs. 1-3. (Fig. 1), itd.

**Dijagrami, crteži, mape i grafikoni** moraju biti izrađeni dovoljno debelim i tamnim linijama na belom papiru (ili paus-papiru), odnosno treba da budu štampani na laserskom štampaču ako su kompjuterski obrađeni.

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**Lista objašnjenja (legendi) za priloge** treba da bude data na posebnom listu hartije prema redosledu.

**Literatura u tekstu** citira se na jedan od sledećih načina: Košanin (1929), (Košanin, 1929), Černjavski & Soška (1937), itd. Ako je autor objavio više radova u istoj godini, oni se, prema redosledu, obeležavaju godinom i slovom, na pr. 1989a, 1989b, itd. U radovima troje autora pri prvom citiranju pominju se sva imena, a zatim, samo ime prvom autora i *et al.* Radovi više autora (četiri i više) uvek se citiraju imenom prvog autora i *et al.*

**Literatura u bibliografskom spisku** sređuje se po azbučnom redu imena autora na svim podacima o radu, izdanju i izdavaču, i to na sledeći način:

Pančić, J. (1874): Flora Kneževine Srbije. – Državna štamparija, Beograd.

Nikolić, V. (1973): *Paniccia L. in Flora SR Srbije V* (ed. M. Josifović). – SANU, Beograd.

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Košanić, N. (1929): Die Koniferen Sudserbiens. – Bull. Inst. Jard. Bot. Univ. Belgrade 1(2): 176-190.

U literaturi se mogu navesti i radovi koji su u štampi (*in press*), ali ne i usmena saopštenja (samo u tekstu). U bibliografskom spisku navode se imena svih autora jednog rada, s tim što se imena predposlednjeg i poslednjeg vezuju znakom &. Imena časopisa treba skraćivati prema internacionalnim standardima ili na sličan način za domaće časopise.

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## INSTRUCTIONS TO AUTHORS

The papers must be written in English or Serbian language and should not normally exceed 15 pages. The manuscript should be submitted to the Editor in an original, two copies and on a diskette in Word for Windows, 2.0, Word Perfect 5.1 and ASCII.

The manuscripts will be reviewed and modified according to referees suggestions.

**The authors receive 30 free reprints.**

The text must be typed double-spaced throughout, with wide margins. All pages must be numbered including all illustrations and list of legends. The authors should suggest a **running title** of not more than 5 words. The same results may not appear as both tables and figures.

**Title page** comprises: **Authors name(s), Title, affiliations, and Address of the corresponding author.** An **Abstract** in English with headline should be less than 150 words. Bilingual **Key words** should not exceed 10 words.

The main sub-divisions of the papers are: **Introduction, Material and Methods, Results, Discussion, Conclusion, Acknowledgments** (if any), **References, Summary** in English for the papers written in Serbian, or **Rezime** in Serbian for the papers written in English. The text in Serbian must have bilingual captions.

**Latin names** (genus, species) and authority must be cited when first mentioned. Further on, the generic name may be abbreviated to its initial except where reference to other genera with the same initial could cause confusion. Latin names should be underlined or typed in *italics*.

**Tables** should be numbered in arabic numerals and submitted on a separate sheet and accompanied by a title and appropriate legend at the top. Each table must be referred to in the text and the indication of preferred position in the text should be given. Citation in the text should be Tab. 1, Tab. 1A, Tabs. 1-3, (Tab. 1), etc.

**Illustrations** may be black and white photos, diagrams or drawings, maps, graphs, labelled with the figure number and author's name in soft pencil on the back identifying the top edge. The position in the text should be indicated by arrow on left margin. In general, the size of each figure must be planned for publishing without reduction or they may be twice the linear dimensions desired in the final reproduction: the maximum space on a page is 12,5x18,5 cm. The figures must be cited in the text as Fig. 1, Fig. 1A, Figs. 1-4, (Fig. 1), etc.

**Diagrams, drawings, maps or graphs** should be drawn boldly in black ink on stout white paper or computer-drawn of the highest quality to stand reduction to the desired size.

Black and white **photographs** must be printed on glossy paper of good contrast.

A separate typewritten double-spaced **list of legends** of all figures must be supplied.

**Literature citation in the text** should take the form: Košanin (1929). (Košanin, 1929). Černjavski & Soška (1937), etc. If several papers by the same author(s) in the same year are cited, they should be lettered in sequence (1989a), (1989b), etc. When papers are by three authors, use all names on the first mention then abbreviate to the first name and *et al.* For papers by four or more authors use *et al.* throughout.

**Literature in References** must be typed with double spacing, without serial numbering and placed in alphabetical order according to the authors' names. Full references must be given according to the type of publication cited, as follows:

Pančić, J. (1874): Flora Kneževine Srbije. – Državna štamparija, Beograd.

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In the References the names of all authors of one paper must be indicated and the last two linked by &. Other citations such as papers „in press” may appear in the References. A „personal communication” may be cited in the text, but not in the References.

All citations in the text should appear in the literature list and vice versa.

Abbreviated journal names are used according to the standards, or may be formed analogically.

**Abbreviations** for widely accepted terms may be used in the text, but for the new ones the full explanation should be given.



