



# Significance and need of powdery mildew control (*Microsphaera alphitoides* Griff. et Maubl.) in the process of regeneration of the pedunculate oak (*Quercus robur* L.) stands in the Ravni Srem area

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## Abstract

**Background and Purpose:** Powdery mildew (*Microsphaera alphitoides* Griff. et Maubl.) was studied from the aspect of its influence on regeneration of the pedunculate oak stands in the Ravni Srem area. Possibility to control the fungus with chemical protection measures was also investigated. During the three-year research period (2006–2008), efficiency of eight fungicides (*Microthiol special*, *Stroby DE*, *ZATO 50-WG*, *Cabrio Top*, *Quadris*, *Alert S*, *Duett Ultra*, *Sabithane*) and their influence on the plant growth increase were tested.

**Material and Methods:** The research was conducted on seedlings and juvenile oak plants in a newly regenerated pedunculate oak stand in the southwestern Srem area, in the Vinična forest section (dept. 15a). Level of plants infection and evaluation of fungicide efficiency were determined by visual evaluation of the presence of powdery mildew on leaves of 25 randomly picked plants within each experimental field. Plant growth elements were determined on the basis of the length of multistage formed shoots and root neck diameter.

**Results and Conclusion:** Suppression of powdery mildew on pedunculate oak seedlings and juvenile oak plants is an important protective measure which is essential for regeneration areas in the period of regeneration cutting. The observed fungicides showed high efficiency in control of the fungus. A biological feature of pedunculate oak to form multistaged shoots by activating terminal buds during the growing season came to the fore. Majority of plants formed 3 or 4 stage growths during the first year. Chemical protection of seedlings from powdery mildew should be conducted in the period of plant growth in height. In such proceedings, the application of fungicides is reduced to minimum and thereby provides a rational seedling protection. In the second and third year of oak development, applied fungicides had little effect. Under a canopy of herbaceous weed species, plants had more or less intense growth in height, depending on climatic conditions and the amount of moisture in the soil.

## INTRODUCTION

Powdery mildew (*Microsphaera alphitoides* Griff. et Maubl., sin. *M. quercina* (Schw.) Burr.) is a significant pedunculate oak fungal disease. Infection with it appeared as an epidemic disease in France in

1907 from where rapidly spread in other parts of Europe. There are two opinions on powdery mildew origin in Europe. According to the older view, the fungus was brought from North America (1) while according to (2) powdery mildew had been present in Europe before only in weakly virulent form so it was not noticed until a significant damage was made. It is assumed that the fungus mutated in to more virulent pathotypes. In pedunculate oak forests in Posavina, this fungus has been chronically present for more than a century (3, 4, 5). The harmful significance of this fungus in forest regeneration and cultivation was pointed out by many authors (6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18). Above mentioned authors state that powdery mildew is one of the most important factors of juvenile oak plants survival and as well as oak forest regeneration. It is believed that the fungus does not cause significant damage to cultivated plants in older stands. It is considered that only in cases of total browse, powdery mildew plays an important role as a causal factor that reduces the vitality of the oak forests.

Powdery mildew attacks oak seedlings and juvenile oak plants in a wide range of environmental and structural conditions in the stands. In the Ravni Srem area, powdery mildew was first monitored and registered with special attention during the early 1990's. For the purpose of successful natural regeneration of pedunculate oak stands, favorable conditions for preservation and development of seedlings and juvenile oak plants in regeneration areas were defined in that period (13). From the year of 1993 until today, as part of the modern system of pedunculate oak stand regeneration in the Ravni Srem area, preventive chemical protection has been provided against the powdery mildew (19).

A long tradition of pedunculate oak cultivation in Ravni Srem and well known quality of its wood make this area the economically most important oak forest area in Serbia. Considering the high value of the pedunculate oak in Ravni Srem area and its vulnerability to powdery mildew in the initial stage of growth, it has become necessary to study this important factor. By dealing only with individual factors and determining a segment of biological principles, in this research we endeavored to point out the importance of powdery mildew in the process of oak stand regeneration as well as the time, the need and justification for chemical protection of juvenile oak plants.

According to (15, 20) in last years there was a lack of phenological observations related to the stage of plant development and forming of new shoots. Beside that, the lack of information about age limit of juvenile oak plants until powdery mildew suppression is justifiable were the reasons of numerous applied protection measures unnecessary from both biological and economical point of view.

The protection of the seedlings and juvenile oak plants until 3 years of age in the Ravni Srem area is performed as needed by using different fungicides (Rubigan, Anvil, Karathane, Stroby DF, sulfur). Forest professionals do not lose interest in achieving the best possible results in

protecting juvenile oak plants from mildew. They are seeking the possibility to use newer preparations in order to achieve the most effective protection.

During the study (2006–2008) which was conducted in the area of the Forest Estate Sremska Mitrovica, in the forest office Morović, the effectiveness of certain fungicides was examined and the importance of oak powdery mildew reviewed as a risk factor in the pedunculate oak stand restoration. In that context, according to current knowledge (15, 20) a multistage growth of the pedunculate oak in regeneration sites and the aspects significant for seedling and juvenile oak plant protection from the mildew were examined. Along with the development of oak seedlings and juvenile oak plants, we analyzed the basic characteristics of the regeneration area. Based on the conducted research, the age limit was defined until which it is necessary to conduct the chemical protection of juvenile oak plants in newly regenerated pedunculate oak stands.

## MATERIAL AND METHODS

The research was conducted in southwestern Srem, in the Vinična forest section (dept. 15a) in the mixed stand of pedunculate oak, narrow-leaved ash and hornbeam which is *Carpino-Fraxino-Quercetum roboris caricetosum remotae* forest type on meadow black soil in the area that is not flooded by the Sava River (21). The stand regeneration was conducted by the procedure described by (15, 22, 23). The regeneration process of the above mentioned stand started in 2004 and in the autumn of 2005 within the existing system of geometrically isolated working fields, the size of 0.9 ha (150 × 60 m) acorn was under-sown in the amount of 700 kg/ha. After regeneration cutting in the winter of 2005/2006, in the regeneration site, a relatively small number of ash and oak trees were detected (10–12 trees/ha), so both seedlings and juvenile oak plants were growing in the light in the opened canopy. Final cutting or removing the trees from the previous stand was done during of the growing season of 2007/2008 when the juvenile oak plants were two years old.

For the purposes of this research, the central part of regeneration area was singled out in the spring of 2006 as the experimental area. In these experimental plots, powdery mildew was suppressed with 8 different fungicides during three growing periods. Used fungicides, active substances and their concentrations are shown in the Table 1.

This experiment was set up according to the random block system of experimental fields (size 8 × 3 m) in 4 replications. Hand sprayer CP 3 manufactured by Cooper, Pegler & Co Ltd was used for the fungicide application. In the experimental fields except the control areas, all pedunculate oak plants were sprayed. The number of plants within the field varied and it was in the range from 35 to 80 plants (1.46–3.33 plants/1 m<sup>2</sup>). In 2006 the oak seedlings were treated 5 times: on May 25, June 2, June 16, July 6 and August 25, while the juvenile oak

**TABLE 1**  
Fungicides used in the experiment.

Fungicide	Active substances	Used concentrations
1. Microthiol special	elemental sulfur	0.5%
2. Strobby DF	kresoxim-methyl	0.02%
3. ZATO 50-WG	trifloxystrobin	0.01%
4. Cabrio top	pyraclostrobin + metiram	0.20%
5. Quadris	azoxystrobin	0.075%
6. Alert S	carbendazim + flusilazole	0.10%
7. Duett Ultra	thiophanate-methyl + epoxiconazole	0.10%
8. Sabithane	dinocap + myclobutanil	0.04%

plants were treated once in the early springs of 2007 and 2008. Plants infection and fungicide efficiency evaluation were determined by visual estimation of powdery mildew presence on all leaves of 25 randomly picked plants within every experimental field. Level of plants infection in individual treatments was determined based on the average percent of coverage of leaves by mycelium of the fungus and it is calculated based on the criteria given in Table 2.

Plant growth elements were determined during the vegetation rest and based on multistage shoot forming and diameter in the level of root neck (15, 20). In this case the plants picked were marked with permanent marks in the root neck diameter area and therefore the subject of evaluation and analysis were the same plants. For data processing we used ANOVA and Duncan test for significance threshold of  $P = 0.05$ .

Changes in the regeneration site by weed species were registered during the research and shown chronologically according to vegetation period.

In order to suppress the weed vegetation, adequate herbicides were used in the whole regeneration area and therefore also in the experimental area. In this way smooth application of fungicides to oak leaves was possible during a prolonged period of time.

**TABLE 2**

Criteria for evaluation of fungal infection of leaves.

Infection scale	Evaluation of leaf coverage with fungal mycelium (%)	Level of infection
0	0	No infection
1	0.1–2	Traces of infection
2	2.1–10	Weak infection
3	10.1–30	Medium infection
4	30.1–50	Medium strong infection
5	50.1–75	Strong infection
6	▶ 75	Severe infection

## RESULTS

### Occurrence and control of powdery mildew in the first year of pedunculate oak development

Powdery mildew attack before and after fungicide treatments in the first year is shown in Table 3. Initial level of plant infection was determined on May 24, 2006 right before the first treatment. The mildew infection was weak in each elementary field and it was in the range of 1.0–1.8%. But after the first treatment highly significant differences were found in the presence of powdery mildew on leaves of treated and untreated plants (Table 3). It should be noted that the powdery mildew infestation of untreated plants during May and June was weak and did not severely affect the development of seedlings. In the rating that was conducted in early July (July 5, 2006), after the third treatment, the average infection rate of untreated plants reached 15.4% and had the character of a strong medium infection. After the fourth treatment and evaluation that followed in mid-July (July 14, 2006) percentage of infected plants in the control area reached the level of 32.6%. Powdery mildew was recorded sporadically on the leaves of treated plants. All the fungicides showed high efficacy in control of powdery mildew, as they were used preventively or when the first symptoms occurred. The last treatment of seedlings was conducted in late August (August 25, 2006) with an average infection rate of the treated plants 10.0 to 22.8%. In plants in the control area more than half of the leaf surface was covered with the mycelium. It was noted that the last plant protection by fungicides was less effective than previous treatments. The reason should be sought in the fact that the protection was conducted in the period when powdery mildew was significantly present on the leaves. Consequently, the effect of fungicides was more curative rather than preventive. We think that the last treatment should be carried out during the second half of July and by early August, a time when the plants have formed the last phase shoot.

The pedunculate oak seedlings have been exposed to the powdery mildew attack from the beginning to the end of the growing season. Such long exposure indicated

**TABLE 3**

The average degree of leaves infection in the percentages before and after treatments with fungicides in 2006.

Product	1st treatment: May 25, 2006		2nd treat- ment: June 2nd, 2006	3rd treat- ment: June 16, 2006	4th treat- ment: July 6, 2006	5th treatment: August 25, 2006	
	May 24, 2006 evaluation before the 1st treatment (1)	June 1, 2006 evaluation after the 1st treatment (2)	June 15, 2006 evaluation after the 2nd treatment (3)	July 5, 2006 evaluation after the 3rd treatment (4)	July 14, 2006 evaluation after the 4th treatment (5)	August 24, 2006 evaluation before the 5th treatment (6)	Sept. 6, 2006 evaluation after the 5th treatment (7)
<b>Average leaves infection with powdery mildew (%)</b>							
Microthiol special	1.8	0.6 b	0.6 b	0.2 b	0.2 b	10.0 bce	8.8 bce
Stroby DF	1.5	0.1 b	0.2 b	0.3 b	0.2 b	18.6 b	11.2 bc
Zato	1.3	0.1 b	0.2 b	0.1 b	0.2 b	16.7 bc	13.7 bc
Cabrio top	1.0	0.1 b	0.2 b	0.2 b	0.2 b	22.8 b	17.5 b
Quadris	1.0	0.0 b	0.3 b	0.1 b	0.2 b	20.4 b	11.6 bc
Alert S	1.0	0.1 b	0.1 b	0.2 b	0.0 b	11.8 bce	9.9 bc
Duett Ultra	1.5	0.1 b	0.2 b	0.2 b	0.0 b	15.6 bc	10.9 bc
Sabithane	1.0	0.1 b	0.2 b	0.4 b	0.3 b	17.9 b	10.2 bc
control	1.1	2.1 a	8.3 a	15.4 a	32.5 a	50.5 a	50.4 a
p blocks	–	0.2130 ns	0.6721 ns	0.4867 ns	0.3441 ns	0.5452 ns	0.3724 ns
F blocks		1.61	0.53	0.84	1.16	1.43	2.35
p treatments	–	0.0004***	0.0000***	0.0000***	0.0000***	0.0000***	0.0000***
F treatments		5.74	12.74	25.00	76.22	31.39	104.81

the need for their multiple protection with fungicides. The presence of powdery mildew on leaves progressively increased and reached a maximum in late August when more than 50% of the assimilation surface of untreated plants was covered with epiphytic mycelium. Used fungicides did not show significant differences in efficiency (Table 3). Only the product Duett Ultra had a mild phytotoxic effect on the oak seedlings. Symptoms of transient chlorosis and leaf corrugation were observed on the plants immediately after treatment with the above mentioned fungicide.

It should be noted that the vegetation period in 2006 was marked by frequent and abundant precipitation. Prolonged heavy rains were unfavorable for powdery mildew spreading as they washed down conidia from the

infected plant parts and slowed the development and expansion of epiphytic mycelium. On the other hand, the conditions for the development of seedlings were favorable due to abundant rainfall. In the experimental plants, the oak biological feature of multistage forming of shoots with leaves by activation of embryonic terminal buds during the growing season was easily observable (Table 4). Presence of the stage growth of the pedunculate oak and the participation of individual shoots were almost equal in treated and untreated plants. The second phase of growth was present in all plants and the property of forming the phase shoots for the third time was noticed in 85% of treated and 82% of untreated plants. Growth in height after the third break was equally manifested both in the treated and untreated plants (Table 4).

In accordance with the multistage growth of plants during the growing period in 2006, we think that the oak seedlings could be adequately protected even with fewer treatments (3–4), if the fungicide protection had been synchronized with the periods of phase growth.

**TABLE 4**

Characteristics of height growth in the first year of development of the pedunculate oak.

Stage growth	Stage development of oak (%)	
	Treated plants	Untreated plants
I	100	100
II	100	100
III	85	82
IV	29	29

**Ground cover formation on the regeneration site in the first year of the pedunculate oak development**

During the the first part of the growing season, oak seedlings were developing in the regeneration area without significant competition of herbaceous and woody



weeds. The regeneration of shoot tree species and appearance of herbaceous weed vegetation were prevented by application of total herbicides in the period of preparing the regeneration stand. In addition, ground and evenly dispersed parts of woody biomass and non decomposed forest ground level on the soil surface significantly prevented the colonization by herbaceous species. Thus, the regeneration area has been relatively little overgrown by weed species until the beginning of July, which enabled the smooth development of seedlings and application of fungicides (Fig. 1).

Ground-cover which appeared in the second part of the growing season in the regeneration area was formed mostly from herbaceous species of terophyte/hamephyte and hemicryptophyte life forms. The highest abundance and ground coverage had species: *Stenactis annua* – vegetative stage, *Solanum nigrum*, *Polygonum lapatifolium*, *Erigeron canadensis*, *Galinsoga parviflora*, *Lamium purpureum*, *Urtica urens*, *Viola arvensis*, *Bidens tripartitus*, *Symphytum officinale*, *Geum urbanum*, *Glechoma hederacea*, *Juncus effusus*, *Carex remota*, *Rumex crispus*, which formed horizontal canopy where seedlings had favorable development conditions. On the other hand, suppression of powdery mildew could still be done (Figure 2).

Only individually or in small oases, the oak seedlings were threatened by weed species which form vertical canopy (*Galium aparine*, *Rubus caesius*, *Rosa canina*). Observations at the experimental plot and habitat conditions indicated that after a well performed start in the development of seedlings (sufficient number of seedlings per area and well preserved assimilation apparatus from oak mildew infestation), subsequent growth of weed vegetation did not represent the dominant limiting factor in seedlings development.

### Occurrence and suppression of powdery mildew in the second year of oak development

Juvenile oak plants were protected with fungicides only once in the second vegetation period and it was conducted in early spring (May 9, 2007). The protection was predominantly preventive because fungicides were



Figure 1. The seedlings and regeneration area (June 2, 2006).

applied before the first symptoms of infection on the leaves. Assessment showed that there was no statistically significant difference in the presence of powdery mildew between the treated and untreated plants (Table 5). Bearing in mind that a weak powdery mildew infestation of the juvenile oak plants during May (during the formation of primary shoots) could not endanger the development of plants, it could be concluded that fungicidal protection was not necessary. The treatment was applied during long spring drought and therefore some fungicides showed phytotoxic effects on plants (Fig. 3).

Duett Ultra fungicide exerted a high phytotoxic effect such as severe chlorosis and partial loss of leaves, while the fungicides Alert S and Microthiol special exhibited less depressive effect on plants as a mild and transient chlorosis.

During the first half of June, the majority of plants formed phase shoots, the so called »second growth« with thin and young mesophylic leaves. Our observations suggest that the leaves formed during this period were to a greater degree infected with fungus comparing to the primary shoots formed in the beginning of the growing season (Table 5). Therefore, the untreated plants showed 2.4% of powdery mildew infection in the beginning of the growing season, but according to the second evalua-



Figure 2. The regeneration area (August 24, 2006).



**Figure 3.** The symptoms of phytotoxic effects of Duett Ultra fungicide (May 24, 2007).

tion performed on June 26, powdery mildew covered 5.6% of the leaf surface of plants. The juvenile oak plants were covered by weed vegetation already in mid-June, which disabled further fungicide protection. The powdery mildew presence and significance when the weeds, with its height and mass, covered the oak juvenile plants were analyzed at the end of June and mid August of 2007 (Table 5). Average coverage of leaves with powdery mildew at the end of June remained at 6.1% and was significantly higher than in the assessment conducted in mid-August (3.1%). Increased presence of powdery mildew on plants in mid-growing season is explained by the formation of phase shoots with young leaves. Since the middle of the growing season the forming of phase shoots was not observed and leaves which had been previously formed by aging developed a thicker epidermis and a

**TABLE 6**

Characteristics of height growth in the second year of the pedunculate oak development.

Stage growth	Stage development of the pedunculate oak (%)	
	Treated plants	Untreated plants
I	100	100
II	46	16
III	3	3
IV	0	0

thicker cuticle, making them more resistant to new infections. The biological defense reaction of plants limited further development and expansion of epiphytic mycelium on the leaves. In this way, the plants were gradually freed from the presence of the fungus (Table 5). It is interesting that the observation in August (August 14, 2007) revealed that the powdery mildew infestation of the untreated plants was the weakest. The reason should be sought in the weaker development of these plants compared to the treated plants. During the year of 2007, only a small number of non-treated plants (16%) had primary axis of shoots elongated and formed new leaves, while this phenomenon was found in 46% of the treated plants (Table 6).

Plants that were protected from the powdery mildew infection at the seedling stage, had a more successful start and more intense growth in the second year of develop-

**TABLE 5**

The average degree of leaves infection in % after treatment with fungicides in 2007.

Product	Treatment: May 9, 2007	June 26, 2007 evaluation in June (2)	August 14, 2007 evaluation in August (3)
	May 24, 2007 evaluation after the treatment (1)		
Average powdery mildew infection of leaves (%)			
Microthiol special	0.9 ab	8.6 a	4.5 a
Stroby DF	0.8 ab	6.8 a	5.3 a
Zato	1.4 ab	4.7 a	3.2 a
Cabrio top	2.1 ab	6.7 a	2.6 a
Quadris	2.1 ab	6.9 a	3.8 a
Alert S	0.9 ab	4.9 a	2.2 a
Duett Ultra	0.7 ab	6.4 a	2.7 a
Sabithane	0.9 ab	4.3 a	2.4 a
control	2.4 a	5.6 a	1.5 a
Average infection of juvenile plants	1.4%	6.1%	3.1%
p blocks	0.3934 ns	0.2177 ns	0.2877 ns
F blocks	0.82	1.59	1.33
p treatments	0.1265 ns	0.9118 ns	0.5662 ns
F treatments	0.74	0.40	0.85

**TABLE 7**

The average degree of leaves infection in % after treatment with fungicides in 2008.

Product	Treatment: May 16, 2008		June 30, 2008 evaluation in June (3)	August 19, 2008 evaluation in August (4)
	May 23, 2008 evaluation after the treatment (1)	June 3, 2008 evaluation after the treatment (2)		
Average powdery mildew infection of leaves (%)				
Microthiol special	5.5 ab	2.4 a	18.5 a	9.6 a
Stroby DF	5.5 ab	4.3 a	16.4 a	11.6 a
Zato	8.1 ab	5.0 a	16.1 a	8.0 a
Cabrio top	10.2 ab	3.8 a	16.1 a	8.8 a
Quadris	10.2 ab	3.8 a	15.4 a	8.4 a
Alert S	5.6 ab	1.7 a	18.4 a	9.5 a
Duett Ultra	5.8 ab	1.0 a	10.5 a	7.0 a
Sabithane	5.2 ab	2.0 a	14.2 a	12.0 a
control	10.6 a	5.1 a	14.7 a	10.7 a
Average infection of juvenile plants	7.4%	3.2%	15.6%	9.5%
p blocks	0.5882 ns	0.5144 ns	0.3721 ns	0.2751 ns
F blocks	0.66	0.78	2.01	1.37
p treatments	0.0820 ns	0.6127 ns	0.4339 ns	0.9705 ns
F treatments	2.01	0.79	0.50	0.27

ment in comparison with untreated plants. The elements of the multistage plant growth point out that fact very clearly (Table 6).

**Ground cover formation on the regeneration site in the second year of the pedunculate oak development**

In the second year of the pedunculate oak development there was a change in qualitative and quantitative participation of certain types of herbaceous vegetation in the regeneration site. From mid June until the end of the growing season in the regeneration site, a phase of »high weeds« was present. The highest frequency and coverage was species *Stenactis annua* (Nes.). The above mentioned

weed species, forming the generative phase, overgrew juvenile oak plants and that prevented further application of fungicides at the regeneration site (Figure 4).

Despite competition at a high level with weed plants at the regeneration area, juvenile oak plants developed well within of that layer and had, depending on climatic conditions and the amount of moisture in the soil, more or less intense height growth. During this period, powdery mildew did not significantly disturb the development of plants.

**Occurrence and suppression of mildew in the third year of the pedunculate oak development**

Juvenile oak plants in the third year of development have been treated on May 16, 2008 when the leaves showed weak intensity of mildew infection. The efficiency of the fungicides was evaluated on May 23, (7 days after the treatment) and on this occasion it was found that, in respect to the presence of powdery mildew on the leaves, there were no statistically significant differences between the treated and untreated plants (Table 7). Summing up the applied fungicides, had little impact on plants. In the treated plants, the infection varied in the range from 5.2 to 10.2% and it was 10.6% in the untreated. According to evaluation done on June 3, (17 days after the treatment) powdery mildew was less present compared to the first evaluation on May 23. This can be

**TABLE 8**

Characteristics of height growth in the third year of the pedunculate oak development.

Growth Stage	Stage development of the pedunculate oak (%)	
	Treated plants	Untreated plants
I	100	100
II	92	94
III	8	11
IV	0	0





**Figure 4.** The weed vegetation and appearance of the regeneration area in the second year of pedunculate oak development (July 26, 2007).

explained by biological response of the plants, where the young leaves from the first phase of growth developed completely in a relatively short period and prevented new infections or penetration of the fungus in the leaf tissue.

The third evaluation of the powdery mildew presence on plants was conducted at the end of June. Powdery mildew infection of plants was several times higher than the evaluation conducted in early June. The plants have in the meantime formed new shoots with young leaves, which was the reason for their increase infection. Therefore, secondary shoots were infected in a greater degree

with powdery mildew comparing to the primary shoots. During July and August, plants formed very small percentage of new shoots so the final evaluation (August 19, 2008) showed significantly less presence of powdery mildew on plants (Table 7).

Juvenile oak plant development in the third year may also be presented through the characteristics of multistage growth. The juvenile oak plants showed no difference in the number of phase shoots, since over 90% of individuals completed their development in height by forming the second increment (Table 8).

**TABLE 9**

Influence of powdery mildew control on height growth and diameter increment of the pedunculate oak in the 2006 – 2008 period.

Product	Height of one year-old plants (cm)	Root neck diameter of one year-old plants (mm)	Height of two-year old plants (cm)	Root neck diameter of two-year old plants (mm)	Height of three-year old plants (cm)	Root neck diameter of three-year old plants (mm)
Microthiol special	27.3 a	6.1 a	43.1 ab	8.3 a	67.9 ab	10.0 b
Stroby DF	25.9 a	6.2 a	43.4 ab	8.5 a	69.5 ab	11.1 ab
Zato	28.1 a	6.6 a	46.4 ab	8.0 a	68.5 ab	11.4 ab
Cabrio top	27.3 a	7.0 a	46.0 ab	8.5 a	65.6 ab	11.3 ab
Quadris	28.2 a	6.9 a	44.6 ab	8.4 a	67.5 ab	10.7 ab
Alert S	26.9 a	6.7 a	44.0 ab	8.6 a	66.8 ab	11.7 ab
Duett Ultra	25.9 a	7.2 a	40.2 bc	8.1 a	60.4 bc	9.9 b
Sabithane	26.0 a	6.4 a	41.7 bc	8.0 a	61.9 bc	10.3 b
control	23.9 b	5.6 b	37.3 c	7.0 b	57.9 c	8.9 c
p blocks	0.1873 ns	0.2390 ns	0.0025*	0.0107*	0.0014*	0.0020*
F blocks	1.39	1.50	3.39	2.19	5.97	6.66
p treatments	0.0144*	0.0101*	0.0017*	0.0058**	0.0021*	0.0159*
F treatments	3.13	3.36	4.57	3.73	4.43	3.06





**Figure 5.** The regeneration site in the third year of the pedunculate oak development (June 30, 2008).

### Ground cover formation on the regeneration site in the third year of the pedunculate oak development

After the final cutting conducted in non-vegetative period in 2007/2008, the regeneration area was completely open. At the end of May, weed vegetation overgrew juvenile oak plants that were till the end of vegetation developing in conditions of high weediness. Dominant weed species was creeping thistle (*Cirsium arvense* L. (Scop.) which covered more than 70% of the regeneration site (Fig. 6). Juvenile pedunculate oak plants in competition with weeds had satisfactory conditions for development, while the influence of powdery mildew was without practical value for cultivated plants.

### Influence of fungicidal protection of seedlings and juvenile pedunculate oak plants on growth and development in height and thickness

Significant differences in the achieved height and root neck diameter growth of the treated and untreated plants indicated the positive effects of fungicidal protection (Table 9). The mildew infection significantly reduced the height and diameter growth of one year old plants and had a negative impact on their later development. Even though it was a surface that had been prepared for regeneration using uniform technology, the differences in repetitions were observed after the second and third year of juvenile oak plant development. These differences suggest that the variability of height and root neck diameter growth of plants is a result of natural processes of differentiation due to different microsite conditions and the effect of of cenological composition of weed species that were not controlled and monitored in our experiment. This fact does not allow us to completely isolate and observe the effect of fungicidal protection in

the later stages of plant development. Significant variations in the actual values of height and root neck diameter growth of the pedunculate oak are primarily the consequence of a broad norm of reaction and the effect of exogenous factors on micro locations. Juvenile oak plants in the second year of development were subjected to the equal impact of fungicides on one hand and other environmental factors on the other (based on gained F values). In the third year of plant growth, environmental factors predominantly influenced the variability of height and root neck diameter increment of the analyzed specimens.

### DISCUSSION

The presence of powdery mildew on oak seedlings and juvenile pedunculate oak plants in a wide range of environmental conditions in the Ravni Srem area launched a series of questions about the real need and justification for fungicidal protection in the initial stages of the oak development. Answer to the questions of the optimal number of treatments and efficiency of used fungicides in controlling powdery mildew have not been fully determined. Also, age limits of juvenile plants, when the pesticide application is justified from biological and economic points of view, are not completely defined. Oak powdery mildew in the previous research conducted in the Gornji Srem area (13, 15, 20, 24) was mostly perceived as an ecopathological phenomenon. Bobinac and Karadžić (13) indicated the need for preventive protection of pedunculate oak seedlings from powdery mildew in the process of natural or artificial regeneration of stands. The experiences gained in control of powdery mildew on seedlings and juvenile oak plants in Ravni Srem over the past two decades also pointed out that the preventive protection has had a positive impact on the development of plants. Since more complex research of

this issue was not conducted in this area, stand that point out the positive effect of chemical protection of the pedunculate oak was mainly based on observations of practitioners, rather than experimentally confirmed results.

Exploring the natural regeneration of the pedunculate oak in the Gornji Srem area and recovery methods depending on the site and stand conditions, Bobinac (15) in his PhD dissertation presents a series of valuable observations on the occurrence of oak mildew and problems related to its control. Indicating that the greatest danger threatening the oak seedlings from powdery mildew at the beginning of the growing season (due to less developed cuticle), the author particularly discusses a feature of multistage height growth of oak and its periodical forming of leaves during the growing season. Studies (13, 20, 25) suggest that environmental conditions are crucial for multistage growth, which is highly expressed at oak in the initial stages of ontogeny. Our studies have confirmed the biological characteristic of the pedunculate oak to give two or more increment in height in the same vegetation period. Growth characteristics of seedlings and juvenile oak plants indicated a different biological response of plants to biotic and abiotic factors, wherein oak powdery mildew was not a limiting factor in all stages of plants development. At the seedlings stage, powdery mildew had a significant effect on height and diameter growth of plants (Table 9), while its importance was suppressed at later stages of the pedunculate oak development. Time and the need for preventive care of seedlings should be defined on characteristics of plant growth, especially multistage growth. Since in our experiment over 80% of plants in the first year of development formed the third, and about 30% them at the fourth phase of growth (Table 4), this indicating that the same number of treatments should be used to protect the pedunculate oak from mildew. The control of powdery mildew should be synchronized with the extension of primary axis of new shoots and formation of leaves, for which phenological observations of multistage plant growth are needed. This is also one of the most important aspects in protecting oak seedlings from powdery mildew infection, because with synchronization of applied protective measures of this type (which follow a multistage growth of plants), the fungus can be effectively controlled and eliminated (smaller number of treatments during the growing season, more rational protection, minimum use of fungicides, etc.). Oak habitats in the area of Gornji Srem, (15, 20) and (13) show that over 90% of individuals extend primary axis and their growth in the period from April to September, takes place in a maximum of four phases. According to Gruber (26), this feature is particularly expressed in the pedunculate oak at the initial stages of its ontogeny. Multistage height growth in our experiment was a generally represented phenomenon, as practically all one-year individuals at least once during the growing season prolonged primary axis of shoots and formed new leaves (Table 4).

Under the favorable conditions and micro conditions for growth, the pedunculate oak as a species with a

significant ability of multistage growth, despite some limiting factors (e.g. oak powdery mildew), may become the dominant type in the initial stages of regeneration (15). In the process, preventive chemical control of phase shoots and leaves during the growing season speeds up the process of ontogenesis where oak becomes the dominant in height growth compared to other generative regeneration trees and shrubs. In our experiment, the oak seedlings developed very well, which can be seen in the participation of phase shoots of untreated plants and also in the total achieved height (Table 4 and 9). Claims of Škorić (27) are also interesting since he believes that the cause for the multi-stage formation of shoots and leaves is oak powdery mildew that causes drying and falling off of previous leaves. Our results indicate that oak powdery mildew was not the cause of the formation of new shoots. The occurrence of multistage growth of the pedunculate oak was in the first place stimulated by environmental factors.

Pointing out that the initial stages of the pedunculate oak development are crucial to the process of regeneration on differently prepared regeneration areas, (15) found that oak seedlings in the second part of the growing season find favorable conditions for development within the newly formed layer of herbaceous plants (mostly therophytic/chamaephytic life forms) that form predominantly horizontal canopy (28, 29). On the regeneration sites prepared in such way, preventive care of the pedunculate oak from powdery mildew is necessarily carried out only in the first half of the growing season until the seedlings are covered with herbaceous vegetation. If young leaf is completely formed before favorable conditions for powdery mildew development, posterior infection cannot destroy the leaf and chemical treatment is not necessary (15). Based on multiannual observations of the vulnerability of seedlings to oak powdery mildew, the above mentioned author concludes that the environmental conditions in the predominantly uniformly regenerated areas are suitable for development of oak and chemical protection measures should be performed if it is necessary. This position also substantially confirms the generally applied system of artificial regeneration of the pedunculate oak in the past (agro-forestry management) in the region of Srem. The knowledge that the pedunculate oak, without preventive protection measures from powdery mildew, develops very well in the canopy of wheat culture, has been used in practice to this day in Srem, and in the countries of Central Europe, the pedunculate oak is regenerated in this way.

In the second and third year of juvenile plant development, powdery mildew attack varied during the growing season and was directly related to the formation of stage shoots and characteristics of the ground cover formation. The leaves on primary shoots from the beginning of the growing season were significantly less infected with powdery mildew compared to the leaves on the stage shoots that were formed later (in June and July). The same conclusions were reached in the research of (30) and (31) stating that the leaves of the second and

third increments formed in the summer months were more susceptible to mildew infection than primary shoots. Preventive protection of juvenile oak plants at the stage of forming primary shoots had a modest effect (Tables 5 and 7), while in the period of forming second increment (when weeds have outgrown oak plants), protection was unreasonable and unnecessary from a biological point of view. The phase »of high weeds« occurred on the regeneration site in the second year of oak development. The largest in number and coverage was the species *Stenactis annua* (Nes.) which is often present in semi-open and open pedunculate oak habitats (15, 28, 29).

In the second year of the pedunculate oak development significant variation in the actual elements of growth in height and diameter was determined. The previous multiannual research (15) also indicated large differences in the height increment of seedlings and juvenile oak plants at the micro areas and in different procedures of pedunculate oak stands regeneration in Srem. The author states that »individual change never occurs at the regeneration areas, but the continuous changes of certain elements which take place and last through several stages of plants development«.

## CONCLUSIONS

Based on the results of three-year experimental research where the importance and need for control of oak mildew in the process of regeneration of the pedunculate oak stands in the selected type of forest in the Ravni Srem area was studied, the following conclusions can be made:

- Control of powdery mildew on seedlings and juvenile oak plants is an important measure of protection that is required in the regeneration areas during the period of regeneration cutting.
- The fungicides studied showed high efficacy in controlling oak powdery mildew and can be recommended for practical use. Duett Ultra product should not be used because of the manifested phytotoxic effects on juvenile oak plants.
- Knowledge of multistage growth is of great importance in the early ontogenesis of the pedunculate oak in terms of preventive care application against powdery mildew infection. It is very important to synchronize the preventive protection of seedlings with multistage height growth of seedlings and juvenile oak plants in conditions suitable for the development of powdery mildew. In such proceedings, the application of fungicides is reduced to minimum and thereby provides a rational protection of juvenile oak plants. In this way, the rapid development of seedlings is enabled, as well as favorable level of development and readiness of shoots for winter and successful growth of plants in the following spring.
- Well-formed one year and two year oak plants tolerate the presence of powdery mildew very well and the fungus is no longer the limiting factor in plant development.
- Seedlings and juvenile oak plants in the regeneration area, within the layer of herbaceous weed species, had more or less intense increase in height depending on climatic conditions and the amount of humidity in soil.
- The results presented in this research can serve as a model for planning protection measures in other complexes of pedunculate oak forests in their restoration process.
- In order to completely perceive the importance of oak powdery mildew in the regeneration process of oak stands in the Ravni Srem area, it would be necessary to continue the research.

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## REFERENCES

- BUTIN H, ZYCHA H 1973 Forstpatologie, Thieme Verlag, Stuttgart, p 1–177
- KIŠPATIĆ J 1991 Šumarska fitopatologija. Sveučilišna naklada Zagreb, str. 1–356
- MANOJLOVIĆ P 1926 Sadašnje stanje hrastovih šuma u Slavoniji, Pola stolecja šumarstva 1876–1926. Zagreb, str. 372–385
- JOSIFOVIĆ M 1929 Pepelnica (medljika) (*Microsphaera quercina* (Schw.) Burr.) i sušenje hrasta u Posavskim šumama. Izdanje Instituta za naučna šumarska ispitivanja pri Šumarskom odseku Poljoprivrednog fakulteta, Beograd, str. 1–14
- VAJDA Z 1974 Uzroci sušenja slavonskih šuma, Zbornik o stotoj obljernici šumarstva jugoistočne Slavonije, posebno izdanje, knjiga 1, Vinkovci-Slavonski Brod, str. 221–238
- LANIER L, JOLY P, BONDOUX P, BELLEMÈRE A 1976 Mycologie et Pathologie Forestières, Tome II – Pathologie forestière. Masson, Paris, p 1–478
- ABGRALL J F, SOUTRENON A 1991 La forêt et ses ennemis. Cemagref, Grenoble, p 1–399
- GLAVAŠ M, HALAMBEK M 1992 Mikoze hrasta lužnjaka i kitnjaka. Šumarski fakultet Zagreb. Glasnik za šumske pokuse 28: 237–244
- SHIRNINA L V 1990 Effect of systemic fungicides on the viability of powdery mildew of oak (*Quercus*). Mikologiya i Fitopatologiya (Voronezh) 23(5): 481–485
- SHIRNINA L V 1992 The biological effect of systemic fungicides in the control of oak powdery mildew. Lesnoe – Khozyaistvo 1: 48–49
- NOVAK-AGBABA S, HALAMBEK M 1993 Značenje pepelnice (*Microsphaera alphitoides* Griff. et Maubl.) u procesu sušenja hrastovih šuma i njeno suzbijanje. Radovi (Šumarski Institut Jastrebarsko) 28 (1-2): 13–24
- NOVAK-AGBABA S, LIOVIĆ B, MATOŠEVIĆ D 1994 Novi fungicidi u suzbijanju pepelnice na hrastovom podmlatku. Radovi (Šumarski Institut Jastrebarsko) 29 (1): 37–47
- BOBINAC M, KARADŽIĆ D 1994 Zaštita ponika lužnjaka (*Quercus robur* L.) od hrastove pepelnice (*Microsphaera alphitoides* Griff. et Maubl.) – mere za smanjenje rizika semene obnove. Zaštita bilja danas i sutra, Beograd, str. 617–627
- SELOCHNIK N N 1996 Effects of powdery mildew and fungicides on oak seedlings. Lesnoe – Khozyaistvo 6: 51–53
- BOBINAC M 1999 Istraživanja prirodne obnove lužnjaka (*Quercus robur* L.) i izbor metoda obnavljanja u zavisnosti od stanišnih i sastojinskih uslova. Doktorska disertacija, Šumarski fakultet Univerziteta u Beogradu, str. 262
- JACOBS P 2003 Mildew (*Microsphaera alphitoides* Griff. et Maubl.) as possible important factor in limiting height growth of oak (*Quercus robur* L.). MSc thesis Forest and Nature Conservation.

- Wageningen University, <http://www.fem.wur.nl/United Kingdom/Publications>.
17. LIOVIĆ B, ŽUPANIĆ M 2006 Ispitivanje djelotvornosti fungicida za suzbijanje gljive *Microsphaera alphitoides* (Griff. et Maubl.) na hrastovom podmlatku. *Radovi* (Šumarski Institut Jastrebarsko) (izvanredno izdanje) 9: 181–188
  18. KARADŽIĆ D 2010 Šumska fitopatologija, Univerzitet u Beogradu. Šumarski fakultet, Beograd, str. 774
  19. AVRAMOVIĆ G, POLJAKOVIĆ-PAJNIK L, VASIĆ V, PAP P 2008 Zaštita šuma tvrdih lišćara od bolesti i štetočina. Monografija 250 godina šumarstva Ravnog Srema, JP Vojvodinašume, Šumsko Gazdinstvo Sremska Mitrovica, poglavlje III: Gajenje i zaštita šuma, str. 147–160
  20. BOBINAC M 1994 Višefazni rast u visinu jednogodišnjih biljaka lužnjaka (*Q. robur* L.) i neki aspekti značajni za semenu obnovu. Beograd, *Šumarstvo* (1-2): 47–57
  21. JOVIĆ D, JOVIĆ N, JOVANOVIĆ B, TOMIĆ Z, BANKOVIĆ S, MEDAREVIĆ M, KNEŽEVIĆ M, GRBIĆ P, ŽIVANOV N, IVANIŠEVIĆ P 1994 Tipovi šuma Ravnog Srema – Atlas. Šumarski fakultet Univerziteta u Beogradu. Geokarta 1–28, Beograd.
  22. BOBINAC M 2007 Oplodna sječa u šumi hrasta lužnjaka i poljskog jasena u Srijemu i njene specifičnosti. *Radovi* (Šumarski Institut Jastrebarsko) 42 (1): 35–46
  23. BOBINAC M 2008 Savremeni pristup obnovi šuma tvrdih lišćara na području Ravnog Srema. Monografija 250 godina šumarstva Ravnog Srema, JP Vojvodinašume, Šumsko Gazdinstvo Sremska Mitrovica, poglavlje III: Gajenje i zaštita šuma, str. 127–135
  24. BOBINAC M 2000 Stand Structure and Natural Regeneration of Common Oak in the Nature Reserves »Vratična« and »Smogva« near Morović. OAK 2000 – Improvement of Wood Quality and Genetic Diversity of Oaks. *Glasnik za šumske pokuse* 37: 295–309
  25. BOBINAC M, VILOTIĆ D 1994 Višefazni rast jednogodišnjih jedinki hrasta lužnjaka (*Quercus robur* L.) sa aspekta morfološke analize poprečnih preseka ose izdanka, Deliblatski pesak – Zbornik radova VI, Pančevo, str. 399–406
  26. GRUBER F 1992 Dynamik und Regeneration der Gehölze. Habilitationsschrift Forstwissenschaftl. Fachbereich der Universität Göttingen. Berichte Forschungszentrum Waldökosysteme, Reihe A/Bd. 86, Teil I: Ergebnisse, p 1 – 419
  27. ŠKORIĆ V 1926 Uzroci sušenja naših hrastovih šuma. *Glasnik za šumske pokuse* 1: 234–246
  28. BOBINAC M, RADULOVIĆ S 2000 Promene u prizemnom pokrivaču podmladnih površina na staništu šume lužnjaka i jasena (*Fraxino-Quercetum roboris aceretosum* Jov. et Tom. 1980). *Acta herbológica* 8 (2): 49–58
  29. BOBINAC M, RADULOVIĆ S 2002 Prilog proučavanju prizemnog pokrivača podmladnih površina posle primene herbicida na staništu šume lužnjaka i jasena (*Fraxino-Quercetum roboris* Jov. Et Tom. 1979). Zbornik rezimea, XII simpozijum o zaštiti bilja i Savetovanje o primeni pesticida, Zlatibor, str. 91
  30. MIRTČEV S, IVANOVA E 1996 Prediction of oak powdery mildew epidemics by dendrochronological analysis, Conférence du Groupe de Travail Européen pour la Dendrochronologie EuroDendro 96, Moudon. [http://www.lrd.ch/congres/lrd\\_lect97.htm](http://www.lrd.ch/congres/lrd_lect97.htm)
  31. MARÇAIS B 2007 Impact of Climate on Oak Powdery Mildew, Acta Silvatica J Lignaria Hungarica. Special Edition, Proceedings of the Conference of IUFRO Working Party 7.02.02, 21–26 May 2007. Sopron, Hungary, p 277