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IMPACT OF OAK LACE BUG *Corythucha arcuata* (HETEROPTERA: TINGIDAE) ON PEDUNCULATE OAK (*Quercus robur*) SEED

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Introduction

Pedunculate oak (*Quercus robur* L.) is the most valuable tree species in Croatia due to the high quality of wood, but management of oaks forests is demanding [1]. Over the past five to six years this particular tree species has suffered heavily from the attack by a leaf sucking insect known as the oak lace bug (OLB), *Corythucha arcuata* [2]. It has established a stable population across Europe, where it is an alien (allochthonous) species [3]. As it has no natural enemies in the ecosystem, it has become an unequaled invasive species [4]. This invasive alien species (IAS) is a very important negative biotic factor which threatens the health of pedunculate oak and hinders sustainable forest management [5]. Foliar damage occurs in spring, after overwintering, when adults start to feed on leaves and lay eggs in late April or early May. The greatest damage occurs after larvae hatch from masses of eggs in early June and feed intensively on the underside of leaves, causing discoloration and destruction of leaf tissue with subsequent reduction of the assimilation surface. Initially, the infected leaves are covered with small black excrements and yellowish dots that represent OLB feeding. Intensive feeding from May gives an autumnal appearance to the entire tree by late July and early August [6]. Consequently, the second and possibly the third OLB generation exerts an additional negative impact [5]. Eventually, in the period of acorn fructification the leaves turn yellow and dry, without any green surface. In combination with other negative abiotic and biotic factors, this results in premature fall of acorns [7]. The negative impact of OLB infestation on reproduction causes particular concern in view of the fact that Croatian lowland oak stands have high ecological and economic importance. Their importance is documented by their share in state owned forests, where pedunculate oak (*Q. robur* L.) accounts for 14.9% (or 45.0 million m^3) of the growing stock and the amount of pedunculate oak in allowable cut reaches up to 13.6% (or 0.79 million m^3) [1].

Reduced seed weight, lower germination rates and poorer seedling vigour could have a detrimental effect on sustainable forest management of pedunculate oak stands in Croatia.

The aims of this study were to establish OLB phenology and quantify the impact of OLB infestation on oak seed in Croatian oak seed orchards and forest stands in the early stages of OLB infestation.

Material and Methods

From February 2017 until November 2017, OLB phenology was monitored in the 'Petkovac' clonal seed orchard and weekly ocular observations of the developing stages were made. The results of monitoring are given in Table 2.
Acorns were collected from two localities in the forest administration of Vinkovci and two localities in the forest administration of Bjelovar. The first and the most severe OLB infestation occurred in the area of Vinkovci, while the latest and the least heavy infestation occurred in the area of Bjelovar. Due to low OLB infestation levels, the latter is considered a control plot. The 'Petkovac' clonal seed orchard (HR-QRO-SP-121/300) and the 'Vrbanjske Šume' seed stand (HR-QRO-SS-121/108) in Vinkovci Forest Administration, as well as the 'Pleščice' clonal seed orchard (HR QRO SP 123/299) and 'Bolčanski - Žabljački Lug' seed stand (HR QRO SS 123/133) in Bjelovar Forest Administration were investigated and acorns were collected during October 2017 (Table 1). The germination test was prolonged for 7 days for samples from the clonal seed orchard 'Petkovac' (HR-QRO-SP-121/300) and seed stand 'Vrbanjske šume' (HR-QRO-SS-121/108).

Table 1: Basic data on investigated pedunculate oak clonal seed orchards and seed stands

Label	Location/Name	Register number/ Type of FRM	Latitude	Longitude	Altitude (m)	Date of collection
53	FA Bjelovar, FO Čazma, Pleščice	HR-QRO-SP-123/299 Clonal seed orchard	45.44N - 45.45N	16.35E - 16.35E	100	October 4, 2017
54	FA Bjelovar, FO Čazma, Bolčanski - Žabljački lug	HR-QRO-SS-123/133 Seed stand	44.55N - 44.55N	16.42E - 16.42E	109-110	October 4, 2017
55	FA Vinkovci , FO Otok, Petkovac	HR-QRO-SP-121/300 Clonal seed orchard	45.08N - 45.08N	18.51E - 18.52E	86-90	October 17, 2017
56	FA Vinkovci, FO Otok, Vrbanjske šume	HR-QRO-SS-121/108 Seed stand	45.01N - 45.01N	18.58E - 18.59E	78-80	October 17, 2017

The most important seed properties tested on acorns included germination capacity, 1000-seed weight and seed moisture content. Seed quality was tested according to the ISTA methodology: germination capacity was determined using the method 'The Germination Test', Chapter 5, 1000-seed weight was determined with the method 'Weight Determination', Chapter 10, and moisture content was determined by means of the method 'Determination of Moisture Content', Chapter 9 of the ISTA manual [8].
In the present study, multivariate analysis of variance was conducted to identify the differences between 1000-seed weight (g) and major properties of laboratory seed germination testing from different locations. Duncan's New Multiple Range Test (MRT) was used to determine differences in seedlings and seeds from different locations. The data were statistically analyzed in STATISTICA 7.1 statistics software [9].

Results and Discussion

After the seasonal activity of OLB in Vinkovci Forest Administration (Table 2), the results of seed quality for different localities were as follows (Table 3 and Table 4).

Table 2: Oak lace bug phenology
Legend**:** + - adults, . - eggs, 0 - larva (nymphs), x - possible third generation

year	month											
1.	I.	II.	III.	IV.	V.	VI.	x VII.	x VIII.	x IX.	X.	XI.	XII.
	+	+	+	+.	+ . 0	+ . 0	+ . 0	+ . 0	+ . 0	+	+	+
2.	+	+	+									

Table 3: 1000-seed weight (g) analysis from different locations

Locality	1000-seed weight (g)	Variance	Standard deviation	Variability coefficient	Mean value	F	Sig.
Pleščice	5800 a	54.857	7.407	2.554	290	469,25	0,0000*
Bolčanski - Žabljački lug	4558 b	35.554	5.963	2.617	227.875		
Petkovac	3495 c	46.643	6.756	3.866	174.75		
Vrbanjske šume	5275 d	33.643	5.8	2.199	263.75		

*Significant at 0.05 level

1000-seed weight (TSW) showed statistical significance for all four sample localities.

Table 4: Results of laboratory seed germination testing from different localities

Locality	Germination energy (%)	Normal seedlings (%)	Abnormal seedlings (%)	Fresh ungerminated seed (%)	Dead seed (%)	Moisture content (%)
Pleščice	6,50 a	32,50 a	9,50 a	0 a	58 a	52,8
Bolčanski - Žabljački lug	1 b	19 b	9,50 a	0 a	71,5 b	56,6
Petkovac	2 b	55,50 c	12,5 a	5 b	27 c	43,6
Vrbanjske šume	3 b	83,50 d	5 a	1,5 a	10 d	42,6
F	5,3922	167,49	0,77929	17,80000	101,3200	
Sig.	0,013933*	0,0000*	n. s.	0,000103*	0,0000*	

*Significant at 0.05 level

Germination tests were conducted during the period of 28 days. For samples from the clonal seed orchard ‘Petkovac’ (HR-QRO-SP-121/300) and seed stand 'Vrbanjske šume' (HR-QRO-SS-121/108) the test was prolonged for 7 more days. At the end of the germination test, the seedlings were smaller than usual. Seed from the clonal seed orchard QRO-SP-123/299 Pleščice showed statistically significant difference in germination energy (after 14 days) compared to the other three localities. The percentage of normal seedlings which represents laboratory seed germination was statistically different for all the localities. No statistical difference was established for abnormal seedlings in any of the four localities despite the high percentage of abnormal seedlings. Acceptable percentage of abnormal seedlings for pedunculate oak should not exceed 5%. The percentage of fresh ungerminated seed from the clonal seed orchard ‘Petkovac’ (HR-QRO-SP-121/300) is statistically different from that in the other three localities. Also, 1000-seed weight in ‘Petkovac’ locality indicates that the seed is very small [10,11]. All the localities show a high percentage of dead seed and are statistically different respectively.

An important factor for assessing the impact of *C. arcuata* on seed production and forest regeneration is acorn quality. The most important seed properties are germination rate and germination capacity, moisture content and 1000-seed weight. In the investigated area the quality of acorns was high and varied from year to year [10-12]. Germination capacity was significantly influenced by the presence of pests (fungi, insects and bacteriosis) and by mast years [13]. Poor seed quality with high levels of dead seeds in Bjelovar Forest Administration (Table 4) made it impossible to establish a correlation between OLB infestation and seed quality.

Conclusions

In all the localities under study, slower germination is the consequence of very low germination energy. The number of abnormal seedlings is higher than usual because the quality of acorns is bad. Bad seed quality could be attributed to inadequate handling of acorns during collection and to poor mast year. High amount of dead seed can also be attributed to the impact of other insects (e.g. genus Balaninus) and pathogens. This is the reason why it was impossible to establish a pattern between OLB infested localities and control localities. OLB phenology shown in this paper emphasizes the activity of OLB and the damage it inflicts on oak leaves during the vegetation period and acorn fructification. Also, inadequate manipulation with seed before sowing makes it hard to ascertain whether seed germination, growth and quality were the consequence of oak lace bug infestation. The real impact of OLB in the area of infestation would be confirmed by lower amounts of dead seed from Bjelovar seed stand and clonal seed orchard ‘Pleščice’. Future research should be conducted on seeds that are manipulated in the same manner. The processes and chemism in acorns could also be correlated with the possible impact of OLB. Since the OLB almost definitely affects tree vitality, the possible impact of this bug on fructification and acorn development should also be investigated.

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