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# Mirjana OCOKOLJIĆ, Dragica VILOTIĆ, Đurđa STOJIČIĆ<sup>1</sup>

## SIGNIFICANCE OF THE ABSENCE OF SEED DORMANCY IN WHITE BARK PINE GENE POOL CONSERVATION

#### SUMMARY

The absence of seed dormancy, i.e. the development of seedlings from embryos without dormancy, occurs most frequently in the Paleotropic, Neotropic and Australian floristic regions. In woody plants of the temperate region, this character is most often latent and occurs in unfavourable environmental conditions. The absence of seed dormancy or vivipary is an adaptive strategy that enables the generative or vegetative reproduction of adult trees in the extreme or specific conditions of vegetation in which the rooting period is short due to the moisture regime. A group of 12 trees of white bark pine were analysed 60 years ago, a group of 8 trees 35 years ago, 5 trees 50 years ago, and 2 trees at present (2012) in the park of the Old Meteorological Observatory in Belgrade using the methods of comparative morphological and genetic-physiological analyses of seed and needle samples. The absence of seed dormancy was confirmed in the continuity of 60 years. Studies of this type have both theoretical and practical significance to the process of white bark pine breeding, as well as to the production of planting stock intended for the very steep and dry southward slopes of the Mediterranean and sub-Mediterranean mountains in Serbia, Montenegro, Herzegovina and Macedonia.

Keywords: Pinus heldreichii Christ., vivipary, breeding, limestone

### **INTRODUCTION**

Recorded and monitored over a period of 60 years, the covered appearance and viviparity of Balkan-Apennine-type white bark pine is of great scientific and practical importance. Namely, permanent and fundamental research of variability of the extreme properties of the woody plants for many years allows the use of new technologies in genetic engineering of whole genomes and chromosomes (Tucović, 1987). Listed researches contribute knowledge about genetic and physiological control of viviparity, which is determined by the laws of inheritance, population and evolution biology (mutation, the principle of the population founder, gene drift, natural selection, crossover relatives, etc.). Also, it enables continuous germination immediately after planting them in the tree and shrub taxon, thereby overcoming some types of seed dormancy.

<sup>&</sup>lt;sup>1</sup> Mirjana Ocokoljić, (corresponding author: mirjana.ocokoljic@sfb.bg.ac.rs), Dragica Vilotić, Đurđa Stojičić, University of Belgrade - Faculty of Forestry, Kneza Višeslava 1, 11030 Belgrade, Srbija (Serbia).

*Pinus heldreichii* Christ. is the tertiary relict and subendemit of the Balkan Peninsula. It is widespread in the high mountain forest zone of the Mediterranean and sub-Mediterranean mountains (up to 2000 m above sea level, very often the upper limit of forest vegetation) of Southern Italy, Bosnia and Herzegovina, Montenegro, Serbia, Macedonia, Albania, Bulgaria and Greece (Vukićević, 1996). It mainly occurs in pure or mixed stands with Molik (*Pinus pence* Griesb). It grows on steep, dry, southern exposed slopes, mostly on limestone substrate. According to the IUCN red list, white bark pine is not listed as endangered, but at the regional level can be included in the endangered, or even highly endangered, type.

Therefore, the evaluation of the genetic-physiological properties of white bark pine stems brought up in the park of the Old Meteorological Observatory on Vračar communal assembly in Belgrade, with which the occurrence of vivipary was recorded, represent the basis for further multidisciplinary research to conserve and preserve the gene pool of the species.

### MATERIAL AND METHODS

Research objects are stems of white bark pine trees in the park the old Meteorological Observatory in Belgrade. 1950<sup>th</sup> the group consisted of 12, then 8 (1977. years) and 5 (2002. year) and today (2012.) only 2 of the white bark pine tree (*Pinus heldreichii* Christ.). Belgrade is situated at 44° 49' eastern longitude width and 20° 32' north geographic latitude. Investigated site is in the central part of the town with the elevation of 132 m. Soil type is brown forest but mostly anthropogenic soil. Habitat belongs to the forest community of Italian oak and Turkey oak with a broom, a variant of hornbeam (*Ass. Quercetum farnetto-cerris aculeatetosum* Job.). The climate is changed moderately continental, with the annual air temperature average of 12 °C (for thirty year period since 1970 until 2000) and with average annual rainfall of 694 mm (according to data of the Hydro meteorological Service of Serbia).

Selected white bark pine trees were compared with the cultures of white bark pine stands in Serbia, with the use of comparative morphological analysis method of natural and cultivated populations of white bark pine and the genetic and physiological analysis methods of samples of seeds of the white bark pine needles and in the laboratory.

Tree heights were measured Blume Leissovim altimeter, a standard diameter with the pelvic diameters. Rating abundance yield was determined by quantification of phenological observations scoring from 0 to 5.

### **RESULTS AND DISCUSSION**

A 60 years period of monitoring white bark pine trees, confirmed the continued occurrence vivipary which contributes to complement genetic research. The hereditary nature of vivipary trees reflected in the appearance on the trees from the generation to generation, while the frequency properties affect the size of the parental population, the nature of the exchange of genetic material

within and between populations (Table 1). This phenomenon leads to the white bark pine in connection with the nature of reproduction in small populations, and the increased possibility of translating a heterozygous genotype in the homozygous state on the basis of inbreeding, the principles of the founders of a population or a random gene drift. Accidental separation of genes in small populations, it explains the differences in many traits between populations are related, but also territorially isolated which may not be due adaptation.

In the long time research on white bark pine viviparity was first discovered in 1950th by Tucović in isolated, small space, of white bark pine population at the former site of the forest of Italian oak in Belgrade. In later researches of Tucović and Stilinović (1971, 1972, 1975) on small populations in natural habitats in Serbia, was confirmed the same. Further research of Tucović and Ocokoljić (2002) confirmed the continued occurrence viviparity. Monitoring and analysis of phenomena, a decade later (2012), is implemented by a group of researchers Ocokoljić, Vilotić and Stojičić. Comparative analysis of the data confirmed that the share vivipary trees are largest in small, spatially isolated populations.

	Type of the	Cine of	, 	Number of	The probability of	
	Type of the	Size of	$\Delta qe$ in years	Number of	The probability of	
Locality	population	population	Age in years	vivipary trees	genetic kinship	
Belgrade	Culture	Very small (8 trees)	45	3	Very big	
Oplenac	Culture	Small (105 trees)	45	1	Big	
Prevalac (Kosovo)	stands	big (thousand trees)	from 30 to mere then hundred	2	Small	
Murtenica	the rest of the stand	Very small (5 trees)	50-60 (4 stables); more (1tree)	2	Very big	

Table 1 Characteristics of white bark pine populations in which vivipary trees are evidenced (Tucović and Stilinović 1975)

Recorded vivipary trees, which are discovered to date, the phenotype and growth elements, types of trees, morphology and colour of bark, twigs, needles and shorn, do not deviate from the typical for the species (Table 2).

White bark pine trees in the past decade has made gains height of 3 m, and their height was 10 to 15 m. Records indicate that the white bark pine realized twice lower height than specified for the species in natural populations (Ocokoljić and Ninić-Todorović 2003). Manifested slower growth of whitebark pine cenosis in urban ecosystems in relation to the natural habitat is expected because the analysed trees are under the influence of many environmental stress factors that lead to major changes: reducing the vitality, height, diameter and volume growth, yield, resistance and long life (Ocokoljić, 2006). The majority of cultivated trees characterize reduced overall life expectancy by at least 30%. Namely, life of trees in urban cenosis is determined by their genetic predisposition, which may be reduced several times under the influence: large areas under the curtains, a large number of tall buildings, ground and overhead installations, climatic conditions (especially temperature), anthropogenic substrate properties, as well as large amounts of aero contamination substances (Ocokoljić, 2011).

Morphometric analysis of the needle indicates that the average length of needles is at the upper limit referred to in the literature for the species, from 6 to 10 cm (Ocokoljić and Ninić-Todorović 2003). Minimum length of the needle is 8.4 cm and the maximum was 11.4 cm. The average value of the length of needles for both trees was 10.1 cm.

Number of trees	Position	Age ( Years)	Height (m)	The scope of the tree (cm)	Length of the tree canopy top(m)	Diameter of the crown (m)	Rating yield	Total seeds per tree	Average seeds per cone	The vivipary appearance		
	2002.											
1.	To the Paster street, left from	72	9	81	1,84	5	2	154	36	rarely		
2.	the walking track To the Paster street, right from the walking track	72	12	95	2,87	5	1	188	58	rarely		
	2012.											
1.	To the Paster street, left from the walking track		10	132	1,95	5,3	3	212	41	rarely		
2.	To the Paster street, right from the walking track		15	179	3,15	5,5	2	234	52	often		

Table 2 Analysed characteristics of white bark pine trees grown in the park Meteorological Observatory in Belgrade

Morphological and genetic variations from typical types are observed in seedlings of vivipary seed (Figure 1). It is reflected in simultaneous appearance of cotyledons and radicle, a greater thickness and length of radicle and cotyledons, and seedlings have very slow growth.

To the woody plants of moderate areas vivipary is the latent trait, as it comes to forming on maternal plant seedlings or immediately after a decline in unfavourable environments (in the snow, the occurrence of low temperatures, etc.). Vivipary phenomenon is characterized by variability, the nature of the genetic and physiological control properties, the impact of population size or population genetic uniformity of representation of the properties, a special form of genetic variability of a series of parallel and convergent evolution. The above confirms that there is genetic control of the extreme properties of seeds and monitoring other than that required further experimental study.



Figure 1. Vivipary (top) and typical (below) seedlings of white bark pine

## CONCLUSIONS

The analysed group of 12 (1950), 8 (1977), 5 (2002) and 2 (2012) white bark pine trees in the park of the old weather station in Belgrade, is proposed for protection as a natural monument.

The study of the age of trees in the Belgrade urban census revealed a small number of trees older than 100 years, making it important to point out the white bark pine tree age of 82 years. They are mostly trees that had shown a broad ecological range and the possibility of existence in the urban environment. For such rare individuals of long-life there arises a need to experimentally confirm aspects of their reproductive function in order to multiply and preserve the archives and establish clonal plantations.

The status of white bark pine seed grown trees, which are extremely rare in nature, in the 21st century is studied from theoretical and applied viewpoints, necessary for the proper use of the extreme properties of the trees and protection and preservation of the gene pool of the species. The vivipary property (absence of dormancy) of white bark pine is genetically only partly explored. The hereditary nature of the analysed trees is reflected in the appearance of vivipary at the level of small cultivated populations from generation to generation.

Research of this type is important to the process of breeding and gene pool conservation of white bark pine, and to the production of planting material for the very steep, dry limestone slopes of the Mediterranean and sub-Mediterranean mountains of Serbia, Montenegro, Herzegovina and Macedonia. Also, it will allow the mapping of genes, fixation of heterosis and the elimination of some types of white bark pine seed dormancy.

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# SAŽETAK

Odsustvo mirovanja semena odnosno razvoj klijavaca iz embriona bez mirovanja se najčešće javlja u paleotropskoj, neotropskoj i australijskoj florističkoj oblasti. Kod drvenastih biljaka umerenog područja ovo svojstvo je najčešće latentno i dešava se u nepovoljnim uslovima sredine. Odsustvo mirovanja semena ili viviparija predstavlja adaptivnu strategiju kojom se obezbedjuje generativno ili vegetativno razmnožavanje odraslih stabala u ekstremnim ili specifičnim uslovima vegetacije u kojima je zbog režima vlaženja kratak period zakorenjavanja.

Stoga se u radu metodama uporedno-morfološke i genetičko-fiziološke analize uzoraka semena i četina analizira grupa od 12 stabala pre 60 godina, zatim 8 pre 35 godina, 5 pre 50 godina a danas od 2 stabla (2012. godine) munike u parku stare Meteorološke opservatorije, na Vračaru, u Beogradu. Potvrdjena je pojava odsustva mirovanja semena u kontinuitetu od 60 godina. Istraživanja ovog tipa imaju teorijski i praktični značaj u procesu oplemenjivanja munike, kao i u proizvodnji namenskog sadnog materijala za vrlo strme i suve, jugu izložene, padine na mediteranskim i submediteranskim planinama Srbije, Crne Gore, Hercegovine i Makedonije.