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SEED PRODUCTIVITY *JUNIPERUS HEMISPAERICA* C. PRESL. *JUNIPERUS SABINA* L. AND THEIR PRESERVATION IN THE CRIMEA

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Data about building of reproductive structures, seeds` productivity and viability of Juniperus hemisphaerica and J. sabina in the Crimea have been given. Possible reasons of low seed production and viability in studied species have been determined.

Juniperus hemisphaerica, J. sabina, microsporophylls, microsporsngiums, pollen grains, ovules, pollination, seeds` viability.

Genus *Junipers* L. (family Cupressaceae) belongs to the gymnosperm plants. Depending on taxonomic viewpoint, there are between 50–67 species of juniper, widely distributed throughout the northern hemisphere, from the Arctic, south to tropical Africa in the Old World, and to the mountains of Central America. The number of juniper species is in dispute, with two recent studies giving very different totals, Farjon [6] accepting 52 species, and Adams [3] accepting 67 species.

There are five juniper species which wildly grow in the Crimea – *Juniperus excelsa* Bieb., *Juniperus foetidissima* Willd., *Juniperus oxycedrus* L., *Juniperus hemispaerica* C. Presl. (=*Juniperus communis* L. subsp. *hemisphaerica* (J.Presl & C.Presl) Nyman), *Juniperus sabina* L. Three of them are trees (*J. excelsa*, *J. foetidissima, J. oxycedrus*) and grow in the foothills of the Crimean Mountains or on the lower part of the slopes. The other two species (*J. hemisphaerica* and *J. sabina*) grow on the top parts of the slopes and on plateaus (900–1400 m above sea level).

Two species *J. oxycedrus* and *J. hemisphaerica* still have an unclear systematic status. R.P. Adams has found out that the two populations of *J. oxycedrus* (Greece, Spain) were different in the four data sets. The plants from Greece were appropriately recognized as a new species – *Juniperus deltoides* R. P. Adams *sp. nov*. [4]. Further investigations showed that the two species are largely allopatric with *J. deltoides* occurring from Italy eastward through Turkey into the Caucasus Mts. and Iran. *J. oxycedrus* var. *oxycedrus* appears to be largely concentrated west of Italy (France, Spain, Portugal, Morocco) [2]. For specimens from Crimean populations such analyses haven`t been made. Another species which is presented as *J. hemisphaerica* C. Presl. in a nomenclatural checklist for Ukraine [11] in Flora Europaer [7] is defined as *Juniperus communis* L. subsp. *hemisphaerica* (J.Presl & C.Presl) Nyman and R.P. Adams and

R.N. Pandey have got the RAPD data that didn`t support the recognition of *J. c.* var. *hemispherica* (from Sicily) [1]. So identity of the Crimean plants *J. hemisphaerica* needs confirmation.

All *Juniperus* species are an important component in the Crimean landscape. They play a significant role in meeting human needs while maintaining the ecological process, protecting watersheds and combating soil erosion.

A great number of scientific works is devoted to the study of treelike junipers (*J. excelsa, J. foetidissima, J. oxycedrus*) which grow along the coast. It has been shown that in nature only seeds propagation is possible for these species and number of normal seeds with mature embryos depends from the environmental conditions but even in the most favorable years they are not enough for successful natural regeneration [19]. Because of this reason and their fragmental Crimean populations *J. excelsa* has got status of "vulnerable" and *J. foetidissima* – "rare" species and they are in "The Red Book of Ukraine" [21]. In spite of it has been shown a low ability of *J. oxycedrus* for natural regeneration [20] it is still beyond the list of preserved species.

As for dwarf junipers (*J. sabina, J. hemisphaerica*) less attention was given for their study. *J. hemisphaerica* communities (with the presence of *J. sabina*) have been put to the "Green Book of Ukraine" [16] with the status "rare". It has been noticed that associations have unsatisfactory potential of natural regeneration, as far as instead of destroyed juniper plants steppe communities are formed. Dominant way for natural regeneration of these species is by seeds. That's why it's important to get information about their ability to produce viable seeds in the conditions of the Crimea.

The aim of our work was to study reproductive structures and seeds of *J. sabina* and *J. hemisphaerica* in the conditions of their natural habitats.

Materials and methods. We studied plants from the plateaus Ai-Petri, Nikitska and Yaltinska Yaila, upper Chatyr-Dag, Demerdzhi Yaila, Tyrke in the zone 900–1400 m a.s.l. and from the park of Nikitsky Botanical Gardens (NBG) (about 300 m a.s.l.). The climate of the plateaus is moderate with the average air temperature +3,5–+6,5 ^{o}C. The warmest month is July (the average air temp. +12,5–+17 ^{o}C), the coldest month is January (the average air temperature –12,5 – -17 ^{o}C). Winter lasts for 105–114 days with strong snow cover for 80-105 days. Year rainfalls are 720–960 mm.

The climate of the southern coast of the Crimea, where NBG is situated, is Mediterranean. The average air temperature is +10,8–+14 °C, the warmest month is July (the average air temperature +20,2–+27,4 ^{o}C), the coldest month is February (the average air temperature -2,5 – +7,3 ^{o}C), year rainfalls 582–621 mm.

Juniperus sabina is dioeciously with separate male and female plants. It is a shrub variable in shape, up to 1 m tall. The leaves are of two forms, juvenile needle-like leaves 5–8 mm long and adult scale-leaves 1–4 mm long on slender shoots 0,8–1 mm thick. Juvenile leaves are found on seedlings and on shaded shoots low in the crown of mature shrubs

Juniperus hemisphaerica (=Juniperus communis subs. hemisphaerica) is a low spreading shrub, 50–70 sm tall, near round shaped, up to 2 m in diameter. It has needle-like leaves in whorls of three; they are green, sessile, keeled and

with a single broad white stomatal band on the upper side, divided by a green line towards the base, 3–14 mm long. It is dioeciously, with male and female cones on separate plants, wind pollinated.

Number of seeds and estimating their viability was carried out in 50 fruits per plant. Seed viability was confirmed by splitting the seeds to check if the embryo was aborted or viable.

Results and discussion. Male reproductive structures of both species differentiate in June. Male cones of *J. hemisphaerica* consists of 8–12 microsporophylls on the abaxial part of each 2–6 microsporangiums are present. The number of microsporophyll and microsporangiums highly depends from growth conditions. On the plateau Tyrke with the most extreme conditions we observed male cones with undeveloped microsporophylls but fully developed microsporangiums. In *J. sabina* male cones consist of 10–14 microsporophylls with 2–4 microsporangiums each. In this species microsporophylls and microsporangiums develop in the year of differentiation (in autumn) and microspores` mother cells form. Meiosis takes place next year, at the end of February – the beginning of March (on the coast) or in April (on the plateaus). In *J. hemisphaerica* only microsporophylls develop in the year of differentiation and microsporangiums start their development next year, in March. Meiosis takes place at the end of March – the first part of April when the air temperature can vary greatly. Seppä *et al*. [10] concluded that the optimum temperature for *J. communis* pollen production was +1,5 °C. In both species pollen grains are one celled and wingless. Pollen viability is 74–95 %. Development of microsporophylls, microsporangiums and meosis are asynchronous so even in the most unfavorable conditions enough number of viable pollen grains is formed.

Female cones in both species differentiate in July. In *J. sabina* they consist of 4–6 fleshy scales (mostly 4) in the axil of each 1 or 2 ovules develop. Our study has shown that 29 % of scales have 1 ovule, 20 % – 2 ovules and 51 % – no ovules and there is no correlation between this index and the place of growth. At the time of pollination female cones contain 1–4 ovules. In *J. hemisphaerica* female cones mostly have 3 fleshy scales, but about 30 % have 6 scales in two rows. Ovules were observed only in the axils of the inner row scales. One ovule forms in the axil of the scale. To the time of pollination female cone, as a rule, contain 3 ovules. P.A. Thomas et al. [15] reported that in England *J. communis* subs. *communis* female strobili normally contain three ovules and thus produce 1–3 seeds (although up to 6 is possible). We haven`t observed female cones with 6 ovules or seeds.

J. sabina pollination occurs at the end of April (on the coast) – May (on the plateaus) and *J. hemisphaerica* – in May (nearly the same terms on the coast and on the plateaus). Pollination is anemophilous with pollen being caught in a pollination drop of sugary fluid secreted by the nucellus. As it has been shown for other Juniper species the appearance of pollination drop highly depends of environmental conditions, such as air temperature and humidity [18]. Mugnaini *et al*. [5] showed that in *J. communis* it appears in the morning and remains in place for up to 7–10 days if pollination didn`t occur and it can reabsorbed in as little as 10–15 min following the arrival of unsuitable pollen or substitute such as talc or coal dust. They have reported that the number of pollen

grains on the surface of the pollen drop doesn`t influence the speed of its absorption. But previous studies showed that the time needed for pollen drop absorption after arrival of pollen grains is in direct correlation with their number [19, 18]. So this question is still open for discussion.

Pollination process is one of the most vulnerable stages in reproduction cycle for many plants. It`s also right for *J. hemisphaerica* and *J. sabina*, especially for the last. The main problem is the asynchronous development of male and female reproductive structures. In *J. hemisphaerica* the main development of ovules and microsporangiums takes place in the year of pollination (March–May) and as a rule finishes at the same time. In *J. sabina* male cones mostly develop in the year of differentiation (only meiosis occurs the next year) but the main stages of ovules` development take place in spring before pollination. At that time the air temperature can rise quickly and as male reproductive structures are more sensible for these changes they mature earlier than females. Microsporangiums open, pollen grains fly but ovules are not ready to get them, so pollination is unsuccessful. If none of the ovules is pollinated, the cone is usually aborted. On the plateau Tyrke we observed that only 5,6 % of female cones *J. sabina* continue to develop after pollination and the other aborted. At the same conditions only 20 % of female cones *J. hemisphaerica* aborted. García [8] in the Sierra Nevada of Spain found an average of 56±2,6 % (SE, *n* = 75, range 5–91 %) of cones *J. communis* aborted. If any ovule in the cone had been successfully pollinated the cone continue to develop. Seed development in *J. sabina* takes about 17–18 months after pollination and they mature in September (on the coast), October – November (on the plateaus). In *J. hemisphaerica* cones are produced annually but it takes 3 years to mature.

Mature fruits of both species are black with a blue bloom. The size (length × width) of mature fruits is - *J. hemisphaerica* 6,9±0,1 × 5,3±0,2 mm, for *J. sabina* 5,5±0,2 × 4,6±0,15 mm. The average seeds number in *J. sabina* is 2,1 per cone and in *J. hemisphaerica* – 2,2 per cone. In a European-wide survey, García *et al*. [9] found the highest number of seeds per cone towards the southern end of *J. communis* range in the Iberian Peninsula; an average of around 2,3 seeds per cone compared to as low as around 1,4 in the Saian Mountains in Siberia within the central range of the species. And for Italy it has been reported 2·23 ± 0·10 *vs* 2·33 ± 0·11 seeds per fruit for *J. sabina* and 2·70 ± 0·05 *vs* 2·69 ± 0·14 for *J. hemisphaerica* [17].

Seed viability for *J. hemisphaerica* varies from 39,1 % (Nikitska Yaila, Demerdzhi) to 72,2 % (Aipetrinska Yaila, Yaltinska Yaila) and in the park it was 42,9 %. *J. sabina* has lower seed viability which varies from nearly zero (Tyrke) to 23,8 % in the park. In Italy the percentage of viable seeds for *J. sabina* 54·1 ± 6·9 *vs* 53·1 ± 2·6 and *J. hemisphaerica* 51·3 ± 8·8 *vs* 49·7 ± 5·5 [17]. García *et al*. [9] reported that percentage of filled seeds (and therefore presumably viable) for *J. communis* averaged less than 11 % in Iberian regions, with a lowest value of 1,4 %, compared to 80 % in the Saian Mountains. They showed that the high proportion of empty seeds is undoubtedly due to sub-optimal climatic conditions affecting pollination and fertilization but suggested that it could also be due to inbreeding depression amongst the naturally fragmented (and now increasingly so) populations. The same conclusion has been postulated with Ruguzov *et al*.

[19] for tree-like Crimean juniper species. They postulated that in successful populations close breeding doesn`t play a significant role but in fragmented populations inbreeding is regular and its results are accumulation of genetic load and inbreeding depression. The reason has been postulated by several workers, who considered that production of seeds and seed fertility decrease dramatically in the populations of most tree species in the northern regions due to stressful climate [12, 13].

Conclusions

Our investigations have shown that in the reproductive cycle of *J. hemisphaerica* and *J. sabina* the most vulnerable stage is pollination success of which depends from weather conditions and populations` status. Seed productivity in both species is quiet low in the Crimea. Seeds` viability for *J. sabina* is very low and for *J. hemisphaerica* it varies greatly. We suggest that the number of viable seeds which produced annually is enough for keeping populations in the same status under unchangeable environmental conditions. But under increasing of anthropogenic influence and climate changes wild populations of these species will decrease. The problem is that juniper plants have long living terms and the result of low (or absent) regeneration we`ll notice only in 25–50 years. That`s why it`s necessary to carry out detailed investigations of all wild Crimean populations of *J. sabina* and *J. hemisphaerica* and to use active management to encourage natural regeneration and to guaranty preservation of these species in the Crimea.

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Приведены данные о строении репродуктивных структур, семенной продуктивности и жизнеспособности семян Juniperus hemisphaerica и J.sabina в Крыму. Определены возможные причины незначительного количества и низкой жизнеспособности семян изученных видов.

Juniperus hemisphaerica, J. sabina, микроспорофиллы, микроспорангии, пыльцевые зерна, семязачатки, опыление, жизнеспособность семян.

Наведено дані щодо будови репродуктивних структур, насіннєвої продуктивності та життєздатності насіння Juniperus hemisphaerica та J. sabina в умовах Криму. Визначено можливі причини незначної кількості та низької життєздатності насіння вивчених видів.

Juniperus hemisphaerica, J. sabina, мікроспорофіли, мікроспорангії, пилкові зерна, насіннєві зачатки, запилення, життєздатність насіння.