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SURVEY OF SEED PROPAGATION OF OPUNTIA HUMIFUSA (RAFINESQUE) RAFINESQUE (CACTACEAE) EX SITU

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Abstract: *Opuntia humifusa* (Rafinesque) Rafinesque belongs to the anthropophyte flora of Bulgaria. The germination properties of *O. humifusa* Rafinesque were tested *ex situ* on fresh and stored seeds with two different soil substrates: 1) taken directly from the location (A) and 2) mixture of compost, sand and gravel in 1:1:1 ratio (B, C, D). The experiments were carried out in 2002 and 2007. Germination registered in 2007 of fresh and stored seeds exceeded several times that recorded in 2002. The stratification of seeds from crops 2002 and 2007, as well the extremely high temperatures during 2007 (above 30°) most probably favoured not only the high germination rate but also the rapid germination of the seeds (12 days versus 23 days in 2002 tests). The germination rate of refrigerated seeds exceeded between the rates recorded for the same fresh seeds 5 and 19 times. The results indicate that storage at low positive temperatures encourages the germinating process. Seedlings growth is slow, but steady and the seedlings are well balanced. The first pad of the seedlings occurs 30 days after germination and three months after sowing their average height is 2,7 cm (max 3,2; min 0,5 cm).

Key words: *Opuntia humifusa*, propagation *ex situ*, seed properties

A. ТАШЕВ, ЗЛ. КАБАТЛИЙСКА, Лесотехнически университет, 1756 София. ПРОУЧВАНЕ ВЪРХУ СЕМЕННОТО РАЗМНОЖАВАНЕ EX SITU ПРИ OPUNTIA HUMIFUSA (RAFINESQUE) RAFINESQUE (CACTACEAE)

Резюме: *Opuntia humifusa* (Rafinesque) Rafinesque е вид от антропофитната флора на България. Направено е проучване *ex situ* на посевните качества на свежо събрани и съхранени семена на *O. humifusa* Rafinesque при два различни почвени събстрата: 1) взет директно от местонаходището (A) и 2) смес от компост, пясък и гребен керамзит в съотношение 1:1:1 (B, C, D). Опитите са изведени през 2002 и 2007 г. Кълняемостта, установена през 2007 г. на свежи и съхранени семена превишава неколkokратно тази, установена през 2002 г. Стратификацията на семената от реколти 2002 и 2007 г., както и екстремно високите температури през 2007 г. (над 30° C) са вероятна причина не само за високия процент на кълняемост, но също така и за бързото покълване (12 дни срещу 23 дни в тестовите през 2002 г.). Процентът на кълняемост на съхранените в хладилник семена превишава този, регистриран при свежи семена между 5 и 19 пъти. Резултатите показват, че съхранение при ниски положителни температури ускорява кълняемостта. Растежът на семеначетата е бавен, но стабилен като младите растения са добре развити. Първият стъблен сегмент се появява 30 дни след покълване и три месеца след засяване средната му височина е 2,7 cm (max 3,2; min 0,5 cm).

Ключови думи: *Opuntia humifusa*,

A lot of the *Opuntia* Mill. (Cactaceae) representatives, including more than 200 species originating from North and South America had been conveyed and are still conveying in different parts of the world, characterized by warm and temperate climate. They are appreciated as ornamentals and regarded for restoration of disturbed sites. Some *Opuntia* species

resistant to moderately low temperatures are planted outdoors in parks and gardens, usually as ornamentals. Almost everywhere, where the climate favours their growth, they enlarge the clumps by layering or seed propagation (Jordanov, 1970). In this way they become part of the anthropophytic flora of a certain area.

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The following species have been introduced in Bulgaria as ornamentals: *Opuntia tortispina* Engelm., *O. ficus-indica* (L.) Mill., *O. vulgaris* Mill., *O. procumbens* Engelm. and *O. compressa* (Salisb.) Macbr. These species are cultivated along the Black Sea coast. *Opuntia tortispina* and *O. Compressa* escaped from cultivation and grow on the Zmiyski ostrov (The Snake Island), and the other species are cultivated in the Botanical Garden – Balchik (*O. ficus-indica*, *O. vulgaris*, *O. procumbens*). Some species were planted in the parks Krichim and Euxinograd (Jordanov, 1970).

According to Delipavlov et al. (2003) *O. compressa* is wild grown in localities near Chirpan, Plovdiv, Harmanly and Jambol – on rigid sand hills and rocks. Assyov et al. (2006) listed the following species as naturalized and belonging to the adventive flora of Bulgaria: *O. tortispina* (South Black Sea coast) and *O. vulgaris* (Black Sea coast, Tundja Hill plane and South Strouma Valley). There are no surveys on the communities of *Opuntia* species in the Bulgarian botanical literature.

O. humifusa is native to North-eastern Great Plains of North America. It is characterized by a good winter hardiness and grows reaches southern Ontario (Canada) northward. It is naturalized elsewhere in north America, South and Central Europe (Austria, Bulgaria Corsica, Croatia, Greece, Herzegovina, Italy, Serbia), Argentina; China; Ethiopia; Haiti; Mauritius; South Africa; It is considered noxious in Australia and Tasmania. It occurs commonly in rocky open glades, dry rocky or sandy grasslands, sandy and gravelly washes of valleys along streams and sand dunes.

The natural propagation of the species is mainly by seeds, although sometimes the vegetative propagation dominates. In moderate climate *Opuntia humifusa* plants are flowering from May to July. The fruits ripe 2–3 month after inflorescence and remain on the plants till next year. The individuals of *Opuntia* are obligate heliophytes and at shady sites are replaced by other species (Benson et al., 1982). In milder climate (in Florida) the plants are flowering throughout all the year (Wunderlin and Richard, 1998).

At extreme conditions plants of *Opuntia humifusa* survive by offsprings from the stem base and from rooted pads, when pressed closely to the ground. The prickly pear colonize post-fire areas by outer sources of seeds (cached seeds by small mammals) or inner source – by seeds, survived the fire (Bunting & al. 1980, Thomas 1991).

Opuntia has the biggest seeds among the Cactaceae family. They reach 3–5 mm and keep germination capacity within 1 to 7–9 years. The seeds of the species of interes are disc-like with hard, regular edges, flattened, rounded, about 4.5 mm in diameter (Benson, 1982). In natural conditions they are spread by birds and a variety of rodents including rabbits, wood rats, prairie dogs, mice and ground squirrels. (Timmons, 1942; Cohn et al., 1982). The germination

rate was reported to be low (Cook, 1942). In laboratory tests, it was found that eastern prickly-pear seeds collected from rabbit scats had higher germination rate than seeds collected from unconsumed fruits. (Birkhead et al., 2005; Cohn et al., 1982; Loucks et al., 1985).

Seeds collected from the scats required an average germination time of 41 days and had germination rate of 25%. Seeds collected from non-consumed fruits required 71 days and germinated at a rate of 18%. (Cook, 1942). The germination problem is not due to the seed cover hardness as it was considered, although pre-sowing treatment improves the seed germination (Orozco-Segovia et al., 2007). Important factors for germination are high humidity and high temperatures (Racine et al., 1974, Orozco-Segovia et al., 2007). Udalova et al. (1977) outlined that the optimum temperature for germination is between 27–35° C and the following soil mixtures are recommended: mould-sand-charcoal in 1:0.5:0.5 ratio and mould, turf, sand and crunched bricks in 1:1:2:1 ratio. Seeds are best sown outdoors immediately after collection without discarding the pulp. Pre-sowing treatment includes only air-drying for a few hours, and storage in sealed, refrigerated containers (Anthony, 1954).

It was found that during the process of cultivation of *Opuntia* the seedlings growth is slow but at the same time it is the fastest among the cacti (Gordon 1978). The seedlings reveal good resistance to drought and the hazard factor for surviving *in situ* is supposed to be the rodents, but not low humidity. (Racine & al. 1974, Nieddu et al 1997). Seed propagation in nature is complicated and depends on the population of rodents, that chase the seeds (Vila et al., 1999). Some observations showed that *Opuntia humifusa* individuals are supposed to be spread widely by intensive grazing (Loucks et al., 1985).

No studies of propagation of *Opuntia* by seeds of local source were found in the Bulgarian botanical literature. Therefore, the aim of the present study was to test seed germination of *Opuntia humifusa ex situ* and the potential of propagation by seeds collected in the surveyed localities.

MATERIAL AND METHODS

Seeds were collected on 4 and 17 May 2002 and 29 of April 2007 from locality of Harmanly region (Tashev, in press). They were sown immediately after gathering. In order to repeat the experiment, part of 2002 seed crop was stored for 5 years in refrigerator at 0–5° C.

The experiment was conducted in greenhouse in the following variants of soil mixture and seed type (fresh and stored seeds):

- Variant A: 5 x 50 6p. Yield 2002. Sowing 29.05.2002. Genuine soil from the natural habitat in Harmanly



Цветовете на *Opuntia humifusa*/*Opuntia humifusa* in blossom



Индивид на *Opuntia humifusa* с плод
Opuntia humifusa specimen with fruit



Характерна туфа на *O. humifusa* с плодове
A typical cluster of *O. humifusa* with fruits



Общ изглед на част от находището с две туфи
Common view of the locality with two clusters of *O. humifusa*

Находище на *Opuntia humifusa*, 16.04.2003 г. Тракийска низина, околности на град Харманли, местност „Приказките“
Locality of *Opuntia humifusa*, 16.04.2003 Tracian lowlands, the suburbs of Harmanly, near the site 'Prikazkite'

- Variant B: 5 x 50 бр. Yield 2002. Sowing 29.05.2002. Soil mixture: sand, disinfected with $KMnO_4$ aggregates < 0.5 mm; crunched bricks, aggregates < 1 cm and compost in 1:1:1 ratio.

- Variant C: 1 x 50 бр. Yield 2002. Sowing 21.04.2007. Soil mixture same to variant B

- Variant D: 4 x 50 бр. Yield 2007. Sowing 21.04.2007. Soil mixture same to variant B

The air-dried seeds were sown at 6 mm depth (1.5 x the seed diameter) in flat vessels with diameter/height 13/4 cm. The air humidity varied between 55 and 80%. The plants were watered once per week with 200 ml water per vessel.

The values of air-temperatures during the experiment of both years were measured with max/min thermometer and varied as follows: in 2002: 18–39° C and 2007 – 13–38° C with record of unspecific high May temperatures.

RESULTS AND DISCUSSION

The results obtained for the germination properties *ex situ* are shown in table 1 and on fig. 1

Таблица 1/ Table 1

Soil germination of fresh seeds of *Opuntia humifusa* – crop 2002

Почвена кълняемост на свежо събрани семена от *Opuntia humifusa* – реколта 2002

Date of sowing Дата на засяване	Variant A (number of germinated seeds/ percentage) Вариант А (брой покълнали семена/процент)				
	I	II	III	IV	V
29.05.2002	1	–	–	–	–
20.06.2002	1	–	–	–	–
02.07.2002	6	2	4	3	2
11.07.2002	6/12%	7/14%	11/22%	8/16%	8/16%
	Variant B (number of germinated seeds/ percentage)/Вариант В (брой покълнали семена/процент)				
29.05.2002	I	II	III	IV	V
20.06.2002	1	–	–	–	–
02.07.2002	2	1	–	1	2
11.07.2002	4/8%	2/4%	–	3/6%	5/10%

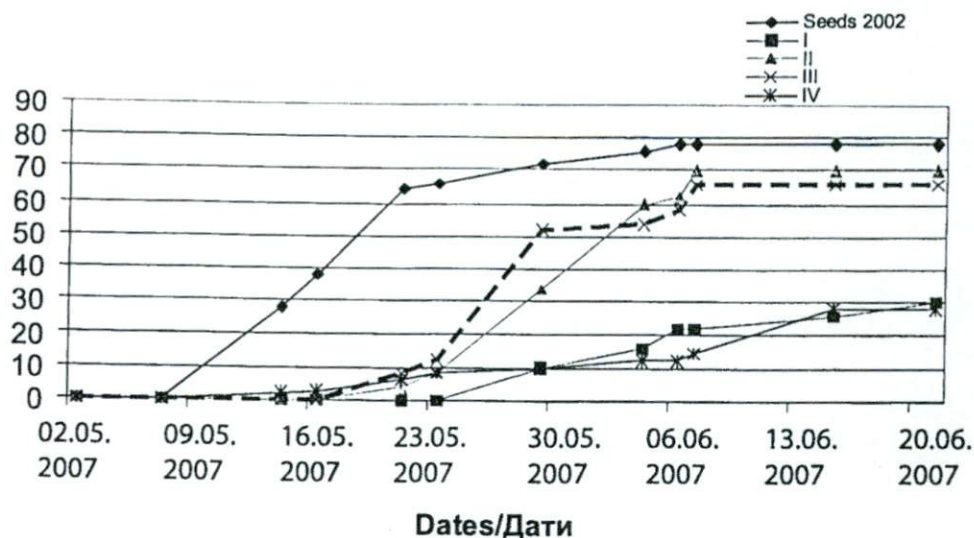


Fig. 1. Germination of seeds of *Opuntia humifusa* (Harmanly locality)
Фиг.1. Покълване на семена от *Opuntia humifusa* (находище Харманли)

The absolute mass of seeds recorded was between 26 and 29 g for 1000 seeds. The seed number per fruit varied between 9 and 31 (average 20.2 seeds/per fruits).

It is obvious that germination rate registered in 2007 of fresh and stored seeds exceeded several times that recorded in 2002. The variants can be classified as follows in ascending order: B, A, D and C. A certain explanation of that fact is the pre-treatment of seeds applied, as the extremely high temperatures during 2007 testing period (above 30° C in May) favoured not only the high germination rate but also the rapid germination of the seeds (12 days versus 23 days in 2002 tests).

A definite result was obtained in Variant C (stored seeds), with germinated seeds exceeding between 5 and 19 times the rate, recorded for the same fresh seeds. The observed period of germination is the shortest for the whole experiment: 23 days. The registered slow growth rate of seedlings coincides with the results, reported in the literature (Vila et al. 1999). Measurements during 2007 test showed that the first

pad of the seedlings occurred between the first and second month after germination, and three months after sowing their average height was 2.7 cm (max 3.2/min 0.5 cm). The seedlings demonstrated high drought tolerance.

CONCLUSIONS

Opuntia humifusa seeds gathered from Harmanly locality are fertile, and the germination of the fresh seeds is at least 5 times lower than the germination, recorded after refrigerating for 5 years. The germination rate in soil from the locality is between 12 and 22 % higher than in the artificial substrate. This confirms that the propagation of the species in the locality by seeds is possible.

The germination of stratified seeds (for 5 years) exceeds between 5 and 19 times the germination of the fresh-gathered seeds.

The cultivated seedlings demonstrate high tolerance to drought, but slow growth. The drought resistance encourages *in situ* cultivation but with good protection of the juvenile plants from animals and trampling down.

Stratification of seed is recommended in seed propagation of *Opuntia*, even when fresh seeds are available, because it simulates the natural winter conditions.

The results of seed germination tests of *Opuntia humifusa ex situ* confirm the hypothesis that the species in natural conditions in the localities is propagated by seeds.

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