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REPRODUCTION AND PROPAGATION OF SOME RARE SPECIES OF THE CRIMEAN FLORA

SUMMARY

The problem of conservation of a biological diversity of the biosphere, including its plant component, is currently very relevant and this is especially important for rare and endangered species of plants. Such rare species, which have to be protected, grow in the Crimea are *Arbutus andrachne* L. (Ericaceae), *Pistacia mutica* Fisch et Mey. (Anacardiaceae), *Campanula talievii* Juz.(Campanulaceae), *Fumana thymifolia* (L.) Spachet Webb (Cistaceae) and *Glaucium flavum* Crantz. (Papaveraceae). As the result of the study of a reproductive biology of these species, the types of formation of their generative structures, especially antekology, seed formation and dissemination have been obtained. It is shown that the main limiting factors for an optimal development of the studied species in the conditions of a natural growth are the meteorological factors in critical phase of formation of generative structures, damage caused by large animals as well as an anthropogenic impact (plowing land, construction, industrial pollution, decoration flowers, etc.). However, conservation of the species for the most part provide: stability of formation of viable male and female generative structures; successful processes of pollination and fertilization as the results of a paired development of elements of a flower, structure of insect-pollinator and pollination mechanisms; features of a seed formation and dissemination. An autogamy and vegetative propagation can be considered as reserve means, contributing to the preservation of these species. Thus, the observed variety of tools for effective processes of reproduction of the studied species of plants indicate the potential and reliability of their systems for reproduction and resettlement, and identification of the causes of the decreasing numbers allow us to develop scientifically based methods and optimize the resumption of types (nature reserves, introduction to culture, repatriation, etc.).

Keywords: Rare species of plants, generative structures, antecology, seeds formation, dissemination.

INTRODUCTION

Currently, actively developing industry, agricultural production with the use of various chemical agents, meliorative measures lead to violations of natural landscapes and plant communities, as well as reduce the species diversity of

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plants. Therefore, problem of preserving gene pool of plant world, together with threat of climate warming, presence of ozone holes in a stratospheric layer, acid rain and accumulation of a toxic waste in the ground, is among the global environmental problems. This is especially important for rare and endangered species of plants, number of populations of which and individuals in them gradually decreases. In this regard, one of the primary environmental problems is to find out the reasons for reduction in a number of rare species and to develop scientifically based techniques for optimizing their renewal and reproduction. Due to its origin and geographical location, the Crimea has a unique flora that, according to V.N. Golubev (1996), has about 2.700 species and subspecies, and nearly 750 of them need protection. Therefore, attention of scientists is directed to a comprehensive study of rare and endangered species of the Crimean flora, as evidenced by numerous publications on the ranges of the species studied, the state of their cenopopulations, seed productivity and breeding characteristics (Kosykh, Golubev, 1983; Krainyuk, 1988; Korzhenevsky, Bagrikova, Ryff, Bondareva, 2004; Shevchenko, Kuzmina, Marco, Yaroslavtseva, 2010; Shevchenko, 2014; Shevchenko, Gafarova, 2015; and others). In this aspect, it is very important to know the reproductive biology of rare and endangered species, including the formation of generative structures, antekologii and embryonic development, which is necessary to create a general picture and assess the prospects for natural renewal of the species under study.

MATERIAL AND METHODS

The objects of these studies were 5 species of flowering plants belonging to different families and growing within the natural range in the: *Arbutus andrachne* L. (Ericaceae), *Pistacia mutica* Fisch et Mey. (Anacardiaceae), *Campanula talievii* Juz.(Campanulaceae), *Fumanopsis laevis* (Cav.) Tzvelev (Cistaceae) and *Glaucium flavum* Crantz.(Papaveraceae).

Arbutus andrachne L. is an evergreen multi-stem tree up to 12 m high. The bark is thin, smooth, dark coral in color, in June-July it cracks and flakes, revealing a young green bark, which by the end of the summer acquires a coral-red color. Leaves are leathery, glossy, ovate-oblong, entire. Tertiary relic, in the Southern Coast of the Crimea grows in the form of small groves or individual trees on dry stony and rocky slopes. In accordance with the classification of V.N. Golubev (1996), according to ecomorphological features this species can be characterized as lithophyte, mesoxerophyte, heliophyte and glycophyte.*Pistacia mutica* Fisch et Mey. - deciduous, often multi-barreled tree up to 15 m high. Leaves pinnate, consist of 3-7 leaves of elliptic form. The species is characterized as hemikserophyte, aeropedophyte, heliophyte, glycophyte. According to E.L. Kordyum and G.I. Glushchenko (1976), *Pistacia mutica* refers to monoecious, dioecious polygamous plants. Most of the Crimean populations of *P. mutica* are represented by dioecious individuals. It grows singly, in small groups or communities in large areas. *Campanula talievii* Juz. - it is a polycarpic, herbaceous, half-rooted plant up to 25-30 cm tall. According to V.N. Golubev

(1996), this is the endeme of the Crimea. Generative shoots are pubescent, numerous. Flowers are also numerous. Above-ground shoots are represented by rosettes of lanceolate leaves. An underground shoot is branched, rod-shaped, fusiform-shaped, light-white in color. The species can be characterized as aeropedophyte and calfyte with a deep rod-root system.

Fumanopsis laevis (Cav.) Tzvelev (syn. *Fumana thymifolia* (L.) Spachet Webb) this is an evergreen Mediterranean summer-winter-green dwarf shrub of loose form, 25-40 cm high. The stem is erect, branchy, with pubescent and protruding branches. The species is characterized as euxerophyte, heliophyte, glycophyte. According to N.I. Rubtsov and G.A. Kupatadze (1978), the only place where the species grows in the Crimea is the southeastern gravelly slope of Mount Koshka (Simeiz village). It grows on dry open rocky and gravelly limestone slopes, among juniper woodlands.

Glaucium flavum Crantz – it is a biennial or perennial bluish-green herbaceous plant with a height of 20-50 cm. The stem leaves are lyrate-pinnately divided, up to 30 cm long. It grows in the littoral belt on seaside sand, shell rock, pebble, less often on seaside limestone rocks, stony-gravelly slopes, clayey and marly cliffs. Xeromesophyte, heliophyte, psammopetrophyte. This species is resistant to sea spray, falling asleep with sand and gravel.

The material for embryological studies was fixed with a Carnoy solution (6: 3: 1) and Chamberlain fixative (ethyl alcohol 70% -90 parts: formalin 40% -5 parts: glacial acetic acid-5 parts). In the preparation of permanent preparations, conventional methods were used (Romeys, 1954; Pausheva, 1990). Sections of 10-12 μm in thickness were obtained using a semi-automatic rotary microtome RMD-3000. The preparations were stained with methylgrunpironin with an Alcyan blue tint (Shevchenko, Ruguzov, Efremova, 1986; Shevchenko, Chebotar, 1992) and Heidenhain hematoxylin with Alcyan blue tint (Zhinkina and Voronova, 2000). The analysis of the preparations was carried out with the help of the Enamed 2 microscope of Carl Zeiss (Jena), photographs were taken with a digital camera A 550. Anecological observations were carried out under conditions of natural growth of the species under study.

RESULTS AND DISCUSSION

In the Crimea, the species studied bloom in the spring - early summer. Thus, *A. andrachne* blooms in April-May at an average daily temperature of +7 - + 10 ° C and can be attributed to the group of medium-spring-flowering plants. Its flowers are bisexual, collected in racemose inflorescences. Androecium is represented by 10 stamens, anthers of which are free, quadrilocular, are opened, like many other representatives of the subfamily *Vaccinioidae* (Yakobson, Terekhin, 1983), with apical pores. The wall microsporangium develops centripetally, and formed consists of the epidermis, endothecium, 3 middle layers and tapetum. The wall of the mature anther is represented by the epidermis, the cells of which are filled with tannin, the fibrous endothecium and the tapetal glume with orbiculs. Mature pollen grains are meridional-3-furrow-aperture,

remain collected in tetrads, 3-cell, consisting of a vegetative cell and two spermiums. Ovary *A. andrachne* upper, 5-locular, ovule anatropic, unitegmal. The embryo sac is monosporous, formed by Polygonum-type from the halazal megaspore. The mature embryo sac is 8-nuclear, 7-cell, elongated with a somewhat enlarged micropylar part (Figure 1).

P. mutica also blossoms in April-May, according to the flowering time can be attributed to the late-spring-flowering plants. Flowers unisexual, male are represented by free stamens, anthers are 4-locular, dithecal, are opened with longitudinal slits. The wall microsporangium develops centripetally, and formed consists of the epidermis, endothecium, 1-3 middle layers and tapetum. The wall of a mature anther is represented by epidermis and fibrous endotecium, sometimes there are remnants of tapetum. Mature pollen grains are single, 2-celled, consisting of vegetative and generative cells. Ovary *P. mutica* upper, ovule crassinucellate, unitegmal, anatropic. However, as the seed develops, the ovules change their position and become campylotropic. The embryo sac is formed from the chalazal megaspore by the Polygonum-type. The mature embryonic sac is large, rounded, 8-nuclear, 7-celled. As the endosperm and embryo develop, it increases and lengthens (Figure 2).

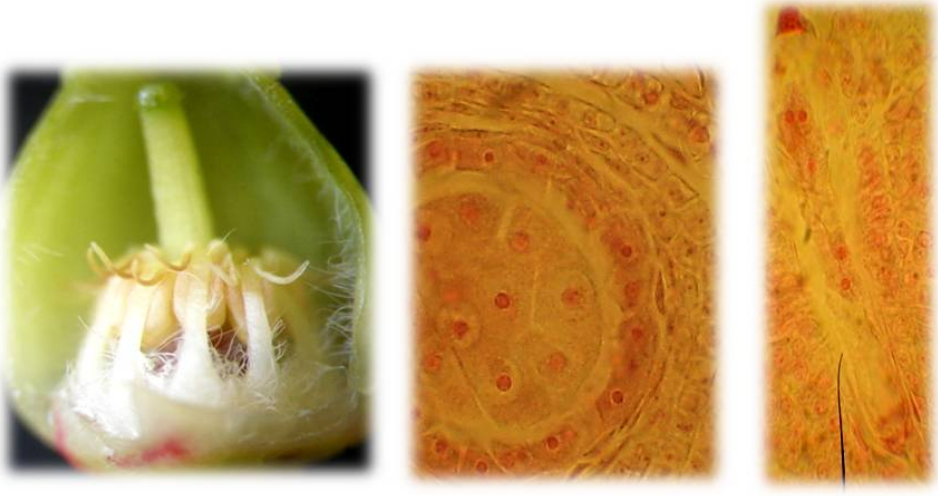

Figure 1. Fragments of *A. andrachne* generative structures

The flowering of *C. talievii* is much longer and lasts from May to August-September. Androecium represented by 5 stamens. The stamens are straight and equal, attached to the nectarean disk, located symmetrically. Stamen filaments have enlarged bases, which, when closed, formed a dome with an opening at the top. The link continues the stamen thread. Anthers are 4-locular, 2-thecal. The wall microsporangium is formed centrifugally. The formed wall of microsporangium consists of epidermis, endothecium, one middle layer and secretory tapetum. The wall of a mature anther consists of squashed cells of the

epidermis, which is covered with cuticle, and endothecium with fibrous thickenings.

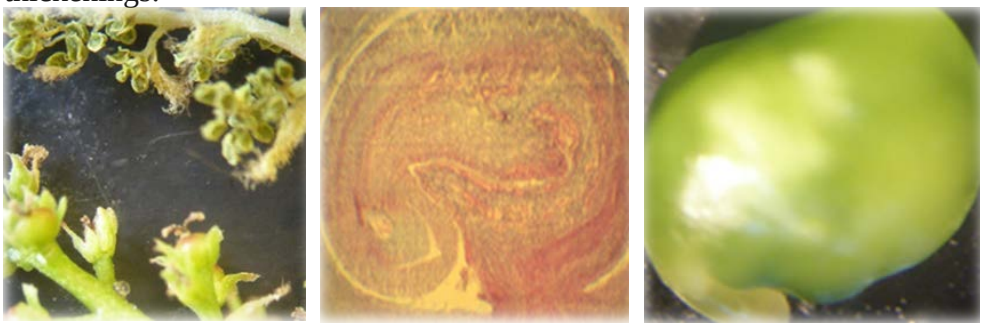

Figure 2. Some elements of *P. mutica* reproductive sphere

Endothecium is often two-row, from the side of the connective it can be two- and three-rowed. The anther is opened introrzno with the help of a longitudinal slit. Mature pollen grains 3-furrow-poral, 3-cell, spermiogenic division often passes on the stigma of the pistil.Ovary of*C. talievii* is lower, ovule is anatropic, medionucellate, unitegmal. The embryonic sac, like many other representatives of the Campanulaceae family (Korobova, Zhinkina, 1987; Zhinkina, 1995), is formed in accordance with Polygonum-type. It is 7-celled, 8-nucleus, elongated, surrounded by tabular cells of the integumental tapetum, reaching the base of the egg apparatus (Figure 3).

Figure 3. Embryo sac and fruits with seeds of *C. talievii*

F. laevis blooms for a long time, from the end of April to July. Androecium is represented by numerous stamens, located in circles. The stamens of the outer circle are sterile (without anther). Anthers 2-thecal, 4-locular, sometimes two-locular. The wall microsporangium develops centrifugally,tapetum is a derivative of the internal parietal layer.

A fully formed wall of microsporangium consists of the epidermis, endothecium, one middle layer and tapetum (Figure 4). The wall of a mature anther consists of the epidermis and endotecium, there is a tapetal glume. Mature pollen grains are two-celled, three-pore. Ovary of F. laevis upper, 3-locular, ovule ortorophic, bitegmal, crassinucellate. The embryo sac is formed by Polygonum-type, 7-cell, 8-nuclear, contains a well-differentiated egg apparatus, the polar nuclei merge before or during fertilization. Antipodes are represented by three cells that can form an antipodal complex.

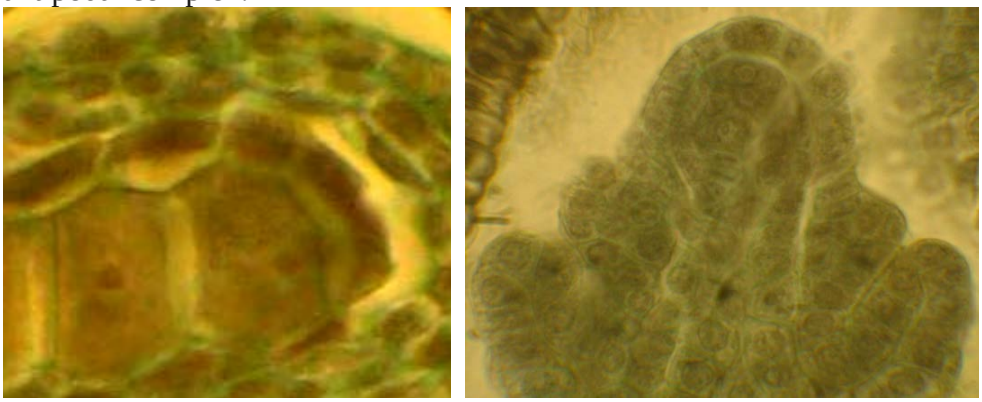

Figure 4. Some stage form of *F. laevis* generative structures

G. flavum blooms for a long time, and it is characterized by the stretching of the phenophases in time and their overlapping. Androecium is represented by numerous free stamens. Anthers 4-locular, 2-thecal.The wall microsporangium develops centripetally, and formed consists of the epidermis, endothecium, 2 middle layers and tapetum, which is a derivative of the secondary parietal layer. The wall of a mature anther consists of the flattened cells of the epidermis, fibrous endothecium and the remains of the middle layer. Mature pollen grains are 2-celled. The ovule of *G. flavum* can be described as camphilotropic, crassinucellate, bitegmal. Embryo sacs Polygonum-type, 7-celled. The egg apparatus is represented by two pear-shaped synergids and an egg. The polar nuclei are located in the central zone of the central cell, closer to the antipodes than to the egg cell. Antipodes are large, occupy more than a third of the embryo sac, often greatly expand, forming an antipodal complex and performing the function of haustorium.

One of the most important stages of the reproductive cycle is pollination, the effectiveness of which depends on the subsequent processes of fertilization and seed formation. Of the species represented, only *P. mutica* is an anemophilous plant, and the rest are entomophiles. However, the processes of pollination are provided by different devices.

Thus, in *A. andrachne* the base of the filamentous filament is strongly pubescent, the anther has excrescence, and the pollen grains have the viscin

filaments (see Figure 1) . All this contributes to the success of pollination. In *C. talievii* the device for pollination is the thickening of stamen filaments at their base. The attraction of insects in *F. laevis* and *G. flavum* is bright yellow flowers, movements of stamens. At the end of flowering flowers in the case of lack of allogamy, they can also have autogamy. A similar phenomenon is observed, for example, in some other species of the genus Fumana (Güemes, Boscaiu, 2001; Carrio et al., 2008).

The presence of viable male and female generative elements, effective pollination provides the subsequent processes of fertilization and the formation of fruits and seeds. Fruit in *A. andrachne* wrinkled orange berry, in *P. mutica* - small turquoise dry turtle, in *F. laevis* the fruit is spherical, trihedral skinny capsule, opening with three valves, in *G. flavum* the fruit is elongated (up to 25 cm) pod. *C. talievii* fruit is a drooping, trilocular, multi-seeded capsule with three rigid bases at the base, through which seeds are scattered.

Seed productivity in *F. laevis*, *G. flavum* and *C. talievii* is quite high, in *A. andrachne* the real seed productivity is only 2-3%, which is a consequence of inbred depression. In *P. mutica*, up to 80-90% of the embryos are formed, which, however, are eaten by the pistachio semeid in the development process, and there is no normal seed in this species.

Dissemination of *C. talievii* and *G. flavum* is provided by ballistochoria (ballistoanemochory and ballistozoochory), as well as epizoochory (with the direct participation of animals). After rash from the capsule, light and small seeds can be carried for considerable distances by gusts of wind (anemochory). The process of dissemination in A. andrachne and *P. mutica* is promoted by birds (ornithochory), the seeds of *F. laevis* are spread with the help of ants (myrmecochory).

CONCLUSIONS

It should be noted, that the main limiting factors in the reproduction of these species are possible extremely low air temperatures, leading to anomalous development of generative structures, the absence of pollinator insects during the flowering period, weak competitiveness, negative anthropogenic impact (trampling, bouquets, Development of territories), damage by animals. However, the formation of effective viable male and female generative structures in the studied species, effective pollination, fertilization and seed formation processes, successful dissemination show that these species have a reliable multiplicative and reproductive system, as well as the potential for species dispersal and colonization of new neighbouring erritories.

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