

Signs of Qualitative Variability of Seeds of *Lonicera Gracilipes* Var. *Glandulosa* Maxim. Under Influence with Ionizing Radiation

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Abstract. The article deals with the study of the biological characteristics of *L. gracilipes* Mig var. *glandulosa* Maxim as a new berry crop. Seeds were treated with ionizing radiation to obtain new forms of crop plants. The identified number of qualitative signs of variability in seedlings exposed to ionizing radiation allows us to preliminarily speak about the identified mutations in *L. gracilipes*. The resulting mutant forms of the species *L. gracilipes* with other biological indicators to varying degrees different from the original species make it possible in the future to isolate promising forms valuable from an economic point of view for domestic berry growing. The currently existing economic indicators of the species *L. gracilipes* in terms of frost resistance (limit -250C), fruit weight - 0.6 g, low productivity (up to 300 g from one bush), put forward specific tasks for the selection of a new rare crop, promising for its cultivation as a fast-growing berry bush.

1 Introduction

The potential of the genotype to adapt to new or changed natural conditions, the key to the prosperity of the species. All signs, features and properties of plants are conditionally divided into two groups: qualitative and quantitative. If the quantitative traits of variability require measurements in their definition, then the qualitative traits are easily distinguishable by eye and become decisive in selection selection. Qualitative signs of plant variability are signs whose development is due to the action of genes with a strong effect, or the so-called main genes. Such signs include the color and shape of flowers, fruits, leaves and other organs [1-2]. Many qualitative features are not subject to environmental conditions and remain in any conditions. This makes it possible to judge the genetic differences of organisms that differ, among other things, in terms of qualitative characteristics. Comparing plant organisms with each other and determining the differences by qualitative signs of

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variability, we can judge one degree or another of mutations, changes in the source material.

Berry bushes are a colossal source of vitamin products that strengthen the health of the population. From early spring to late autumn, they delight a person with their fruits, filling the diet with vitamins. Huge potential in berries is contained in the form of biologically active substances useful for humans [3]. One of the first to bear fruit is *L. caerulea*. Along with the richest set of vitamins and microelements, the early fruiting period of *L. caerulea* is a factor determining its value; it is the first berry of summer [4-5]. Mankind uses the fruits of 8 types of honeysuckle for food [6-10]. They occupy different agro-climatic niches and bear fruit at different times. Another type of early-bearing honeysuckle, ripening at the same time as the blue honeysuckle *L. caerulea*, honeysuckle slender-petiolate glandular foma *L. gracilipes* was found [6-8].

2 Research Methodology

In order to accelerate the stimulation of the process of variability for selection in the rather conservative, in its heredity, ancient (Hara, 1983) species *L. gracilipes*, the seeds were irradiated with ionizing radiation in two versions, 1 Gy (Sv) and 2 Gy (Sv). Small doses of irradiation were chosen, which did not reduce the viability of seeds, but only induced mutations. Information on the reaction of *L. gracilipes* seeds to ionizing radiation is practically absent in the literature, so the dosages were used according to Z.P. Zholobova [11-12], who studied this issue on blue honeysuckle. A fluorograph brand FTS-01 “Electron” was used as an irradiation source (Fig. 1)



Fig. 1. Fluorograph FTs-01 “Electron” (left - rear panel, right - fluorograph screen with attached seeds)

Paper envelopes with dry seeds of *L. gracilipes* were attached to the fluorograph brand FTs-01 “Electron” (Fig. 3). In the first option for 50 days (total dose of ionizing radiation 1 Gy), and in the second for 100 days (total dose of ionizing radiation 2 Gy). The seeds in the first variant, after 50 days of exposure to microdoses on a fluorograph, were soaked in the stimulator “Epin Extra” + “Cytovit” (4 hours at a temperature of +200C; 1 ml “Epin Extra” per 1.5 liters of water + 1.5 ml “Cytovit”) and placed on coconut soil in the refrigerator +40C, 10/17/2021 until single seedlings appear on 12/13/2021, then the container with seedlings was transferred to a room with a temperature of +12-140C, where the seedlings unfolded cotyledon leaves on February 1, 2022.

3 Results and Discussions

In December 1994, in the Siberian Branch of the BSI FEB RAS, from the city of Sendai (Japan), seeds of the slender-petiolate form of the glandular honeysuckle (*Lonicera gracilipes* Mig var. *glanduosa* Maxim.) were obtained, belonging to the order Villusaceae, family Honeysuckle, subfamily Caprifolioideae, genus Honeysuckle. *Lonicera gracilipes* is a deciduous shrub up to 3 m high, endemic to Japan [7-8], found naturally in the south of the islands of Hokaido, Hon-shu and Shikoko. In the taxonomy of honeysuckle, it occupies an isolated place. Sometimes the species is singled out as a separate subsection. Widespread in gardens throughout Japan for a long time. It grows in deciduous forests in the mountains, at altitudes up to 600 m above sea level. The leaves are rounded, oppositely located on the shoot, apical broadly oval with a pointed end. Freshly blossomed leaves have a bright crimson edging. Blooms in Japan in the second half of May [13], in the Northern Branch of the BSI FEB RAS in the first ten days of June [6-8]. Elongated, pink, single, rarely paired bells of flowers up to 19.4 mm long have a pleasant aroma and attract insects. At the end of June, beginning of July, bright red single fruits (rarely fused two) ripen with a glossy-shiny skin covered with glandular outgrowths. The average weight of berries is 0.6 g with a content of dry soluble substances of 12.1%, pH of the juice is 5.1, the taste of berries is sweet, on average there are 4 seeds in a berry, which are 2 times larger than blue honeysuckle seeds. According to [6-8, 13] it is a tetraploid species ($2n=4x=36$).

Seeds received from Japan on January 23, 1995 were placed for stratification for three months at +40C, and sown in April. At the age of 3 years, the plants were planted in the garden collection in 1998. Currently, three specimens of slender-petiolate honeysuckle (*L. gracilipes*) have been preserved in the garden collection of the Japanese population. V.V. Sheiko [7-8] recommended a new type of berry shrub for cultivation in the southern zones of the Russian Federation after an introductory study at the SFBSI.

In the spring of 2021-22, a survey of the state of plants confirmed the freezing of the shoots of the formation above the snow cover. Growing cut shoots in the winter of 2021-22 and 2022-23 showed that shoots thinner than 3 mm die at a temperature of -200 C, shoots thinner than 5 mm and all buds cannot withstand -250C. Shoots thicker than 6 mm withstand temperatures down to -250C under snow cover (20-30 cm) and all buds and shoots winter well. Collection plants (75%) are in a weakened state, and 25% of plants are well developed, have a bush diameter of up to 1.5 m, a height of 0.8 m (at the level of snow cover). Such a different state of plants growing in the same soil-ecological conditions indicates the breadth of the climatic niche of the species and the potential for the introduction of this species in more severe conditions. In 2021, fruit ripening was noted on June 25, and in 2022 on June 30. The fruits are red, shiny, sweet (Fig. 2).



Fig. 2. Honeysuckle fruits (collected fruits - on the left and on the bush - on the right)

The entire crop is located on the shoots in a ground position (Fig. 2). The results of long-term observations of the state of the studied *L. gracilipes* specimens in the SFBSI collection determined the direction of research in the direction of increasing frost resistance, by the method of individual selection from the mass of seedlings of local seed reproduction. Phylogenetically young and old species are characterized by less variability than species that are in the prime of their development [14-16].

The experiment on seed germination showed that the seeds in the control variant, in the first variant, sprouted on February 17, 2022, almost at the same time as the seeds of the second variant, which indicates the need for a dormant period for the seeds of honeysuckle slender-petaled (Table 1).

Table 1. Terms of sowing - germination of seeds

Options	Sowing term	Duration of stratification	Seed germination time	Control germination time
1 option	17.10.21	55 days	13.12.21	17.02.22
2 option	13.12.21	60 days	13.02.22	13.02.22

Seeds in the second variant (after 100 days of exposure to microdoses on a fluorograph) are soaked for 4 hours in a growth stimulator solution (4 hours at a temperature of + 20 degrees C; 1 ml of Epin Extra per 1.5 liters of water + 1.5 ml of Cytovit) and placed on coconut soil in a refrigerator +4 degrees C, 13.12. 2021 Upon detection of a single pecking of seeds on February 13, 2022, a bowl with sown seeds of the second variant of exposure to ionizing radiation was transferred to a room with an air temperature of 12-140C; on March 2, 2022, friendly seedlings were found in the sowing of the variant and control.

Signs of qualitative variability visually determined on the seedlings of *L. gracilipes* confirm the correctness of the chosen dose of irradiation. Artificially provoked mutagenesis will allow for a short period of time to increase the variability of the species and obtain mutations that are not found in nature. It becomes possible to directly use the obtained mutants by selecting specimens with valuable pomological traits or to use the resulting organisms in the further selection process without finding a fully interesting variant of the traits.

When examining the seedlings of the first variant (50 days of exposure, 1GR), it was found that in some seedlings the hypocotyl was colored cherry (Fig. 3), in the control, the hypocotyl was green. The cotyledon leaves of the seedlings of the first variant are wave-like curved along the edges, the control has an even edge of the leaf. In the second variant, all seedlings have a cherry color of different intensity, colored hypocotyls, in the control, the hypocotyl is green.



Fig. 3. Seedlings of *L. gracilipes* (on the left - green hypocotyls, on the right - colored hypocotyls in seedlings)

The cotyledon leaves in the second variant, as in the first one, have a different degree of curvature of the edge (Fig. 3); in the control, the curvature of the edge of the cotyledon

leaves is absent. The next examination of seedlings in both variants revealed uneven appearance of the first pair of true leaves. In all shoots, both in the first and in the second variant, to varying degrees, there is noticeable intensive development from the first pair of true leaves and slow growth of the second true leaf (Fig. 4). In seedlings of control crops of both variants, both leaves develop evenly (photo5). On March 21, 2022, in the sowing of seeds of the first variant dated October 17, 2022, the appearance of the second pair of true leaves was found. One plant from this variant (1Gy) was completely dark green with a cherry hue, and roots were formed on the hypocotyl 4 mm above the soil level. In another plant from the same variant (1Gy), part of one leaf blade of the first true leaf is mosaically lightened. Over time, it becomes more and more noticeable that the hypocotyl is colored in a reddish hue in almost all seedlings in both variants. The venation of the second pair of true leaves is cherry-colored and in shades of cherry in different shoots, of different intensity. The mutant that gave roots above the soil level has a cherry color of the entire plant and a central shoot that does not develop. In many plants in both variants, the growth of lateral shoots from the buds above the first true leaf is observed.



Fig. 4. Intensive development of the first pair of true leaves and slow growth of the second true leaf (left and right)

In seedlings in variant 2 (2Gy), staining of hypocotyls with cherry blossom and leaves is also observed, in varying degrees of intensity in all seedlings. In the control, both in the first and in the second sowing, the seedlings are painted in exactly green color (Fig. 5).



Fig. 5. Control variant (green leaves and shoots)

Seedlings of control crops developed unevenly. In all seedlings from seeds exposed to ionizing radiation, both in the first and second versions, the growth retardation of the main shoot is more or less pronounced (Table 2).

Table 2. Indicators of qualitative variability of *L. gracilipes* seedlings

Indicators	1 option / control		2 option / control	
	PC.	%	PC.	%
Sown	50/14	100/100	50/14	100/100
Ascended	9/3	18/21	20/4	40/8

With stained hypocotyl	4/no	44/0	16/no	80/0
With a curved edge of cotyledon leaves	6/no	66/0	16/no	80/0
With variegated leaves	1/no	11/0	1/no	5/0
With stunting of the central shoot	5/no	55/0	12/no	60/0
With a cherry tint of real leaves	4/no	44/0	16/no	80/0
With green leaves (normal)	4/3	44/0	3/4	15/0
With roots above ground level	1/no	11/0	No no	0/0
With no glandular pubescence	3 /no	33/0	12/no	60/0

In both variants of the experiment, there are no seedlings corresponding to the control in terms of quality, i.e. all seeds exposed to ionizing radiation in a total dose of 1 Gy and 2 Gy gave presumably seedlings with an altered genetic set, seedlings in both variants are mutants in varying degrees of genetic deviations from the parental form (Fig. 6).



Fig. 6. Mutants with varying degrees of genetic abnormalities

4 Conclusions

A number of qualitative signs of the variability of seedlings exposed to ionizing radiation allows us to preliminarily speak about the resulting *L. gracilipes* mutations. Received View Forms *L. _ gracilipes* with other biological indicators differing to varying degrees from the original species make it possible in the future to single out promising forms valuable from an economic point of view in the offspring.

The currently existing economic indicators of the type *L. gracilipes* in terms of frost resistance (limit -25 °C), fruit weight - 0.6 g, low productivity (up to 300 g per bush), put forward specific tasks for the selection of a new rare crop, promising for cultivation as an early-fruiting berry bush.

Acknowledgments

The work was carried out within the framework of the program “Formation, conservation and study of bio-collections of the gene pool of various directions in order to preserve biodiversity and use them in health-saving technologies” (FGUU-2022-0014)

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