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RESEARCHES CONCERNING THE MULTIPLICATION *IN VIVO* OF LISIANTHUS FOR PROMOTING IN ROMANIAN GREEN HOUSES

Maria CANTOR, Rodica POP, Iudita Elisabeta CSETE, Buta ERZSEBET, Anca HUSTI

University of Agricultural Sciences and Veterinary Medicine, Faculty of Horticulture, 3-5 Manastur Street, Cluj-Napoca, Romania

Corresponding author email: marcantor@yahoo.com

Abstract

Current concerns of plant growers in Romania are to improve the flowers assortment with new species. Our researches focused on herbaceous ornamental plant Lisianthus russelianus Hook., (Eustoma grandiflora L.). It is a relatively new species in the range of cut and potted flowers for her beautiful colored and vase life. The new species introduced requires establishing efficient multiplication techniques. The biological material used in the experience were seeds from Sakata company (USA), represented by three cultivars: 'Echo Lavender', 'Flamenco White' and 'Mirage Pastel Pink'. Also vegetative multiplication by cuttings was experimented on different culture substrate. Plants were grown in greenhouses of USAMV Cluj-Napoca and Botanical Garden "Alexandru Borza" Cluj-Napoca. The results obtained will be using for Romanian growers for diversification their assortment in greenhouse.

Key words: *eustoma, cultivars, propagation, seeds, cuttings, Gentianaceae.*

INTRODUCTION

Lisianthus is a relatively new species in the range floricultural, Japan being the largest producer. *Lisianthus* is a species with great horticultural potential in Romania.

Lisianthus is a moderately cold-tolerant annual or biennial plant native to the southern part of the United States and Mexico (Roh and Lawson, 1988), belong to *Gentianaceae* family. In Texas is popularly known as 'blue bell', 'prairie rose' or 'prairie gentian'. Genus name alludes to the beauty calyx and corolla, the flower look is glossy, satin and beautiful border („lisieé" in French means smooth, glossy). From crossing wild forms, which shows blue flower petals, resulting in improvement works form a large flower perfection extremely varied in color and shape.

In Europe started to be cultivated only after the 1970s. After France, the Netherlands ranks second in the culture of this beautiful flower. In Romania the first cultures were established from Companies Codlea and early cultures were initiated at University of Agronomic Sciences Bucharest since 1989 (Cantor, 2009).

This plant grows to 50-75 cm in height with 20-40 flowers. By nature, *Lisianthus* initially forms a rosette and grows very slowly during the winter, stems elongate in the spring, and it flowers in summer (Roh et al., 1989). In recent decades, breeders have developed a variety of cultivars with respect to many traits such as uniform flowering throughout the year, lack of rosetting, heat tolerance, flower color, and flower size and form, including double flowers etc (Harbaugh, 2006). The importance of this ornamental flower is due to its beauty, diversity of colors, excellent keeping quality, and wide range of different forms (Ali et al., 2008, Kanwar and Kumar, 2009). It is known that to this genus *Lisianthus* (*Eustoma*) are belonging 27 species, herbaceous and woody plants. It is a relatively new species in the range of cut and potted flowers.

Due decorative qualities lisianthus culture's expansion took in most of Europe and thus flower growers and enthusiasts have expressed a particular interest in this crop. *Lisianthus* flowers are much used by those who deal with flower arrangements and bouquets for different occasions. Lisianthus is highly regarded not only as a cut flower, and as the plant pot. Currently, in our country, lisianthus culture is not widespread, although there is market demand. Most flowers sold and used by florists are imported from Holland.

For the new species introduced are required efficient multiplication techniques to be established.

Eustoma randiflorum is commonly propagated by seed or cutting. Sexual propagation used to obtain seedlings seeds brought from abroad (Netherlands, USA, France, etc). From a gram of seeds can be obtained about 8000 plants (Bala, 2010).

Generally, sowing the plants of Lisianthus is in autumn or January-February. Industrial culture, sowing can be done in half, and according to this period, the number of days to flowering may vary.

Propagation by cuttings is less practiced. Cuttings are cut to a length of 10 to 15 cm (which have three pairs of leaves) and placed on the substrate of peat and perlite, sand or perlite simple. Rooting takes place for 40-50 days. The disadvantage of this method is that the percentage of shoots of the plant is weaker, we get a few plants and can easily transmit diseases and pests (Toma, 2009).

MATERIALS AND METHODS

For the new species introduced are required efficient multiplication techniques to be established. The study was about conventional propagation methods, by seeds. The biological material used in the experience comprised in seeds from Sakata Company, represented by three F_1 hybrids of *Lisianthus* russelianus: „Echo Lavender", „Flamenco White" and „Mirage Pastel Pink". These were used to obtain plants to start the *in vitro* and *in vivo* experiments in order to establish the germination and the rooting rates. Seeds were treated thiuram pelleted. The biological material presents the following characters:

„Echo Lavender". It is one of the most popular cultivars with early flowering. Plants are vigorous in spring, while in summer droughts become more fragile. The leaves are 5-8 cm long, elliptical, slightly sharp and succulent; they look dull and bluish-green color, with 3-5 ribs clearly visible. Summer flower buds open in cones than 5 cm in diameter, showing many shades of purple.

„Flamenco White". The flowers are simple, is suitable for cultivation in summer when temperatures are high and days are long. Stems are vigorous and flowers on top of them are white. Leaves are skin, colored bluish green, located opposite the stem. Stem height is 50 cm. Blooms two weeks earlier than hybrid *Mirage Pastel Pink.*

„Mirage Pastel Pink". Flowers are simple, ideal for summer flowering. Has smaller petals, and are resistant to transport. The flowers are pink, with the little white. The leaves are green and vigorous stems are about 40-50 cm in length. It is resistant to high temperatures and botrytis.

Experience was made in greenhouses of USAMV Cluj-Napoca and the greenhouses of the Botanical Garden Alexandru Borza Cluj-Napoca.

Propagation by seeds

Sowing was done on 04.03.2011, in small pockets trays using the neutralized peat substrate. The seeds were sown on the surface because they are photosensitive (germinate in the presence of light). Greenhouse temperature was 20-22°C during germination, and then decreased to 18-20°C. Relative humidity was 70-75% and 80-85% of the substrate, maintaining this level through daily watering. Germination occurred at 21 days, after which observations were made on the percentage of germination. During the period of germination were assured maintenance, or weeding, watering and ventilation greenhouse. Lisianthus from each hybrid were sown one hundred seeds.

After about six weeks on 4/24/2011 plants produced two little leaves true. Four days later, on 05.28.2011 was carried out first transplantation in alveolar larger trays, then on 6/10/2011 moved in pots 7x7cm (Figure 1).

Figure 1. Plants in cellular trays and pots (original)

Vegetative propagation by cuttings

Cuttings were made from plants grown in the greenhouse of the Botanical Garden 'Alexandru Borza' Cluj-Napoca from the seeds of the three hybrids studied. Leaves were removed from cuttings length exempting those in the apical zone (Figure 2).

Cuttings were trimmed to the size of about eight cm and planted in wooden boxes on different rooting substrates.

For rooting of cuttings were used three types of substrate:-S1-sand river; -S2-peat + sand, mixture of 1:1 and; -S3-peat + sand + garden soil, mixture of 1:1:1.

After 40 days there have been observations on rooting percentage. *In vivo* plants were grown and maintained in the greenhouse of the USAMV Cluj-Napoca and served to multiply by cuttings and *in vitro* propagation.

Figure 2. Aspects of in vivo propagation by cuttings (original)

RESULTS AND DISCUSSIONS

Results of germination rate

In vivo experience in seed germination percentages first comment on were made at about 21 days, at which time it was considered the emergence over. Data on the percentage of germination are shown in Table 1. The performed analyzes on the percentage of germination is a weak seed germination in all hybrids analyzed, with an average rate of 65% (Table 1).

Table 1. The percentage of germination in vivo three hybrids examined

Hybrid name	No of seeds sown	No of seeds germinated	Percentage of geminated seeds (%)
Mirage Pastel Pink	100	64	64
Echo Lavander	100	69	69
Flamenco White	100	62	62
Average	100	65	65

Analyzing the data in Table 1 it can be seen that the highest percentage of germination was obtained in the F_1 hybrid *Echo Lavender*

respectively 69%, followed by hybrid *Mirage Pastel Pink* with a germination rate of 64%. The lowest percentage of germination was obtained in hybrid *Flamenco White* of only 62%.

The data obtained are in accordance with the literature, which mentions a low germination in this species, both because of the very small size of seeds and germination special conditions, namely the presence of light (Cantor, 2009).

Results for rooting cuttings

Data on percentage of rooting cuttings from three hybrids analyzed are presented in Table 2.

Table 2. Rooting percentage in the three Lisianthus hybrids

Variant[1]	Number of rooted cuttings used	Number of rooted cuttings	Percentage of rooting (%)
S1E	30	0	0
S2 E	30	5	16
S3E	30	3	10
S1F	30	0	0
S2F	30	4	13
S3F	30	3	10
S1M	30	0	0
S2M	30	8	26
S3M	30	4	13
Total number	270	27	10

1Substrate 1Hybrids;
S1-river sand; E-*Echo Lavender;*
S2-peat + sand, mixture 1:1; F-*Flamenco White;*
S3-peat + sand + garden soil, mixture 1:1:1; M-*Mirage Pastel Pink*

From the table it can be seen that the highest percentage of rooting was obtained in hybrid *Mirage Pink Pastel* on sand and peat substrate mixed 1:1 with 26%, followed by hybrid *Echo Lavender* on the same substrate, 16%. A small percentage of rooting was recorded and the substrate composed of soil-peat-sand in a ratio of 1:1:1. *Pastel Pink Mirage* hybrid resulted in a 13% and other hybrids with a percentage of 10%. If the substrate consists of river sand has no rooted cuttings.

CONCLUSIONS

Based on the experiences made the following conclusions are:

Data obtained shows that the seeds size and ecological conditions influenced negatively the germination percentage, registering an average of only 65% for the analyzed hybrids.

The experiments conducted shows that species *Lisianthus russelianus* Hook. is quite recalcitrant to conventional vegetative propagation, by cuttings, regardless of the substrate used.

The small percentage of rooting, probably due to the fact that greenhouse facility was not equipped with 'mist system' that does not ensure a high air humidity nor stimulators were used rootedness.

We recommend the promoting culture in our country, of the species *Lisianthus russelianus* Hook., by enriching assortment with new cultivars.

Biological material obtained by applying conventional propagation methods lead to obtaining parent plants which can be used as a basis for *in vitro* multiplication that will produce an unlimited number of plants in a relatively short time.

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