# Technology of growing orchid flowers from seeds

*Surayyo* Misirova*

Namangan Engineering-Technological Institute, Namangan, Uzbekistan

**Abstract.** For the first time, the introduction and adaptability of the Felionopsis variety, an orchid (Orchidaceae J.) imported from Malaysia, was analyzed. When using 0.4 l/mg of BAP (quinitine) as part of a nutrient medium for growing orchids in vitro from seeds and vegetative parts, the number of flowers at the 18th month was 10 or 2 times compared with the preparation Trichoderma vrayde and it was found that it is possible increase activated carbon by 2.5 times. It has been proven that up to 60% of explants can be obtained in 2 weeks using the MS+1NAA+1BAP hormone when cultivating an orchid flower stem in vitro.

## 1 Introduction

To date, a number of scientific and practical works are being carried out on a global scale to develop floriculture and landscape design, landscaping roads, settlements, parks and gardens. In landscape design, orchids are one of the leading flowers of the world, and they are grown in open (up to 30%) and closed (up to 80%) ground. The creation of new varieties, as well as the development of technology for breeding orchids used in landscaping, is of current importance.

According to the International Florists Association, there are currently 290 species in the existing range of flowers, 50 of which are cultivated as the main species. Among the main species, orchids are of particular importance, and large-scale research is being carried out to create new varieties of them in countries such as Holland, Germany, Italy, Spain, Kenya. As a result, more than 30,000 varieties of orchids have been created, which are widely used in floriculture. The countries that grow these species are doing a lot of research to create their new species. As a result, about 500 varieties of orchids are created annually [1,2].

In our country, a number of measures are being implemented to create and expand effective plantings of flowering plants in the system of urban planning and landscaping. The basis for scientific research are the goals and objectives arising from the Decree of the Cabinet of Ministers of the Republic of Uzbekistan No. 830 dated October 16, 2017 “Prospective development of floriculture in the Namangan region”, instructions during the President’s visit to Andijan region dated June 6, 2021 “On organizing the creation of a cluster on the basis of the Andijan Flowers Garden enterprise and the creation of an Andijan orchid flower sample”, as well as other regulatory legal acts of the relevant industries [3].

* Corresponding author: samisirova@mail.ru

The purpose of this work is to study the morphology of seeds of orchid varieties (Orchidaceae J.), introduced from abroad, used for landscaping in Uzbekistan, as well as to develop a technology for growing these flowers from seeds and vegetatively, and from seeds of seedlings - new, in addition, to select and evaluate promising varieties.

## 2 Materials and methods

Research on biodiversity, distribution, bioecological characteristics, as well as breeding of orchids, on the creation of new varieties, breeding technology, cultivation and selection using scientific achievements are conducted by the world's leading scientific centers and higher educational institutions such as the International Seabuckthorn Association (China), Himachal Pradesh Agricultural University (India), Technical University of Berlin (Germany), University of Turku (Finland), Biological research Institute of Romania (Romania), All-Russian Research Institute of Medicinal and Ornamental Plants (Russia), Academician Siberian Research Institute of Horticulture M.A. Lisavenko (Russia), Botanical Gardens of the National Academy of Ozarbaijon and Samarkand State University (Uzbekistan). As a result of these studies, the distribution of orchid species, decorative features and methods of intensive reproduction, diseases and pests have been determined, agro-techniques for growing the above-mentioned plants in plantations have been developed, more than 30,000 orchid varieties have been created to date, and a technology for their reproduction and cultivation on plantations has been developed. In the field of floriculture, research work is carried out mainly in the following priority areas: selection of promising forms of orchids with valuable economic and biological characteristics suitable for each soil and climatic condition and the creation of new varieties; development and improvement of effective methods of their reproduction and resource-saving technologies for growing seedlings, caring for plantations [4-7].

## 3 Results and discussion

On the selection of orchids, improving the technology of caring for plantations and growing seedlings, large-scale studies were carried out by such researchers as I.V. Belitsky, V. Morozov, G.L. Kolomeytseva, S.O. Gerasimov (Russia), Chiba Masaaki (Japan) , R.L. Dressler, Harper Tom, White Taylor, Judy Taylor (USA), Leroy-Terquem, Gerald and Jean Parisot (England), Lyuis Knudson (Netherlands) Andżelika Byczyńska, Agnieszka Zawadzińska, Piotr Salachna (Poland). In order to further develop and coordinate scientific research in the field of floriculture, the International Flower Trade Association was established in Brussels, which brings to the general public the achievements of flower growers of the world. In Uzbekistan, such scientists as Z.P. Bochantseva, K.Sh. Tajiboev, V.P. Pechenitsin, M.T. conditions, natural stocks and phenology of species growing in local conditions. These scientists did not conduct research aimed at the introduction, adaptation and cultivation of foreign varieties widely used in landscaping. Research on growing orchids in Uzbekistan has not been carried out before. Analysis of the results of this study showed that in Uzbekistan there were separate studies on orchid breeding, but no studies were carried out on cultivation technology. As for the orchid, not only has the technology of reproduction and growing seedlings not been developed, but this flower has been scientifically studied for the first time in the climatic conditions of Uzbekistan. In this regard, scientific research on the development of technology for breeding and growing seedlings of these flowers becomes relevant [8-10].

Seed separation is carried out in laminar boxes sterilized with bactericidal lamps. The seeds are placed on the surface of the nutrient medium in the test tube. There are several

studies on orchid seed germination and temperature. As with many other species, orchid seeds grow over a range of temperatures, but maximum germination is achieved only within a narrow range. Orchid seeds germinate between 10 and 30°C, but the optimum temperature range appears to be between 23 and 24.5°C. Germination percentages decrease below 15°C and above 27°C [9,10].

BAP (kinitin), Trichoderma vride, Activated charcoal were used for extracting callus tissue from orchid seeds, and the nutrient medium prepared with BAP (kinitin) was the most optimal variant, and the process of extracting callus from seeds was accelerated (Table 1,2,3). Viability of callus tissue obtained from orchid seeds was determined in incubation and greenhouse. After 50 seeds germinated in incubation, 48 remained in 7 days, which is 96%. 48 surviving orchid flowers were transferred to the greenhouse and kept for 14 days, 48 flowers survived and 100% was adaptated (Table 4). The development of roots and stems of orchid seeds grown in nutrient media was observed for 18 weeks, and the most favorable variant was also obtained in media prepared with BAP (kinitin). It was observed that the maximum roots grew to 12.20±9.40 cm and stems to 55.10±15.20 cm in 18 weeks (Table 5). The flowering dynamics of the grown orchid was observed for 18 weeks, and variant 1 was found to be the most optimal variant. It was observed that 100% of orchid flowers bloomed in 18 weeks (Table 6). The sequence of extracting callus tissue from orchid flower seeds is shown (Figure 1).

**Table 1.** Effect of the amount of BAP (kinitin) in the nutrient medium on the dynamics of plant germination during in vitro propagation of orchid flowers from callus tissue.

| **Options** | **MS (mixture of mineral salts) 1l/gr** | **Carbohydrate (Sucrose) 1l/gr** | **Inositol 1l/gr** | **Amino acid protein substances (Casein) 1l/gr** | **BAP/kinitin 1l/mg** | **Antibiotic (benzylpenicillin) 1l/mg** | **Agar agar 1l/gr** | **Neutralizing reagent** | **Solution environment** | **The average number of root pieces** | **The average length of the root cm** |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | | **10 weeks** | **10 weeks** |
| control | 4.4 | 30 | 0.01 | 0.01 | 0.0 | 100 | 6 | NaOH | 5.8 | - | - |
| 1st experiment | 4.4 | 30 | 0.01 | 0.01 | 0.5 | 100 | 6 | NaOH | 5.8 | 5±4 | 1.25±0.20 |
| 2nd experiment | 4.4 | 30 | 0.01 | 0.01 | 1.0 | 100 | 6 | NaOH | 5.8 | 6±5 | 1.31±0.22 |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 3rd experiment | 4.4 | 30 | 0.01 | 0.01 | 1.5 | 100 | 6 | NaOH | 5.8 | 7±4 | 1.35±0.20 |
| 4th experiment | 4.4 | 30 | 0.01 | 0.01 | 2.0 | 100 | 6 | NaOH | 5.8 | 9±7 | 3.44±2.46 |

**Table 2.** Effect of the amount of Trihoderma vride in the nutrient medium on the dynamics of plant germination during in vitro propagation of orchid flowers from callus tissue.

| **Options** | **MS (mixture of mineral salts) l/gr** | **Carbohydrate (Sucrose) l/gr** | **Inositol l/gr** | **Amino acid protein substances (Casein) 1l/gr** | **Trihoderma vride** | **Antibiotic (benzyl penicillin) l/mg** | **Agar agar 1l/gr** | **Neutralizing reagent** | **Solution environment** | **The average number of root pieces** | **The average length of the root cm** |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | | **10 weeks** | **10 weeks** |
| control | 4.4 | 30 | 0.01 | 0.01 | - | 100 | 6 | NaOH | 5.8 | - | - |
| 1st experiment | 4.4 | 30 | 0.01 | 0.01 | 1.5 | 100 | 6 | NaOH | 5.8 | 3±2 | 0.70±0.65 |
| 2nd experiment | 4.4 | 30 | 0.01 | 0.01 | 1.3 | 100 | 6 | NaOH | 5.8 | 2±1 | 0.61±0.42 |
| 3rd experiment | 4.4 | 30 | 0.01 | 0.01 | 1.2 | 100 | 6 | NaOH | 5.8 | 2±1 | 0.32±0.29 |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 4th experiment | 4.4 | 30 | 0.01 | 0.01 | 1 | 100 | 6 | NaOH | 5.8 | 2±1 | 0.32±0.25 |

**Table 3.** Effect of the amount of Activated charcoal in the nutrient medium on the dynamics of plant germination during in vitro propagation of orchid flowers from callus tissue.

| **Options** | **MS (mixture of mineral salts) 1l/gr** | **Carbohydrate (Sucrose) 1l/gr** | **Inositol 1l/gr** | **Amino acid protein substances (Casein) 1l/gr** | **Activated charcoal** | **Antibiotic (benzyl penicillin) 1l/mg** | **Agar agar 1l/gr** | **Neutralizing reagent** | **Solution environment** | **The average number of root pieces** | **The average length of the root cm** |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | | **10 weeks** | **10 weeks** |
| control | 4.4 | 30 | 0.01 | 0.01 | - | 100 | 6 | NaOH | 5.8 | - | - |
| 1st experiment | 4.4 | 30 | 0.01 | 0.01 | 1.0 | 100 | 6 | NaOH | 5.8 | 3±2 | 0.60±0.55 |
| 2nd experiment | 4.4 | 30 | 0.01 | 0.01 | 0.8 | 100 | 6 | NaOH | 5.8 | 2±1 | 0.51±0.42 |
| 3rd experiment | 4.4 | 30 | 0.01 | 0.01 | 0.7 | 100 | 6 | NaOH | 5.8 | 2±1 | 0.32±0.29 |
| 4th experiment | 4.4 | 30 | 0.01 | 0.01 | 0.5 | 100 | 6 | NaOH | 5.8 | 2±1 | 0.31±0.25 |

**Table 4.** Viability rate in cultured callus tissue from orchid seeds.

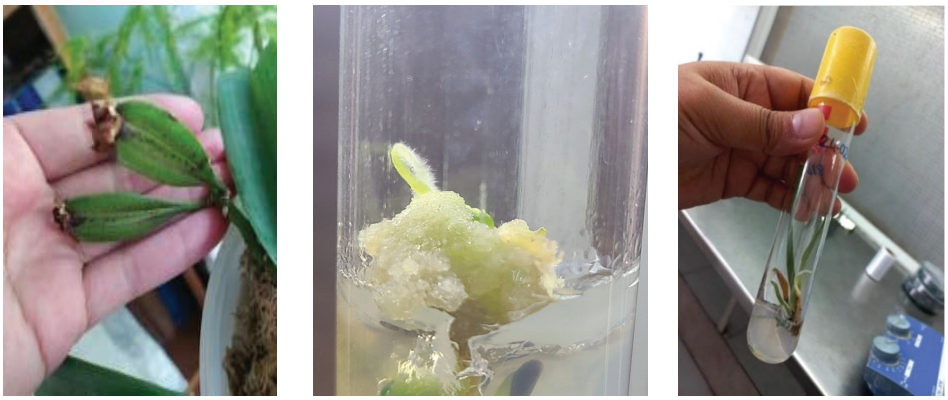
| **Growing room** | **The number of plants transferred to the second nutrient medium** | **Checking days** | **Amount of surviving plants** | **The degree of surviving %** |
|---|---|---|---|---|
| Incubation (laboratory) | 50 | 7 days | 48 | 96 |
| Greenhouse | 48 | 14 days | 48 | 100 |

**Table 5.** The effect of reactants on the growth dynamics of roots and stems in obtaining callus tissue from orchid seeds in method of in vitro.

| **Options** | **Length of 10-week-old vegetative organs sm** | | **Length of 30-week-old vegetative organs sm** | | **Length of 50-week-old vegetative organs sm** | | **Length of 70-week-old vegetative organs sm** | |
|---|---|---|---|---|---|---|---|---|
| | **root** | **stem** | **root** | **stem** | **root** | **stem** | **root** | **stem** |
| control | - | - | - | - | - | - | - | - |
| БАП 1l/gr | 3.44± 2.46 | 25.9±18.20 | 4.80± 3.10 | 38.4± 31.20 | 10.80 ±6.20 | 49.35 ±36.20 | 12.20 ±9.40 | 55.10 ±15.20 |
| Trihoderma vrida l/gr | 0.32± 0.25 | 11.9±4.40 | 0.80± 0.70 | 13.4± 12.20 | 2.75± 2.15 | 17.54 ±16.30 | 3.20± 2.75 | 18.12 ±14.34 |
| Activated charcoal % | 0.31± 0.25 | 9.80±3.20 | 0.90± 0.50 | 12.5± 10.10 | 2.30± 2.10 | 16.34 ±15.25 | 3.30± 3.10 | 16.10 ±13.30 |

**Table 6.** Effect of reagents on the dynamics of flowering in the extraction of callus tissue from orchid seeds in method of in vitro.

| **Options** | **Number of 15th month flower, piece %** | | **Number of 16th month flower, piece %** | | **Number of 17th month flower, piece %** | | **Number of 18th month flower, piece %** | |
|---|---|---|---|---|---|---|---|---|
| БАП l/gr | 1±2 | 20 | 3±5 | 50 | 9±10 | 100 | 10 | 100 |
| Trihoderma vrida l/gr | 1±2 | 20 | 1±3 | 30 | 3±4 | 40 | 5±7 | 70 |
| Activated charcoal % | - | 0 | 1±3 | 30 | 2±4 | 40 | 4±6 | 60 |



a b c

**Fig. 1.** Fungal microbes in the process of growing orchid seeds on a hormone-free medium. a-orchid callus formation process; b - orchid fertilisation process; c - the process of cultivation an orchid on a nutrient medium.

## 4 Conclusions

By using 0.4l/mg of BAP (quinitin) in the nutrient medium for growing orchids in vitro from seeds and vegetative parts, the number of flowers in the 18th month increased by 10 pieces or 2 times compared to Trichoderma vrayde preparation, and 2.5 times compared to activated carbon was determined. It was proposed to add 1 mg of BAP and 1 mg of NAA to the nutrient medium for growing Orchids (Orchidoseae J.) in vitro. It was recommended to store orchid buds on the stems with cytokinin oil in a room with a temperature of 24-250C and 50% humidity.

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