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Research Article

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Production of Microtubers From in Vitro Potato on the Heavy Clay Soil in Georgia

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ABSTRACT

In Georgia, potatoes, both seed and food material, are mainly produced in mountainous regions, at an altitude of 1000-2500 meters above sea level, where the black soil allows to obtain high-quality food potatoes.

The main goal of the study was to obtain potato primary microtubers from in vitro plants in the village of Ajamet, Imereti region of Georgia, which is located at an altitude of 104 meters above sea level.

Potato variety “Sante” was used from the in vitro collection of Biotechnology Center. Apical Meristem method was used for establish in vitro culture on the Murashige ans Skoog (MS) medium.

Out of 1500 in vitro potato variety “Sante” were placed in plastic pots filled with soil for strengthen the root system which regularly was irrigated with water and 1% YaraTera™ Kristalon™ under laboratory conditions during 4-5 days.

Potato seedlings were planted at the demo plot in the village of “Ajamet”, Imereti region of Georgia at the end of May. 2.5 months after transplanting the potato variety "Sante", microtubers with typical maturity but non-standard shape were obtained, which is caused by the heavy soil in the lowland area of Georgia.

Despite the non-standard size, it is quite possible to obtain a certain amount of edible potatoes from these microtubers in the following years, by cultivating the local soil (loosening, adding fertilizers), especially since the starting material was in vitro potato obtained from the apical meristem.

First time in Georgia the potato microtubers were produced from potato seedlings obtained from in vitro plants (without greenhouse) on the heavy clay soil in Georgia.

Keywords: Potato, Microtuber, In Vitro, Seedling, Open Field, Production

Introduction

Cell technologies are based on in vitro cultivation of cells, tissues, and organs. The process of clonal propagation consists of three stages. First, the isolation of an explant from the initial plant tissue. At this stage, it is necessary to obtain an infection-free culture, survival-adaptation of the explant on a medium, and rapid growth. The second -micropropagation -the growth of number of microclones and third- rooting. In this technology, the transfer of microplants from sterile to non-sterile conditions is the most

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important stage of production. Therefore, first of all, it is necessary to place in vitro plants in various types of containers field with soil in laboratories to strengthen for their further growth in open filed.

Such enhanced in vitro plants' ability to adapt to stressful situations in the open field. According to scientific research, the combination of plant adaptation and rooting should begin in laboratories, which will subsequently increase the yield of tubers obtained from such plants in open filed.[1]

In vitro conditions are fundamentally different from ex vitro conditions. For getting a high multiplication factor, it is necessary to provide plants with a different composition of salts in the soil, change the concentration of the soil solution, increase the amount of growth regulators. The photosynthesis is practically not carried out in in vitro stage; a large amount of ethylene accumulates in the plant tissues. Long-term conversation of plant tissues in such conditions leads to various physiological abnormalities. Many authors note a reduction in the cell layers in the leaf parenchyma, their porosity with large air spaces. [2]

According some scientist's in vitro plants develop non-functional stomata that do not close under the influence of a number of specific factors. Roots formed in vitro do not have stomata, the vascular system is poorly developed, and the cells are much enlarged. Development of in vitro plants are is depended by in vitro conditions, namely, the composition of the medium, lighting, etc. [3] According to a number of scientists, such plants cannot adapt to different environmental conditions. Plant stress during transplantation into open filed leads to the death of a large number of plants. Adaptation in ex vitro conditions is complicated for plants; therefore, it is necessary to extend the period of temperature, light and atmospheric humidity for them, which is possible only in laboratory conditions. However, in most cases, the death of plants is observed 10-15 days after transplantation. Therefore, it is necessary to create all the conditions at the in vitro rooting stage so that at the moment of transplantation the plants are strong in ex vitro unfavorable conditions. A certain part of the scientific community, based on research, focuses on the morphological parameters of plants during this period (stem length, leaf diameter, number of nodes) [4].

In addition, plants have a special ability to adapt to the environment in which they have to develop. They are affected by drought, excessive humidity, soil composition, unfavorable conditions for plant development cause plants to develop genetic adaptations to existing conditions [5]. Which is carried out by the variation of genetic mechanisms, inheritance and selection. Scientists associate this ability of plants with their genetic adaptation, in terms of adaptability to adverse environmental conditions. The program for the development of the characteristics of all plant species is embedded in the genetic material. The material in which the encoded program is transmitted from one generation to another remains unchanged, species look absolutely the same. In the population of organisms of all species, small changes in the genetic material occur. Every plant has the ability to adapt, which is determined by its genotype. These properties distinguish resistant varieties of agricultural crops [7].

In vitro plants must be prepared for transplanting into the open field. Rooting of micro plant shoots, their subsequent adaptation to the soil, and transplanting into greenhouses or open ground are the most difficult stages on which the success of clonal micropropagation is based.

The production of minitubers form seedlings was performed first time in Georgia Mountain potato region, The research needs to be expanded to different regions of Georgia for future improvement potato seed production system according varieties and regions [8].

The main goal of the study was to obtain potato primary microtubers from in vitro potato variety "Sante" in the village of Ajamet, Imereti region of Georgia, which is located at an altitude of 104 meters above sea level.

Materials and Methods

Potato variety "Sante" was used from the in vitro collection of Biotechnology Center. Murashige and Skoog medium (MS) was used to propagate potato in vitro explants [9]. Medium with 60 g/L-1 sucrose, 7 g/L-1 agar autoclaved to 121°C during 20 min at 15 psi, pH was adjusted to 6.1. 2-3 cm long single nodes were separated from 4-5 weeks old explants with 5-6 nodal segments under laminar flow chamber and used as explants for in vitro propagation.

The Potato virus A (PVA), Potato virus M (PVM), Potato virus S (PVS), Potato virus X (PVX), Potato virus Y (PVY) and Potato leaf roll virus (PLRV) infections were study using Double Antibody Sandwich (DAS)-ELISA using commercial Kits (BIOREBA AG, Switzerland) according to the manufacturer's Instructions: leaves were crushed ((w/v) 1:5)in extraction buffer (pH 8.2) containing 2% polyvinylpyrrolidone (PVP MW 24,000) 0.02% NaN3 and 0.05% Tween 20 [10]. Absorbance was determined at 405/450 nm on ELX800 Microplate Reader (Bio-Tek Instruments, Winooski, VT) and Samples with absorbance values greater than or equal to three times of the average of the kit negative control samples were considered infected.

Out of 1500 in vitro potato variety "Sante" were placed in plastic pots filled with soil for strengthen the root system which regularly was irrigated with water and 1% YaraTera™ Kristalon™ under laboratory conditions during 4-5 days.

Potato seedlings were planted at the demo plot in the village of Ajamet, Imereti region of Georgia at the end of June. Potato plants growth and development were stronly controled during the vegetation period.

Results and Discussions

For in vitro propagation of potato variety "Sante" plants were placed in phytotrone regulate condition (temperature-24-260 C; humidity-70-75%; 4500-5000 lux, 16/8h). The formation of in vitro plants was completed after 15-17 days with 7-8 nodes. The plants were reproduced a second and third time for their massive propagation.

To strengthen the obtained in vitro tube plants, we transferred the plants to polyethylene cups and covered them with fertile soil so that only one leaf remained above the soil and placed in this condition during 6-7 days (Figure 1).

Figure 1: The process of planting plants to polyethylene cups

During this period, the plants were treated twice with a 1.0% green crystallon solution. Once immediately after planting the plants in the glass and the second after 5days. Before transferring to open field, the plants had already developed strong stems and roots with 3-4 nodes (Figure 2).

Figure 2: Strengthened in vitro plants with 3-4 nodes

Potato seedlings were planted at the demo plot in the village of Ajamet, Imereti region of Georgia at the end of May. The distance between the rows was 40 cm, The distance between the plants was 20 cm. During the entire potato growing season, three hoeings were carried out, the plants were treated every fourth day with a 2% "Biocatena" working solution, 50-100 ml of the working mixture was applied to each root of the plants (Figure 3).

Figure 3: Planting in vitro potato seedlings into open filed

Potato plants growth and development were stronly controled during the vegetation period. Due to climatic conditions and soil structure the growth of plants was different. During the study period, June, July, and August 2024 were marked by unusually high temperatures (+39+42°C) and low rainfall for this area. Despite irrigation, most of the plants failed to complete the growing season, however, some surviving plants continued to grow and develop, which is related to the agricultural practices used on the plot and the genotype of the potato variety.

The potato variety "Sante" was introduced to Georgia in the 1990s and has remained one of the most popular varieties. "Sante" is quite resistant and has easily adapted to the environmental conditions of Georgia. (Figure 4).

Figure 4: Seedlings of potato variety "Sante" in open filed

2.5 months after transplanting the potato variety "Sante", microtubers with typical maturity but non-standard shape were obtained, which is caused by the heavy soil in the lowland area of Georgia.

No viral diseases were observed in the received mini-tubers. The production of virus free potato microtubers is related the fact that the potato plantations are quite far from the village, and there are no fruit orchards which is one of the important sources of the spread of viruses.

Despite the non-standard size, it is quite possible to obtain a certain amount of edible potatoes from these microtubers in the following years, by cultivating the local soil (loosening, adding fertilizers), especially since the starting material was in vitro potato obtained from the apical meristem (Figure 5).

Figure 5: Microtubers received from in vitro potato variety "Sante" in open filed

Conclusion

First time in Georgia the potato microtubers were produced from potato seedlings obtained from in vitro plants (without greenhouse) on the heavy clay soil located at 104 meters above sea level for further reproduction of super-super elite, and ultimately elite potato planting material in Georgia.

It is noted that the local population produces only vegetables and berries. Based on the results of the study, we can conclude that with proper soil cultivation, it is also possible to produce both food and seed potatoes, which is important for farmers living in the lowland zone of Georgia.

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