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FEATURES OF GROWING PINEAPPLE (ANANAS COMOSUS) IN VITRO CONDITIONS

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ABSTRACT

This article provides information about sterilization and the choice of in vitro culture medium for the introduction of pineapple.

Keywords. in vitro, clone, vegetative propagation, rosette of leaves, sucker, medicinal, explant, sterilizing reagent, nutrient medium

INTRODUCTION

Relevance of the topic. There is no area where plants and their products are not used. This is due to their biological, ecological, agronomic and social significance. Biological significance is related to the satisfaction of human needs for natural substances, including vegetable protein, vitamins and minerals, and ecological importance is related to ensuring the purity of atmospheric air. At the same time, crop production (agricultural Culture is one of the main areas of income for the population and contributes to the improvement of their social situation. According to scientific sources, once people used about 3 thousand species of plants, now more than 10 species are considered the most used. It can be seen that the targeted use of plants and their products is one of the most important tasks facing humanity. Because the world of plants is one of the main sources of satisfying the growing need of the population for food.

In the "Development Strategy of New Uzbekistan for 2022-2026" of the President of the Republic of Uzbekistan, increasing the income of farmers and farmers by at least 2 times through the intensive development of agriculture on a scientific basis, further development of fruit and vegetable growing, and the pharmaceutical industry are identified as priority goals. Instead, it should be noted that meeting the growing demand of the population for quality food is essential to improving their health. In particular, the propagation and use of medicinal plants is one of the important tasks of our time. Decree of the President of the Republic of Uzbekistan "On measures for the protection of medicinal plants growing in the wild, cultural cultivation, processing and rational use of available resources" As noted in the Of the more than 4,300 plants belonging to the local flora, 750 species are considered medicinal, of which 112 are listed for use in scientific medicine, of which 70 species are actively used in the pharmaceutical industry. on the application of scientific achievements. Instead, it should be noted that the rapid development of science and technology and the lack of activity among the population lead to an increase in the number of people. At the same time, excessive consumption of products and sweets made from animal fats and high-quality flour products, non-compliance with the diet and rhythm cause an imbalance between the main components of food and, as a result, obesity. This situation leads to the occurrence of cardiovascular and other diseases among the population. One of the main reasons The onset of this disease is an increase in the amount of cholesterol (fat particles) in the blood, high blood pressure,

myocardial infarction, cerebral hemorrhage or ischemic stroke (due to the accumulation of fats in the vessels of the brain and neck), diabetes causes diseases. One of the most pressing problems today is the use of plants and the search for a cure for such diseases instead of synthetic drugs.

To solve these problems, the President of the Republic of Uzbekistan has adopted special resolutions, with the help of which it is possible to grow fruits, vegetables and fruit and berry products with nutritional properties in our country. This, in turn, creates a full opportunity for the effective operation of peasant farms operating in these areas. In In particular, the issues of growing fruits and vegetables and their export in our republic today have become much more advanced, which allows ordinary farmers to freely export their products abroad, which further expands the opportunities of the population to export their own products abroad. But despite this, some tropical and subtropical fruits are still bought abroad for foreign currency. This is one of the pressing issues facing our scientists today.

Unconditionally, in our country, high-quality and high-quality products are grown from fruit and vegetable crops. However, in the healthy life of a person, there are such species of plants that are mainly grown in other climatic regions and are annually imported to our republic. These plants are not only medicinal, but also loved by people for their beauty. Such plants include the pineapple Ananas somosus (L.) Merrplant. Pineapple contains vitamins (C, carotene, B1, B2, B5, B12, PP), calcium, phosphorus, iron, copper, iodine, zinc, magnesium, manganese. At the same time, the pineapple fruit contains 86% water, protein, sugar, and citric acid. In addition, it contains more than 60 aromatic substances that give pineapple a unique smell. Bromelain, a biologically active substance contained in pineapple fruits, prevents the formation of blood clots. The enzyme bromelain in the composition improves digestion. Pineapple has antimicrobial and anti-inflammatory effects. It has diuretic, blood-thinning properties. It is useful for avitaminosis (avitaminosis), when the walls of blood vessels and vessels are brittle[11; pp.144-148,12; P.115-118]. At the same time, pineapple can be used as an ornamental plant. One of its important biological properties is drought resistance.

MATERIALS AND METHODS

The research was carried out in the greenhouse of the "Field Experimental Field" of the Gulistan State University, the laboratory of "Experimental Biology" and the Academy of the Academy of Sciences of the Republic of Uzbekistan. It was carried out in special laboratories of the Sodykov Institute of Bioorganic Chemistry. Cultivation of pineapple seedlings (Ananas somosus (L.) Merr) in vitro. For in vitro micropropagation

The following sterilizers were used for sterile and living tissue material of pineapple plants: Thimerosal 0.0005-0.001 mg/l, Hypochlorite (salts Na or Ca) 1-10 % solution, Silver nitrate (Ag NO3) 0.001-0.005 % and ethyl alcohol 30-70 % solution. Of these sterilizers, thimerosal at a concentration of 0.0005-0.001 mg/L gave satisfactory results, and different concentrations were used. In the course of the research, MS (Murashige-Skoga), DKW (Kuniyuki Nut Driver), WPM (Woody Plant Medium) nutrient media with different macro-micronutrient composition were used. Hamburg vitamins B5 -thiamine, pyridoxine, glycine; sucrose -2%, agar-agar 0.6%. The research was carried out in special laboratories of the Sodykov Institute of Bioorganic Chemistry. Cultivation of pineapple seedlings (Ananas somosus (L.) Merr) in vitro.

RESULTS AND DISCUSSION

The pineapple family belongs to the bromeliad family (Bromeliaceae) and is a perennial plant with basal leaves. Pineapple leaves have many special functions. For example, it collects water, nutrients necessary for the growth and development of plants. Plants belonging to the pineapple family are epiphytes, they live amicably in the above-ground organs of the plant. However, this is not the case with pineapple. The pineapple plant has a developed root system, through which it receives from the soil all the nutrients it needs for growth and development. Important aspects of the pineapple plant have attracted the attention of scientists around the world. This can be seen from the scientific research conducted by world scientists for many years [1; p. 550-587, 2; p. 295].

These studies have provided information about the biological and physiological characteristics of the pineapple plant and how it is propagated, and it has also been noted that pineapple can be used commercially and grown at home. In the conditions of our republic, there is no complete information about the physiological and biochemical properties and methods of pineapple breeding. Of course, during 2015-2020, professors of the Department of Biology of Gulistan State University conducted some research on the pineapple plant [3; p. 6-8, 4; p. 69-107]. In these studies, some results were found about the methods of propagation of the pineapple plant, including the possibility of growing it at home. However, methods for propagating pineapple in vitro have not been developed.

For the propagation of the pineapple plant in vitro, its variety "Maspin" was used.

The "Maspine" pineapple variety is considered one of the promising varieties not only in biotechnology and production in Malaysia. Due to the delicious fruits, the demand for this variety is very high [5; pp. 3859-3866].

Pineapple is one of the good objects of biotechnology. It is noted that it can be reproduced in vitro. It took an average of 8 years [6; pp. 296-300].

As you know, it usually takes 12-14 years to create new varieties by breeding methods. For this purpose, a complex crossing is carried out over many years. At the same time, after vegetative propagation, new varieties were created as a result of interbreed crossing [7; pp. 499-508].

For this reason, the use of biotechnological methods in the cultivation of pineapple provides an opportunity for rapid and large-scale reproduction and selection of elite plants [8; pp. 296-300].

Propagating pineapple in vitro has a number of advantages. First of all, this method allows you to propagate pineapples quickly. It is known that 30 to 1,250,000 explants can be obtained from a pineapple plant in 8 months.

Another important aspect of pineapple propagation in vitro is the ability to control the "natural flowering" process. Several factors affect the reproduction of pineapple in vitro. For pineapple propagation in vitro, a "dormant bud" was used, and with the help of a leaf, the plant can be repeatedly induced [9; pp. 117-119].

6-benzylaminopurine (BAP) solutions of 3.0 mg/L and *indoleacetic* acid (IAA) 2.0 mg/L have been reported to be effective in growing pineapple seedlings in vitro. Optimal MD2 conditions in liquid and solid media for the production of pineapple seedlings Moore, cige and Skook (MS)

obtained good results when 7.5 mg/l of BAP and 2 mg/l of naphthalic acid (NRA) were added to the culture medium [10; pp. 614-619].

It was noted that at a low concentration of BAP (5.0 mg/l) in a liquid medium, the number of sprouts increased compared to a solution with a solid medium (7.5 mg/l). The solutions used in the culture medium and their concentration are of paramount importance in the propagation of plants in vitro. It has been reported that high concentrations of cytokines adversely affect the formation of leaves in the explant, for this regeneration was obtained from the leaf and flower of the plant at a solution concentration of 1.3 µM BAP or 4.6 µM kinetin and 5.4 m.

The low concentration of the hormone in the solution led to the production of healthy pineapple sprouts to M solutions of naphthyl acetic acid.

In addition, the relatively low concentration of cytokines made it possible to obtain a large amount of regeneration from the Begonia x plant. The results of the studies showed that when a good environment was chosen, the growth of the pineapple plant was from 3 cm to 7 cm. The concentration of the BAP solution is 5 mg/l. 36 seedlings were grown at the same time. The use of the hormone (BAP) to induce in vitro regeneration has been studied in other plants, including Artemisia annua, a member of the Asteraceae family. Studies conducted on these plants have shown the formation of healthy roots within two weeks at this concentration of solution. But at a high concentration of the solution, the development of the root system slows down. It has been established that auxins (NUC, BCI, or a combination of them (NAA and BCI)) are more effective in propagating pineapple plants in vitro.

Scientific work on the cultivation of the pineapple plant was carried out on the territory of Uzbekistan. The necessary scientific materials were used in in vitro culture for the cultivation of pineapple plant seedlings. They have been studied and separated for presentation in practice.

In vitro sprouts were prepared from lateral shoots of pineapple (Ananas somosus (L.) Merr). The following sterilizers were used to sterilize the pineapple plant in vitro and obtain living tissue material:

1. *Thimerosal 0.0005-0.001 mg/l*
2. *Hypochlorite (Na or Sa salt) 1-10% solution*
3. *Silver nitrate (Ag NO3) 0.001-0.005 %*
4. *Ethyl alcohol 30-70% solution*

Before the pineapple plant (Ananas somosus (L.) Merr was placed in a sterile state, the plant growth point was checked for damage. The leaves around the pineapple stem are carefully torn off and placed in running water for a day. Remove from water and mix with laundry soap powder on a magnetic stirrer for 1 hour. Hypochlorite (salt Na or Ca) 1-10% solution is kept for 10 to 15 minutes, and thimerosal solution 0.0005-0.001 mg/l is placed for 20-25 minutes.

During this time, the premises of the laminar flow box were prepared for work, pre-prepared distilled-sterilized water and filter paper, ethyl alcohol and working tools, i.e. tweezers and a scalpel, were needed. At the end of the sterilization time, the material is taken from the thimerosal solution 0.0005 - 0.001 mg/l, immediately soaked in ethyl alcohol, placed in sterilized-distilled water, washed 3 times and laid out on sterilized filter paper to remove excess moisture. Since the stem of the pineapple is a complex stem, there is a high chance of

fungal and bacterial spores being present at the point where each leaf blade attaches to the stem. With this in mind, The sterilization process is carried out in stages.

At the first stage, the effect of solutions that transfer plant tissue to a sterile state is studied, at a high concentration, there is a high probability of tissue burning, therefore, in order to avoid damage to the tissue, it was kept in small concentrations. Therefore, the ratio of concentrations above is not explicitly indicated. The transition of the plant to a sterile state is known from the fact that fungal or bacterial spores do not multiply in the nutrient medium.

In some cases, a 1-10% solution of hypochlorite (Na or Ca salt) is sufficient and the desired result can be obtained. In our practice, each method was used to sterilize the pineapple plant, and Thimerosal 0.0005-0.001 mg/l of these sterilizers gave a satisfactory result, and different concentrations of this preparation were used in the course of the work.

During in vitro culture, the tissue to be sterilized may undergo necrosis under the influence of sterilizing agents. Especially when using mercury sterilizers, it is important to choose the right exudation time for these substances. For this purpose, a solution of ethyl alcohol was used.

After sterilization of pineapples selected for in vitro micropropagation, it is necessary to determine the optimal nutrient content for

Growing. In the course of our studies, the composition of macro-micronutrients varied;

MS (Muracigue-Skuk).

1. DKW (Kuniyuki Nut Driver)
2. WPM (Woody Plant Medium) nutrients were used:

(I) composition, NH4NO3 – 1650 mg/l, KNO3 – 1900 mg/l and CaCl2 – 0,400 mg/l.

(II) content, NH4NO3 400.0 mg/l, $Ca(NO3)_2$ - 471.26 mg/l, CaCl2 - 72.50 mg/l.

(iii) content, NH4NO3 to 1416.0 mg/L, $Ca(NO3)_2$ to 1664.0 mg/L and CaCl2 to 112.50 mg/L.

Hamburg vitamins B5-thiamine, pyridoxine, glycine; Sucrose reagents -2%, agar-agar 0.6% were used. Of the nutrients listed, the MS culture medium (Mura shige-Boredom), which was selected for further work, gave good results.

Phytohormones that are part of auxin and cytokinin during the growth of the pineapple plant in vitro;

1. Auxins include α-naphthyl acetic acid (NUC), indolylbutyric acid (IMA), adenine hemisulfate, indole acetic acid (IAA).

2. Cytokinins - benzylaminopurine (BAP), furfuralami no purine (kinetin) were used in different combinations, studied in different concentrations and the most optimal one was chosen.At the same time, the effect on the plant was studied, and the compounds were applied in different concentrations.

In the Cell Technology Laboratory, the pineapple plant was rooted and stimulated to grow until the rosette leaf touched the surface of the water.

The growing plant is separated from the root and sterilized. Once sterility was achieved, the leaves were removed and sterilized again. Cuttings were harvested, shoots of 0.2-0.5 and 1 cm in size were separated from the original shoots and planted on a nutrient medium. The color of cuttings (explant) planted on the nutrient medium was white, light yellow, light green and reddish (Fig. 1). They had an effect on cytokine compounds of phytohormones and studied their effect on the growing season of the plant. The concentration influencing the vegetation period

has been selected. A separate experiment was carried out with each joint of the genotype, since the apical part stops growing, as indicated above, and the 2nd joint is relatively exogenous. The difference from other genotypes was the shortening of the joint space, no more than 0.2 cm. They had an effect on cytokine compounds of phytohormones and studied their effect on the growing season of the plant. The concentration influencing the vegetation period has been selected. A separate experiment was carried out with each joint of the genotype, since the apical part stops growing, as indicated above, and the 2nd joint is relatively exogenous.

Fig. 1 Pineapple growth on sections of living tissues and nutrient media

The difference from other genotypes was the shortening of the joint space, no more than 0.2 cm. Living pineapple tissue is sown on a non-hormonal nutrient medium, changed every 3-5 days, clean tissue is selected (the explant, free from fungal and bacterial infection, is selected and transplanted). After 14 days, it is transferred to a harmonious environment. 16/8, i.e. 4 p.m. and 8 a.m., illumination 1200-1500 lux, air temperature 20-220 C degrees. After 15-20 days, the explant began to grow. To propagate the seedling, the growth point of the lawn was cut off, and after a while, new shoots began to grow from the side of the lawn. After the well-grown grass explant exceeds 5 cm, the root part has developed and the seedling can be planted in the soil.

CONCLUSION

In our country, vegetative propagation of pineapple is possible. It is advisable to use pineapple fruits for this. Work has been carried out to obtain sterile explants from the stems and seeds of 3 different varieties of the pineapple plant. Pineapple, imported from China, has been recommended for use in in vitro cultivation. Using 3 different reagents for sterile transfer, the

most optimal one was chosen. A solution of silver nitrate (AgNO3) of 0.001-0.005% and ethyl alcohol of 30-70% gave the best result. The expediency of using Moore, whitefish-Boredom (MS) and Gamborg (B5) nutrient media for pineapple propagation in vitro is noted. At the same time, normal conditions were: 16 o'clock in the afternoon, 8 o'clock in the morning, illumination 2000-3000 lux, room temperature 20-220C and relative humidity 60-70%. It has been found that after 21 days, 3 micro cuttings can be propagated by cuttings from micro cuttings planted on a new medium, and their growth rate accelerates during the third cloning compared to the second due to the adaptation of the plant during microcloning.

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