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INFLUENCE OF SUBSTANCES REGULATING THE GROWTH OF LEMON IN MEYER VARIETIES FROM CITRUS PLANTS AND EARLY CULTURE PROCESSES.

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Abstract:

In the Meyer variety of lemon, using L. Osbek's clonal micro-multiplication methods, the following order of use of sterilizers for sterilization of implants was found to be appropriate: 96% ethanol (5 sec) and 4% sodium hypochlorite solution (20 min). The yield under sterile conditions was 70%. A solid Murasige and Skuga (MS) nutrient medium with the addition of 1.0 mg / l benzylaminopurine and 0.5 mg / l naphthyl acetic acid was found to be optimal for in vitro propagation of the Meyer variety of lemon. Plants are planted in the soil substrate: peat, perlite, vermiculite, sand 7: 2: 3: 2 ratio. Plants adapted to the growing environment in option 1 - ½ MS +3.0 mg / l NUK plant viability was 68.2%, and in option 2 this indicator -½ MS MS = 1.0 mg / l NUK + 1.0 mg / l IUK o ' the viability of the wires was 87.5%.

Keywords: Limon, In Vitro Rooting, NAA, Media, Surface Sterilization

Abbreviations: **BAP** - 6-Benzylaminopurine**, NAA** - Naftilsirka kislotasi **MS** - Murashige and Skoog Media [Murashige and Skoog, 1962]**, DKW** - DKW Media [Driver, Kuniyuki, 1984].

INTRODUCTION.

In recent years, the world, as well as in our country, is actively researching the in vitro cultivation of virus-free planting material. The results of many years of experience have shown that the most optimal and effective way to grow seedlings from green and semi-woody cuttings in greenhouses with special film controlled microclimate. Depending on the age of the species, it is important to determine their rooting, growth and development.

Therefore, the selection of nutrient media for the cultivation of quality and virus-free healthy lemon seedlings, the expansion of scientific research on the development of optimal methods of their adaptation to environmental conditions is an urgent task of both theoretical and practical importance.

MATERIALS AND METHODS

Our research was carried out in the in vitro laboratory of the Research Institute of Horticulture, Viticulture and Enology named after Academician M. Mirzayev using MS nutrient medium of Meyer variety of lemon. The plant selected to carry out this research was brought to the laboratory in November-December 2020 from the central experimental nursery of the institute.

The leaves of the imported plants were cut. The side and tip growth points were not damaged during the cutting process.

CONDITIONS OF CULTIVATION.

During the in vitro culture, the seedlings were inoculated with low amounts of potassium nitrate-free ammonium nitrate (250 mg / l) in Murasige and Skuga (MS) and KMO-MS nutrients, but ammonium sulfate 1500 mg / l. [Murasige, Skuga, 1962], added 2.5% sucrose, 0.7% agar-agar and growth regulators - naphthyluxic acid (NOS), benzylaminopurine (BAP) and vitamins: thiamine, pyridoxine, niconic acid . The DKW [Driver, Kuniyuki, 1984] medium was used in the micropropagation phase with the phytohormones BAP and GK (hyberellinic acid). At the rooting stage, the micro-buds were placed in ½ MS medium filled with auxins - NAA 2.0 mg / l and IAA 1.0 mg / l.

Sterilization of nutrients was performed in a Market Forge sterilmatic autoclave at 120C, 1 ATM pressure, for 15 min. The implants were stored under artificial light (2790 lux) during the 14/10 period at a temperature of +20 ± 2C. Some of the plants obtained in vitro were transferred to the air conditioning room for observation in August 2021.

RESULTS AND DISCUSSION.

Micro-propagation of the Meyer variety of lemon was carried out in four stages, the isolation of which was carried out using generally accepted methods [1].

The first step is to sterilize the implants and solidify them. Includes planting in MS environment. Sterilization of the material was performed in three stages with different effects on the main sterilizing agent (Table 1).

Table 1

The main substance that affects the survival rate under sterilization conditions is the sterilizing agent.

Basic sterilizers.	Sterilization time, min.	Yield under sterile conditions,%
Hypochlorite Na ("Whiteness"), 4%	15	49.2
	20	70.1
	25	58.4

Based on the data in the table, it can be concluded from Table 1 that the treatment of the implant with 4% sodium hypochlorite solution is at least It takes 20 minutes. The difference with exposure proved to be significant for 15 minutes ($p = 0.04$, $p < 0.05$). The highest percentage of yield under sterile conditions was observed at 20 min exposure. - 70.1%. According to OV Doroshenko [2016], Approximately the same percentage of twigs was observed under sterile conditions in 10% Na hypochlorite solution for 15 min, i.e., viability was 68.2%.

Results of the development of lemon seedlings in MS solid nutrient medium Shown in Table 2. When analyzing the results of the production of Meyer variety of lemon from citrus plants in MS solid nutrient medium, it was found that the optimal medium was 1.0 mg / l BAP and 0.5 mg/l NAA added medium. The highest percentage of live implants in it is 77.5. In a KMO environment of 2.0 mg/l BAP and 2.0 mg/l added, HA showed the lowest percentage of yield of living seedlings - 47.3. The difference between the first and fourth environmental options turned out to be significant ($p = 0.03$, $p < 0.05$). The results obtained differ from the results of L.S. According to Samarina [2013] and other scientists, for lemon varieties, the yield of seedlings grown in KMO environment was high and was 84.7%.

Table 2

Yield of seedlings living in MS solid nutrient medium,%

Concentration of regulators in the nutrient medium and growth control	Living implants
MS+1.0 mg/l BAP+0.5 mg /L NUK	77.5
MS+1.0 mg/l NUK	63.6
MS+1.0 mg/l BAP	69.2
KMO+BAP 2.0 mg/l + GK 2.0 mg/l	47.3

In the first variant of the environment, the onset of microbut growth begins one week after planting, the second and third - at 1.5 weeks, in the fourth - observed in 2 weeks. The appearance of the first leaf was noted in the first variant of the medium after 2–3 weeks, and after 3–4 weeks - in other variants. Growth of seedlings stopped 5–7 weeks after planting, regardless of the medium variant. The development of the implant in the MS nutrient medium is shown in Figure 1.

The second stage was the activation of bud growth, which was then carried out in a DKW environment. After a week, recovery of growth was observed, after 4-5 weeks, the formation of micro-buds began. From each mother plant transplant we were able to obtain micro-tumors in 2.45 ± 0.78 conditions.

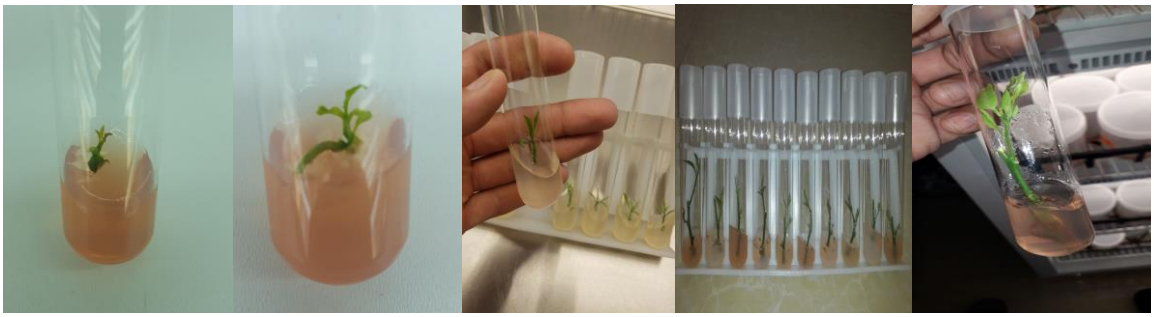

Figure 1. Propagation of lemons in the nutrient medium of Meyer variety
MS + 1.0 mg / l BAP + 0.5 mg / l NAA.

Concentration of mineral salts and sucrose (½MS), used with half of the MS nutrient medium for rooting of plant seedlings, in two versions:

NAA with the addition of 3.0 mg / l and 1.0 mg / l NAA + 1.0 mg / l IAA. The results show the effect of aoxins on rhizogenesis in Figure 2.

To evaluate the effectiveness of rhizogenesis, the frequency of rooting, i.e. the number of roots, was calculated in the first stage of development. During the first development of the environment, 65.1% of the rooted micro-buds and the root length was 3.6 ± 0.81 cm and the number was 2.6 ± 0.49 PCS. In the second phase of our study, it was observed that the root length was 62.8% and the roots of rooted micro-plants were 4.2 ± 1.16 cm and the number was 1.2 ± 0.34. Using data from Samarina [2013] and other scientists, we studied the effects of auxin and cytokines with the best 1.0–2.0 mg / l NAA and 1.0 mg / l added for rooting of lemon varieties. As a result of the study, rooted buds ranged from 83.7% to 95.1%. The length of the roots was 4.2-5.2 cm, and the number was 3.1-3.8.

In vitro plants were transferred to a closed room for acclimatization. Plants are planted in the soil substrate: peat, perlite, vermiculite, sand in the ratio 7: 2: 3: 2. Plants adapted to the growing environment in option 1 - ½ MS +3.0 mg / l NUK plant viability was 68.2%, and in option 2 this indicator -½ MS MS = 1.0 mg / l NUK + 1.0 mg / l IUK o ' the viability of the wires was 87.5%.

Figure 2. Effects of oxides on rhizogenesis Micro-cut Meyer lemon.

CONCLUSION

As a result of our research, the following scheme of microclonal propagation of the Meyer variety of lemon can be proposed. With the addition of growth regulators 1.0 mg / l BAP and 0.5 mg / l NAA, you can use MS, a solid nutrient medium. In the micropropagation phase, good results were obtained by adding BAP 2.0 mg / l and HA 2.0 mg / l in DKW 60C medium. Nutritional medium for the rooting stage ½MS 1.0 mg / l NAA + 1.0 mg / l IAA auxin double concentration of mineral salts and sucrose is proposed. To transfer to the soil in an enclosed room, you can use a soil substrate of the following composition: peat, perlite, vermiculite, sand in a ratio of 7: 2: 3: 2.

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