"ROOT SYSTEM FORMATION AND ADAPTATION TO EX VITRO CONDITIONS IN REGENERANT LAGOCHILUS INEBRIANS BUNGE SAMPLES"

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Abstract
When leaf axillary buds and leaf fragments were used as primary explants during experiments a combination of BAP (5 mg / l) + NAA (0.4 mg / l) in the MS culture medium was found to be the optimal indicator for formation of callus tissue. It was also found that the intensity of callus tissue formation in the combination of BAP (3-4 mg / l) + NAA (0.4 mg / l) in the MS medium was high when using buds and leaf fragments located in the tip of stem .

In experiments, it was detected that the intensity of root formation was relatively high in combinations of NAA (0.5-3 mg / l) + IBA (0.5-3 mg / l) or NAA (0.5-3 mg / l) + IBA (0.5-3 mg / l)+ activated charcoal (3 g / l) in ½ MS medium. The average rooting frequency in the samples was 81.3%. 100% rooting in Lagochilus inebriance shoots was obtained only on the basis of ½ BDS and ½ MS medium in the presence of auxins NAA, IAA, IBA at a concentration of 5.0 µM.

Keywords: Lagochilus Inebriance leaf axillary buds, MS medium, callus tissue, average rooting frequency

Today in our republic, special attention is paid to the inventory of medicinal plant species, the assessment of their resources, the creation of technologies for the reproduction of promising species and the production of natural medicines based on local plant raw materials. [7-10] On the basis of the program measures implemented in this direction, certain results have been achieved. In particular, substances have been isolated from local medicinal plant raw materials, from which drugs for the treatment of viral, infectious, cardiovascular diseases have been developed, the raw material base of medicinal plants based on farm zones

has been strengthened.[10-18] The strategy of actions for the further development of the Republic of Uzbekistan defines the tasks of "further development of the pharmaceutical industry, provision of the population and medical institutions with affordable, high-quality medicines." Based on these tasks, including recommendations for the breeding and production of one of the medicinal plants, the protected species Lagochilus inebrians, acquires important scientific and practical significance.

Accordingly, the purpose of this research work is: to obtain and put into practice microclonal reproduction, storage and pathogenic seedlings of a promising medicinal plant - Lagochilus inebrians bunge in vitro.

The results obtained and their analysis

The following is an analytical review of the results of studies on the adaptation of non-pathogenic Lagochilus Inebrians Bunge seedlings obtained in vitro to ex vitro conditions.

When growing the plant in culture in vitro, 6-BAP - 1-3 mg/l; GC - 0.5-2.0 mg/l; NSC - 0.1-0.3 mg/l or BMI - 0.1-0.3 mg/l and a complex of vitamins were added to the nutrient medium. To stimulate the process of rhizogenesis in the culture medium, the concentration of macronutrients and sucrose was reduced. [18-22] Auxin was added to the feed medium at a concentration of 0.2-2 mg/l or incubated in an explant solution of BCI (50 mg/l) for 18 hours and at the next stage was sown into the feed medium without the addition of phytohormone (pH =5.5-5.7). In experiments, tidiazuron (N-phenyl-N'-(1,2,3-thiadiazole-5-Yl urea); TDZ), zeatin (1.65-5 mg/L), 2,4-D, ISC (0.1-0.5 mg/L) in various combinations were tested as a stimulant. The used phytohormones and vitamin complexes belong to the production of the company "Serva" (Germany). Cultivation of the culture was carried out in the laboratory for 16 hours in the light mode of 3000 lux per day at a temperature t = 24-26 ° C, humidity 60%.[4-9]

Experiments have shown that under the conditions of using a combination of BAP (5 mg/l)+NAA (0.04 mg/l) in the composition of the MS nutrient medium under in vitro conditions, the intensity of formation of callus tissue from the leaf sinus is higher than under BAP conditions (2 mg/l)+NAA (0.1 mg/l)+GA3 (30.5 mg/l) in combination, it was found that growths in the maximum amount and length are formed in the culture, as well as a high intensity of the root formation process if IBA (3 mg/l) is added to the incubation medium [22]. It is noted that the intensity of propagation of shoots is higher in the case when element 6 is added to the composition of the nutrient medium.

In vitro, phytohormones are used to form callus tissue. The correct choice of phytohormone types and their combinations in the composition of the nutrient medium at this stage is important to ensure the optimality of the process. In our studies, combinations of BAP (1-5 mg/l) were tested+NAA (0.4 mg/L) and MS+2.4-D (1-5 mg/L) in the MS nutrient medium for the induction of callus tissue in various initial explants listed above. In the experiments, plant extracts were placed in glass containers (275 ml) with a 25 ml nutrient medium, while the culture was in standard conditions (+20...Incubate for 4 weeks at a temperature of + 25 ° C, 16 hours of illumination during the day and 8 hours in the dark).[12-14]

In studies, it was found that in the nutrient medium, the intensity of development and proliferation of callus tissue in combination with BAP (0.5-2 mg/l)+NAA (0.25-1.5 mg/L) occurs at a relatively high level.

In studies, it was noted that in a nutrient medium, MS has a higher intensity of tumor formation in a combination of BAP (0.5-2 mg/l)+NAA (0.1-1 mg/l)+kinetin (0.5-1 mg/l), GA3 (0.5 mg/l)+adenine sulfate (40 mg/l).Pp. 16-22. Studies show a high intensity of root formation in combination with NAA (0.5-3 mg/l)+ IBA (0.5-3 mg/l), as well as NAA (0.5-3 mg/l)+ IBA (0.5-3 mg/l)+activated carbon (3 g / l) in a nutrient medium for ½ ms.

L.Inebrians under in vitro conditions by microcloning, the intensity of the stages of the reproduction process depends on such factors as (formation and proliferation of callus tissue; formation of growths, root formation), the type of explant, the method of sterilization of the explant, combinations of phytohormones used in the nutrient medium. The experiments revealed optimal parameters for the formation of callus tissue of the BAP combination (5 mg/l)+NAA (0.4 mg/l) as part of the MS nutrient medium in a variant in which buds and leaf fragments of the axillary cavities of the leaves were used as the initial explant. It was also found that when using shoots and leaf fragments located at the ends of stems, the intensity of callus tissue formation is higher in the composition of the MS nutrient medium in combination with BAP (3-4 mg/l)+NAA (0.4 mg/l).[4-8]

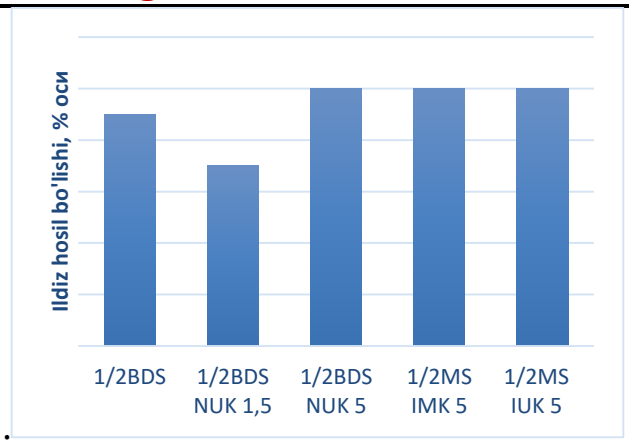
In experiments, it was found that the proliferation of callus tissues is carried out optimally in a combination of VAR (1 mg/l)+NAA (1 mg/l) as part of the MS nutrient medium. It was also noted that the intensity of root formation is relatively high with the combination of BAP (2 mg/l) +NA (0.1 mg/l)+GA3 (0.5 mg/l) (Fig.1).

Fig.1. Intensity of root formation under in vitro conditions of Lagochilus inebrians

Seedlings obtained under in vitro conditions were gradually transferred to the soil. To do this, the plant obtained in vitro is planted in a substrate in a greenhouse. The substrate consists of two layers of sand and sawdust. The top of the substrate is an ordinary substrate, sawdust is laid on the surface layer.

Under in vitro conditions, it was found that the proliferation of callus tissues L.Inebrians optimally carried out in the MS nutrient medium in a combination of BAP (1 mg/l)+NAA (1 mg/l). In the BAP combination variant (2 mg/l)+NAA (0.1 mg/l)+GA3 (0.5 mg/l) there is a relatively high intensity of root formation. It was noted that in the conditions of the WPM nutrient medium, in the presence of a combination of kinetin (2.3-18.4 microns) +1-naphthalene acetic acid (0.54 microns), the regeneration intensity is higher than in the MS. L nutrient medium. For the introduction of L.Inebrians, in vitro reproduction methods were created.

It should be noted that the plants in the studied specimens easily take root in an environment with added hormone and in a non-hormonal environment. The average rooting frequency in all tested samples was 81.3% (Fig.9). In L.Inebrians, 100% rooting in the microclimates of inbreeders was obtained only in the presence of auxins at a concentration of 5.0 microns on mineral bases of ½ BDS and ½ MS - NAA, IAA, IBA. The most effective means for stimulating rhizogenesis in L.Inebrians was ½ B5 filled with 5.0 microns of NAA (Fig.2)



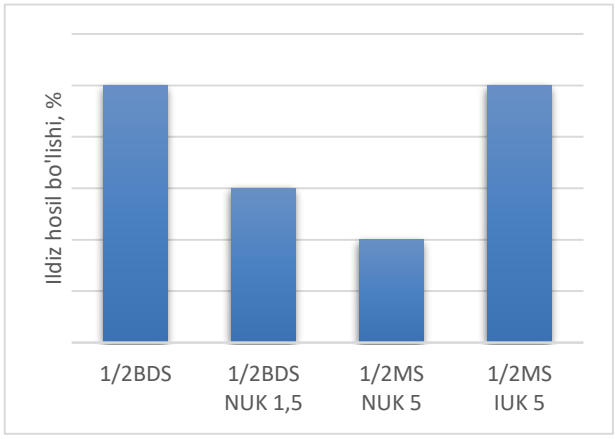


Fig.2. Samples grown in laboratory sterile conditions (a) and grown in greenhouse conditions (b)

It can only be shown that the presence of 5.0 microns of NAA, IAA or IMC in the nutrient medium compared to a medium without hormones with a low content of NAA (1.5 microns) the formation of roots in the medium was observed .

Conclusions

1.The optimal environment was chosen for the natural reproduction of the studied Lagochilus inebriance samples; it is more effective to introduce 0.1 microns of BAP into the nutrient medium according to the B5 recipe. For Lagochilus inebriance samples grown under laboratory conditions, use a BDS medium containing 5.0. for samples grown in greenhouse conditions, the optimal medium is nutrient medium B5, supplemented with 0.4 microns of bap, 3.2 microns of NAA and 2.3 microns of IAA. The most effective cytokinin in the proliferation phase is BAP compared to TDZ and Kinetin.

2. For the first time, the influence of the mineral composition of the medium on the path of morphogenesis of Lagochilus inebriance was shown using the same

combination of growth regulators: growing segments on B5 causes direct hemogenesis, and the use of BDS causes indirect hemorrhogenesis.

3. Growing in the rooting phase at low positive temperatures (+7 ° C) promotes more intensive rhizogenesis and plant growth, as well as accelerates the germination and development of restored plants, followed by their transfer to ex vitro conditions. The most optimal mode of adaptation of the microclimate of the studied specimens is adaptation in a greenhouse using a mixture of crushed coconut fiber and sand as a substrate (3:1), which provides a high frequency of adaptation (up to 82.7%).

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