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Microbial Stimulation of Growth of Lucerne

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Abstract
From the soil samples outside the areas of intensive agriculture, were allocated 145 isolates: 80 cultures growing on medium nutrient agar, 28 – on 79 medium for fixing microorganisms and 37 isolates on MRS medium, by forming zones of hydrolysis of chalk. The influence of selected microorganisms were researched on seed germination and seedling growth of lucerne. Stimulation of the growth of lucerne by some cultures reached 35% (5, R11) - 45% (1, 9, R5, R28) compared with the control.
Key words: biopesticide, growth stimulation, lucerne, plant protection.

I. Introduction

In biological condition farming focuses on legumes, which are able to create organic nitrogen through its symbiotic apparatus. Using of legumes, especially lucerne, is essential, as is the culture of multi-application, which is used for food in the form of green mass, hay and silage, as a raw material compound feed and irreplaceable component mixtures to create cultivated pastures. Properties of alfalfa as a nitrogen storage, reducing agent and water-structuring physical characteristics of soils, implemented for inclusion in crop rotations as a precursor. Its plants are rich in protein, carbohydrates, minerals, vitamins and other biologically active substances. Alfalfa has a very wide distribution area due to the high yield and low cost derived from her feed [1]. Introduction alfalfa in all types of crop rotation has a huge impact on the growth of productivity subsequent crops, improving the quality of derived products and improvement of soil fertility, which is especially important in arid climates of southern Kazakhstan as a zone of risky agriculture, in which two-thirds of households are unstable area, and the third - the lack of moisture. Of 10 years, typically 5-6 are dry [2,3]. Therefore, based on the technology of high and stable yields of forage crops should be developed system of feed production, which reduces to the use of various remedies the crop.

Application of biological solutions for this problem takes priority, they do not carry a negative impact on the system of the entire plant [1]. However, until now in conditions of South Kazakhstan untested local strains of bacteria promising to enhance the growth promoting activity in order to improve productivity and quality of Lucerne forage.

Particular interest is the use of seperate strains of microorganisms to find but the impact of the process on the whole plant. We out the selection and introduction of microorganisms having antagonistic to phytopathogenic microflora and growth stimulating properties, especially in the early stages of plant development.

II. Materials and Methods

To isolate microorganisms, the soil samples were taken outside the zone of intensive land use. Microorganisms were isolated on media nutrient agar (MPA), MRS with chalk and on 79 medium to nitrogen-fixing bacteria. For research were used lucerne seeds varieties "Akmal", from Zhambyl branch "Kazakh Research Institute of Agriculture and Plant." Plant seeds were soaked for 1 h in suspension of isolated microorganisms (concentration 1.times.10.sup.8 cells / ml) then were seeded in soil, placed in a Petri dish of 20 pieces of 3 replicates. Test seeds incubated for 1 hour before seeding sterile tap water. After 7 days, we determined the quality of germinated seeds, length seedling, root length of each plant alfalfa. Then the roots were separated from the green part, and dried at room temperature and then identified the average total dry weight of roots and greens. Statistical processing of the results of research carried out by standard methods, using Student's t test for the level of $p < 0,05$.

Environment

Environment for saprophytic microorganisms Nutrient agar contains:

Agar – 15,00 g; peptone – 5,00 g; sodium chloride – 5,00 g; beef extract – 1,50 g; yeast extract – 1,50 g; distilled water – 1 ltr.

Environment for lactobacilli MRS agar contains:

Dextrose – 20,00g; agar – 15,00 g; proteose peptone №3 g; beef extract – 10,00 g;yeast extract – 5,00 g; sodium acetate – 5,00 g; ammonium citrate – 2,00 g; potassium phosphate dibasic – 2,00 g;

sorbitan monooleate complex – 1,00 g; magnesium sulphate – 0,10 g; manganese sulphate – 0,05 g; distilled water – 1 ltr.

Environment for environment for fixing microorganisms №79 contains:

NaCl – 0,10 g; K2HPO4 – 0,50 g; MgSO4 – 0,20 g; CaCO3 – 0,01 g; mannite – 10,00 g; yeast extract – 1,00 g; agar – 20,00 g; distilled water – 1 ltr.

III. Results and Discussion

From the soil samples of outside the zone of intensive land use, were allocated 145 isolates: 80 cultures growing on medium MPA, 28 medium 79 on for fixing microorganisms and 37 isolates on medium MRS, forming a zone of hydrolysis of chalk. The influence of selected microorganisms on seed germination and seedling growth of alfalfa. Of the 145 isolates, 9 isolates inhibited germination of seeds, 3 had no effect and 133 stimulated seedling growth (from 9 to 45%). Seed germination of Lucerne by treatment of the isolated microorganisms was increased to 35% (5, R11) - 45% (1, 9, R5, R28) (Fig. 1).

Figure 1 - Stimulation of germination and growth of lucerne seeds

To create a biological product that stimulates plant growth, greater interest in the conditions of the Republic of Kazakhstan with the often arid climate, are microorganisms that not only stimulate the growth of the green parts of plants, but also increase the length of the roots. Therefore, the development of our biological product for stimulating the growth of microorganisms selected safflower not only stimulate the growth of the green part (1, 2, 5, 9, 15, 20, R1, R3, R5, R11, R26, R28, M1, M7) but causing extension of the root system (1, 3, 5, 7, 8, R5, R11, R28, R35, R69, M6). The study was selected few cultures that stimulated the growth of the entire plant (1, 5, 9, R5, R11, R28) (Table 1).

Table 1 Effect of microorganisms isolated from soil on germination and seedling growth of lucerne

Culture	Germination,%	The length of the green parts	Length of roots	Dry weight per plant greenery	Dry weight of roots per plant
		cm	cm	g	g
Control	55	4,5±0,4	3,0±0,2	0,023	0,015
1	95	4,8±0,1	3,7±0,4	0,034	0,021
2	85	5,5±0,6	2,4±0,5	0,033	0,016
3	70	4,2±0,4	2,4±0,2	0,026	0,014
5	90	4,9±0,7	3,3±0,6	0,038	0,023
7	65	3,5±0,2	2,7±0,2	0,029	0,017
8	75	4,4±0,4	2,4±0,5	0,025	0,014
9	100	4,8±0,2	3,8±0,6	0,039	0,022
15	35	5,2±0,1	1,8±0,6	0,022	0,019
20	50	5,3±0,6	2,2±0,2	0,023	0,018
R5	95	5,1±0,4	3,8±0,3	0,036	0,023
R11	90	4,7±1,1	3,4±0,2	0,039	0,021
R26	75	3,8±0,5	2,5±0,4	0,027	0,016

R28	95	4,9±0,7	3,5±0,1	0,033	0,024
R35	75	3,1±0,6	3,2±0,1	0,030	0,014
R51	65	3,4±0,6	2,0±0,1	0,030	0,016
R55	65	2,8±0,1	2,1±0,6	0,024	0,017
R69	70	3,4±0,6	2,0±0,2	0,027	0,015
M1	65	4,8±0,3	2,4±0,8	0,026	0,016
M6	60	4,3±0,4	2,1±0,4	0,025	0,017
M7	50	4,8±0,1	2,6±0,2	0,027	0,016
Notes: 1 R- microorganisms isolated from the Nutrient agar (МПА) 2 M – microorganisms isolated from the MRS agar 3 without designation microorganisms isolated from medium number №79					

All active strains of bacteria belong to the genus of *Azotobacter*. In conclusion we can say that from the active strains will be compiled to create associations based on their high-performance environmentally safe drugs to stimulate the growth of lucerne.

The next step will be identify the ability of bacteria, stimulating the growth of lucerne seeds, inhibit the development of pathogenic and opportunistic fungal microorganisms isolated from the surface of lucerne seed in the formulation laboratory experiments. Selected cultures will comprise, which developed in the laboratory of complex multicomponent bacterial preparation comprising groups of micro-organisms of various physiological stimulating the growth of lucerne and other crops.

References

[1] A.A. Sinitsyna. *Protection against pests of lucerne seed and the preservation of its pollinators in terms of the Krasnodar Territory*, PhD., 2000, V.167.

[2] E.A. Akhmetov, N.N. Karmanova, S.R. Karbaeva, B.K. Asubaev. *Economic and social geography of Kazakhstan, Methodological guidance*, Almaty, 2005, V.93

[3] T.L. Kazanovska. *Teleconumicatii and social geography of Kazakhstan: methodological guidance, Methodological guidance*, 2013, V.105

[4] M.V. Shternshis, F.S. Jalilov. *Biologics in plant protection*. *Novosibirsk*, 2000. – V.128

[5] M.G. Sokolova, G.P. Akimov. *Impact on plant phytohormones synthesized rhizosphere bacteria* // *Applied Biochemistry and Microbiology.* 2011. T. 47. № 3. P. 302-307

[6] J.T. Botbaeva, I.E. Mustafina, A.Z. Aiupova, A.P. Nauanova. *Selection of strains of the genus Bacillus with antifungal activity to create effective biologics* // *Herald Karaganda University. Biology Series. Medicine*. 2011. № 2 (62). P.29-33