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INTERACTION OF HIGHLY BIOTECHNOLOGICAL STRAINS OF MICROORGANISMS WITH THE ROOT SURFACE OF PLANTS

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Abstract

The soil microbiome of agricultural crops is a fundamental element of agroecosystems, determining the growth, development and stability of plants in the agrocenosis. Interactions between plants and microorganisms are the result of long-term coevolution, which contributed to the formation of stable, mutually beneficial associations. One of the key strategies for stable colonization of plants is the formation of biofilms by microorganisms on the surface of roots.

Biofilms are organized communities of microorganisms immersed in a self-synthesized extracellular matrix. This structure provides microbiomes with protection from negative environmental influences and reactions of the plant's immune system. An in-depth understanding of the processes of biofilm formation allows us to develop new strategies for plant-microorganism interactions to increase plant resistance to abiotic stresses, including drought, salinity, heavy metal pollution, and also contributes to the formation of stable and highly productive agroecosystems.

During the experiment, it was found that the level of formation and maturity of biofilms on the roots of seedlings of different test crops depended to a large extent on their species, while the influence of the studied strains was less pronounced. After 48 hours of exposure, biofilm formation was observed on the roots of all studied plants, although the level of their maturity varied significantly. In particular, the minimum density of biofilms was characteristic of representatives of the Fabaceae family, while the maximum was characteristic of crops of the Poaceae family.

Keywords: *biofilms, adhesion, roots, soil, microbiome, community of microorganisms, consortia.*

All plant-microorganism interactions are not random and are the result of long-term coevolution, often leading to associations in which the host (plant) and its microbiota cooperate in a mutually beneficial manner [1]. Recently, data have been published in the scientific literature indicating cases of extreme dependence of host plants on the microbiome [2, 3] to the complete absence of any interaction with a particular microbiota [4].

However, the first step in establishing an interaction between microorganisms and plants is the adhesion of microorganisms to plant roots. This process involves weak hydrophobic and electrostatic interactions, which can progress to more stable attachment induced by specific environmental signals

and the host plant [5]. The next step in this interaction is the expression of genes responsible for the synthesis of exopolysaccharides with subsequent biofilm formation. The formed biofilms have the appearance of mucous complex-organized membranes around the roots, which provide a long-term and favorable environment not only for the attachment of microorganisms to plants, but also for their existence [6].

Given all of the above, the purpose of our study was to identify the ability to form biofilms on the roots of various agricultural crops by strains of microorganisms that have high biotechnological characteristics and are promising for production.

The study was conducted in the Laboratory of Microorganism Ecology, Department of Agroecology and Biosafety, Institute of Agroecology and Environmental Management of the National Academy of Sciences of Ukraine (Kyiv, Ukraine). In the laboratory experiment, the adhesive properties of 5 strains of microorganisms (Bacillus subtilis K4, B. subtilis F6, B. subtilis D24, B. subtilis M7, B. subtilis A9) isolated from the rhizosphere of spring wheat in 2024 were determined. The following test plants were used for the experiment: spring wheat Tokata, barley Sebastian, corn Khorol, cucumber Rodnychok F1, sweet pepper Ivanhoe, tomato Sanka, zucchini Eleonora F1, beans Shahinya, chickpeas Triumph, vegetable peas Dragon, seed peas Maecenas, pumpkin West.

The ability to form biofilms on plant surfaces was studied by the express method 48 hours after contact between the seedling and the microorganism. The level of biofilm formation was assessed using the plus system, where 0 - no adhesion of bacterial cells was recorded, (++++) - biofilm of medium or greater thickness, information is specified in Table 1 [7, 8]. 10 seedlings from each study variant were studied; biofilm formation was assessed in 10 fields of view. Data processing was carried out using MS Excel, Statistica 12.0 software.

In laboratory studies, the level of biofilm formation on the roots of test crop seedlings was determined by the action of test microorganisms presented in Table 1.

Table 1. Biofilm formation by strains of microorganisms on seedlings of agricultural crops

№	Option of research	*Bacillus subtilis*				
		A9	**K4**	**M7**	**D24**	**F6**
1	Spring wheat	++++	++++	++++	++++	++++
2	Barley	++++	+	++++	+++	+++
3	Corn	++++	+	++++	++++	++++
4	Beans	+	++	+	+	++
5	Chickpeas	+	++	+	++	++
6	Vegetable peas	+	++	+	++	++
7	Seed peas	+	+	+	++	++
8	Tomato	++++	++++	++	++++	++++
9	Sweet pepper	+++	+++	+	++++	+++
10	Cucumber	++	+++	+	++++	++++
11	Zucchini	+++	+	++	+++	++
12	Pumpkin	++	+	+	++	++

During the research, it was found that the level of formation largely depended on both the species of the test culture and the strain under study. After 48 hours of study, biofilm formation was observed on the roots of all the tested test cultures, but with different levels of formation. The lowest level of biofilm formation was characteristic of test cultures belonging to the legume and cucurbit families. However, the formation of biofilms of medium and greater thickness was observed when the studied strains were used on cereal and solanaceous crops.

In general, the most intensive formation of biofilm structures was observed on seedlings of wheat (Triticum aestivum), corn (Zea mays), barley (Hordeum vulgare) and tomatoes (Solanum lycopersicum), sweet pepper (Capsicum annuum L.), which indicates a high level of microbial colonization under optimal microbiological conditions and is most likely associated with the activity of primary roots (high secretory capacity) of cereal crops. It was also found that microorganisms exhibit a high ability to attach to the surface of the rhizodermis due to the production of extracellular polysaccharides, which contribute to the formation of biofilms and increase the resistance of the microbial population to adverse conditions. Of particular note are the results on the formation of biofilms on the roots of legumes - peas, field peas, chickpeas and beans. It was found that these test cultures demonstrated a lower density of biofilms, which is most likely due to the fact that the bacterial test strains are not symbiotic nitrogen fixers, but belong to free-living microorganisms.

According to the results of studies of the strains of microorganisms, it was found that all bacterial strains were able to form colonies and biofilms of varying degrees of thickness on the roots of seedlings, which indicates their ability to be introduced into the root zone of plants during the formation of the microbiome.

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ВЗАЄМОДІЯ ВИСОКОБІОТЕХНОЛОГІЧНИХ ШТАМІВ МІКРООРГАНІЗМІВ ІЗ КОРЕНЕВОЮ ПОВЕРХНЕЮ РОСЛИН

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Анотація

Ґрунтовий мікробіом сільськогосподарських культур є фундаментальним елементом агроекосистем, що визначає ріст, розвиток та стабільність рослин в агроценозі. Взаємодії між рослинами та мікроорганізмами є результатом тривалої коеволюції, яка сприяла формуванню стабільних, взаємовигідних асоціацій. Однією з ключових стратегій стабільної колонізації рослин є формування біоплівок мікроорганізмами на поверхні коренів.

Біоплівки – це організовані спільноти мікроорганізмів, занурені в самосинтезований позаклітинний матрикс. Така структура забезпечує мікробіомам захист від негативних впливів навколишнього середовища та реакцій імунної системи рослини. Глибоке розуміння процесів формування біоплівки дозволяє розробляти нові стратегії взаємодії рослин і мікроорганізмів для підвищення стійкості рослин до абіотичних стресів, включаючи посуху, засолення, забруднення важкими металами, а також сприяє формуванню стабільних та високопродуктивних агроекосистем.

Під час експерименту було виявлено, що рівень формування та зрілості біоплівок на коренях розсади різних тестових культур значною мірою залежав від їх виду, тоді як вплив досліджуваних штамів був менш вираженим. Після 48 годин експозиції формування біоплівок спостерігалося на коренях усіх досліджуваних рослин, хоча рівень їхньої зрілості суттєво варіювався. Зокрема, мінімальна щільність біоплівок була характерна для представників родини Fabaceae, тоді як максимальна – для культур родини Poaceae.

Ключові слова: біоплівки, адгезія, корені, ґрунт, мікробіом, угруповання мікроорганізмів, консорції.