BIOTECHNOLOGY OF PROBIOTIC MICROORGANISMS' MICROENCAPSULATION

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Introduction. The microbiome plays an important role in various physiological processes, including digestion, metabolism, immune responses, biosynthesis of many compounds, elimination of toxins, regulation of gut-brain axis function, and even disease pathogenesis. Most of these microbial communities in the gut are influenced by the mode of birth, feeding of the child, genetic background, and lifestyle, including diet, exercises, stress, medications, and the general health of the host. Intestinal microbial populations can vary significantly from person to person, including healthy individuals. Unfavorable changes in the microbial composition, and hence in its functions, are a characteristic of dysbiosis. In COVID-19, the gut microbiome has been shown to be disrupted after SARS-CoV-2 infection. Existence of a gut-lung axis in which the gut microbiota is metabolically capable of influencing lung function is evidenced.

Discussion. Modern approaches, such as the introduction of pro-, pre-, para-, synbiotics and their other derivatives, along with transplantation of fecal microbiota, can restore the disturbed microbiota of the gastrointestinal tract (GIT). There is currently growing interest in functional innovative food products as ideal carriers for probiotic microorganisms. However, many commercial probiotic products are ineffective because the beneficial bacteria they contain do not survive food processing, storage, and passage through the upper gastrointestinal tract. Therefore, modern effective strategies are needed to improve the stability of probiotic microorganisms, both in food products or medicines and during their passage through the human gastrointestinal tract. One of such strategies is microencapsulation as an effective means of protecting probiotics under aggressive conditions. Microcapsules allow programmed release under certain conditions. An effective microencapsulation system maintains the stability of probiotic microorganisms during storage, protects them from aggressive conditions in the upper GI tract, releases them in the large intestine, and then promotes their colonization of mucous surfaces. To achieve better protection and controlled release of probiotics, alginate microgels are widely used.

There are several types of microgels for the of probiotic microorganisms' delivery:

1. Simple microgels usually consist of small spherical particles containing a network of cross-linked biopolymers inside, with pores filled with an aqueous solution.

2. Core-shell microgels. The performance of microgels can be improved by coating them with one or more layers of biopolymer.

3. Biopolymer complex microgels. It is possible to use both one biopolymer to create microgels, and it is advisable to combine two or more biopolymers to improve their stability or functionality. Microgels can be made by combining biopolymers using complex coacervation (mixing a negatively charged biopolymer with a positively charged one).

4. Microgels resistant to gastrointestinal conditions. An interesting approach has been developed to increase the viability of encapsulated probiotics during the gastrointestinal tract. It consists in controlling the pore size and internal pH of the microgels. Many probiotics are inactivated when exposed to gastric juice due to its high acidity and the presence of digestive enzymes. This effect can be reduced if the internal pH of the microgels remains neutral in the stomach, and the impossibility of penetration of digestive enzymes into the microgel is ensured. Simple microgels are not very effective in protecting probiotics in GI conditions because small hydrogen ions (H+) and digestive enzymes can easily diffuse into them due to the relatively large pore size of the biopolymer network.

5. Nutrient supplemented microgels. Another approach to increase the survival of encapsulated probiotics during storage and in the gastrointestinal tract is to provide them with a sufficient amount of beneficial nutrients - prebiotics. It is known that the encapsulation of prebiotics in the microgel core improves the viability of probiotic microorganisms.

Biomaterials used to encapsulate probiotics include natural and synthetic polymers. These biomaterials are in direct contact with living cells. For this reason, criteria have been developed for their selection:

- physical and chemical properties (chemical composition, morphology, mechanical strength, stability in the gastrointestinal tract and intestinal fluid;
- toxicological analysis;
- manufacturing and sterilization processes.

The most common biomaterial used to encapsulate probiotics is alginate. Other supporting biomaterials include carrageenan, gelatin, chitosan, whey proteins, cellulose acetate phthalate, locust bean gum, and starches.

The selection of a suitable biomaterial is a preliminary study requiring careful methodological research. The search for new materials for encapsulation is of paramount importance. These materials must meet the requirements of non-toxicity, resistance to gastric acidity and compatibility with cells of probiotic microorganisms.

To evaluate the effectiveness of a probiotic delivery system, it is very important to characterize its structural organization, physicochemical properties, functional characteristics, and sensitivity of probiotic parameters. Statistics are performed by a combination of *in vitro* and *in vivo* models. *In vitro* models for rapid detection of various compounds, but they often do not accurately model the complex processes that increase them within the human gut. *In vivo* methods are more expensive and time-consuming, but they allow more accurate assessment of the potential effectiveness of the delivery system in rare cases.

Conclusions. Increased interest in understanding the importance of the intestinal microbiota in normal and pathological conditions, as well as the increasing number of negative factors affecting the microbiota of various biotopes of the host organism, the development of systems for the oral delivery of microencapsulated active viable probiotic microorganisms to the large intestine is one of the important tasks of modern biotechnology.

Microencapsulation of probiotics into polymeric microcapsules successfully protects them from the harsh and changing conditions of the gastrointestinal tract, and also allows the delivery of living cells of probiotic microorganisms without losing their functionality to the target biotope of the host organism. The microcapsules also protect the probiotic cells during the stabilization process and storage over a wide range of temperatures, and therefore can significantly extend the shelf life of the final product. Joint microencapsulation of prebiotics with probiotic microorganisms can additionally increase the survival rate of the latter during storage and in the gastrointestinal tract.

It has been shown that Alginate is an ideal biopolymer material for probiotic microorganisms' microencapsulation for their targeted delivery to the intestine. Alginate is biocompatible, environmentally friendly, has a low cost, and most importantly is characterized by ease of use.

Thus, the development of biotechnologies for pro- and synbiotics, as well as functional foods enriched with microencapsulated probiotic microorganisms as effective means of maintaining and restoring the intestinal microbiota, is one of the urgent and important tasks of modern science.