

## **ISOLATION OF STRAINS AND DEVELOPMENT OF STARTER CULTURES FOR FERMENTED MILK PRODUCTS**

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### **Abstract**

Fermented milk products are widely consumed because of their high nutritional value, probiotic properties, and technological versatility. The quality and stability of these products largely depend on the characteristics of lactic acid bacteria used as starter cultures. These microorganisms play a key role in rapid acidification of milk, formation of texture and aroma compounds, and inhibition of undesirable microflora during fermentation.

The formulation of growth medium composition for lactic acid bacteria is an important step of starter culture technology development. It should be cost-effective and nutrient-rich, allowing bacteria to reach high cell density. For industrial use, starter cultures must be preserved in a way that maintains their viability and metabolic activity for long periods. Freeze-drying (lyophilization) is one of the most effective methods for microbial preservation. However, the dehydration process can cause cellular damage, including membrane disruption and protein denaturation, which may reduce the survival rate of bacteria after drying.

Therefore, the composition of the growth medium and the use of effective cryoprotective systems are critical factors influencing the properties of bacterial culture. Developing simple, food-grade, and technologically accessible growth media and cryoprotective formulations is an important task, especially with the increasing demand for locally produced starter cultures in the dairy industry.

### **Keywords**

Lactic acid bacteria, fermented milk products, starter cultures, growth media, freeze-drying.

## **ВЫДЕЛЕНИЕ ШТАММОВ И РАЗРАБОТКА СТАРТОВЫХ КУЛЬТУР ДЛЯ ФЕРМЕНТИРОВАННЫХ МОЛОЧНЫХ ПРОДУКТОВ**

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### **Аннотация**

Ферментированные молочные продукты пользуются высокой популярностью у потребителей, благодаря высокой пищевой ценности, пробиотическим свойствам и разнообразию ассортимента. Стабильность качества этих продуктов во многом зависит от свойств молочнокислых бактерий, входящих в состав заквасок. Эти микроорганизмы играют ключевую роль в быстром снижении активной кислотности молока, формировании вкуса, аромата и текстуры готового продукта, а также повышают безопасность за счет подавления нежелательной микрофлоры в процессе ферментации.

Разработка состава питательной среды для молочнокислых бактерий является важным этапом разработки технологий производства заквасок. Питательная среда должна быть экономически

эффективной и богатой питательными веществами, позволяя бактериям достигать высокой плотности клеток. Для промышленного использования необходимо использовать технологические режимы, позволяющие сохранять жизнеспособность и метаболическую активность бактерий в течение длительного времени. Лиофилизация является одним из наиболее эффективных методов сохранения жизнеспособности микроорганизмов. Однако этот процесс может вызывать повреждение клеток, включающее разрушение мембран и денатурацию белков, что может снизить выживаемость бактерий после сушки.

В связи с этим, состав питательной среды и использование эффективных криопротекторов являются критически важными факторами, влияющими на свойства бактериальных культур. Разработка простых, применимых в пищевой промышленности, технологически доступных питательных сред и криозащитных составов является важной задачей, особенно в условиях растущего спроса на закваски местного производства для молочной промышленности.

#### **Ключевые слова**

Молочнокислые бактерии, ферментированные молочные продукты, закваски, питательные среды, лиофилизация.

Growing the pure culture of cells is the first step for the preparation of starter cultures. The culture media and conditions can influence viability of the starters during preservation. Lactic starter cultures grow generally better in the rich media than in milk, but the media are too expensive for the industrial-scale preparation of bulk starters. Given the rapidly expanding market for bacterial starters in the food industry and the growing focus on maintaining a healthy intestinal microbiota, the development of new competitive products and the continuous renewal of lactic acid bacteria strains collections are essential.

The study was carried out using a stepwise experimental design integrating microbiological isolation, physiological screening, technological evaluation, and preservation trials.

**Isolation and Identification:** A total of eight strains of lactic acid bacteria were successfully isolated and identified from samples of traditional fermented dairy products. Cultivation was performed on MRS and M17 agar at 36–37 °C for 24–72 hours. All isolates were confirmed as Gram-positive and catalase-negative, matching the phenotypic profile required for technological LAB, a promising probiotic strain was also detected [1].

**Technological Screening:** The acid-forming capacity of isolates was assessed in sterilized skim milk. Fermentation was conducted at 37 °C for 18–24 hours. Strains demonstrating rapid acidification, homogeneous clot structure, and acceptable sensory characteristics were selected for further experimentation [2].

**Development of a Food-Grade Nutrient and Protective System:** To ensure applicability for fermented milk production, a simplified nutrient medium free from peptone was formulated. The medium contained milk-derived substrates and lactose as a carbohydrate source. Lactose was selected instead of glucose to better simulate dairy fermentation conditions and reduce risk of excessive by-product formation. For preservation studies, a cryoprotective solution consisting of skim milk powder and lactose was prepared and added directly to the culture medium without centrifugation. The absence of centrifugation minimized mechanical stress and simplified the process, making it scalable for industrial application. The medium consisting of dairy-based showed the most significant results. It not only ensured high bacterial survival rates during the freeze-drying process but also demonstrated excellent functional activity upon rehydration, yet the media is still under development to get the best possible results.

**Freeze-Drying Procedure:** Pre-freezing was conducted at low temperature to ensure uniform ice crystal formation. Lyophilization was performed under controlled vacuum conditions. The process included primary drying (sublimation phase) followed by secondary drying to reduce residual moisture. The protective effect of skim milk proteins is associated with membrane stabilization, while lactose functions as a glass-forming agent reducing molecular mobility during dehydration [3] [4] [5].

**Evaluation of Viability and Survival Rate:** Viable cell counts were determined before and after freeze-drying using the colony-forming unit (CFU) method according to standard

microbiological protocols [5]. The results demonstrated satisfactory preservation efficiency, confirming the protective capacity of the formulated system.

The research successfully resulted in the isolation of eight technologically promising LAB strains. A simplified, peptone-free protective medium was developed that facilitates high survival rates during freeze-drying. The final products showed excellent fermentation capacity and organoleptic properties, making this system a viable candidate for industrial starter culture production.

### **Literature**

1. Coeuret V., Dubernet S., Bernardeau M., Gueguen M., Vernoux J.-P. Isolation, characterization and identification of lactobacilli focusing mainly on cheeses and other dairy products // *Le Lait*. 2003. Vol. 83. P. 269–306. DOI: 10.1051/lait:2003019. 83. 10.1051/lait:2003019.
2. Bintsis T. Lactic acid bacteria as starter cultures: an update in their metabolism and genetics // *AIMS Microbiology*. 2018. Vol. 4, No. 4. P. 665–684. DOI: 10.3934/microbiol.2018.4.665.
3. Foerst P., Santivarangkna C. Advances in starter culture technology: focus on drying processes // *Advances in Fermented Foods and Beverages* / ed. by W. Holzapfel. – Cambridge: Woodhead Publishing. 2015. P. 249–270. DOI: 10.1016/B978-1-78242-015-6.00011-6.
4. Widyastuti Y., Febrisiantosa A., Tidona F. Health-promoting properties of lactobacilli in fermented dairy products // *Frontiers in Microbiology*. 2021. Vol. 12. Article 673890. DOI: 10.3389/fmicb.2021.673890.
5. Saikia R., Borpuzari R., Borpuzari T. Recent advances in dairy starter cultures: a review // *IndiabIn Journal of Animal Health*. 2022. DOI: 10.36062/ijah.2022.03922.