ISSN: 0975-766X
CODEN: IJPTFI
Available Online through *Research Article*
www.ijptonline.com

MASTER SEED MICRO-ORGANISMS SELECTED INTHE GORSKY STATE AGRARIAN UNIVERSITY AND THEIR PRACTICAL USE

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Received on 12-11-2016 *Accepted on 02-12-2016*

Abstract

Background / Objectives: The search for new promising microbial strains is a topical and preferred direction for the development of various food industry branches. Thereby this article is aimed at finding domestic competitive starter cultures.

Method: The main approaches to the study of this problem are to analyze the diversity of lactic microorganisms and select the best of them and that can be achieved by studying the complex of their cultural, physiological and biochemical and technological properties. Currently, when there is an intensive development of biotechnology in food production, including functional products, it is indisputably necessary to search for and develop scientific approaches to the production of probiotic food using physiologically active strains of lactic microorganisms that can inhibit the evolution in the body of pathogenic microorganisms and opportunistic pathogens as well as to improve the accessibility of ration ingredients and normalize micro flora of intestinal tract.

Results: The article presents the results for selection of master seed microbial strains selected in various parts of the Republic of North Ossetia – Alania, including in the high mountain regions. It was found that lactic bacteria are widespread in the Republic of North Ossetia - Alania. It is conclusively established that the strains selected in the regions in which they are used are more likely to adapt to domestic raw materials and have a positive effect on the development of the microorganism; this became the basis for seeking master seed microbial strains in North Ossetia-Alania. The scientific value of the studies undertook in this area is that high active strains of lactobacillus were selected, deposited in the Russian Collection of Industrial Microorganisms of the State Research Institute of

Genetics (GosNIIGenetika), and are used in practice in formulation of starter cultures in those regions of North Ossetia-Alania, where the microorganisms are not subjected to mutative changes as a result of human impact. A number of issues related to the identification and selection of starter cultures with high and standing potential, studying their biology and processing properties, the creation of functional products, which have an important role in human life, were considered. Motivation for the use of biologically-active probiotics in the diet of farm animals and poultry was scientifically established and proved.

Application/Improvements: The information contained in this article serves a useful purpose for the dairy industry, poultry and livestock enterprises aimed at the expansion of the range of functional foods and the use of probiotic additives.

Key words: Selection, strains, microorganisms, probiotics, antagonistic activity, functional food, diet.

1. Introduction

The most important promising and high-demand area of microbiology is the search for new strains of lactic microorganisms to create probiotics and functional foods [11].

In recent decades, a new direction has been developing - is functional food. To this end, probiotic lactic acid bacteria are usedto increase the bioavailability and therapeutic action of functional foods [13].

The dairy industry uses a huge variety of strains, and therefore studies for the detection of domestic high yielding lactobacillus strains are carried out through their comprehensive analysis[5].

Cultured dairy products manufactured with the use of lactic acid bacteria starter cultures form a high-growth segment of dairy industry - production of probiotic products [6].

In the agricultural sector it is reasonable to use lactic acid bacteria capable to survive in the gastrointestinal tract and to enhancethe productivity of farm animals and poultry [7].

A. Bezkorovainy[14]statedthatonly 20-40% of microbial strains survive in the stomach. In the opinion of K. Kailasapathy[15]many lactic acid bacteria strains do not reach intestinal tract, or survive in it only for several days. Consequently, it is necessary to search for bacteria strains, which can withstand the high stomach acidity.

Direct anti-toxic and anti-microbic action of probiotics can be successfully used when treating mild and moderate forms of enteric infections, virus diarrhea, antibiotic-enteropathogeniccoli bacillos is and antibiotic diarrhea [16-17].

The most important properties of probiotics are their ability to adhere to the intestinal epithelium through glycoconjugate receptors, thereby enabling colonization resistance and preventing the adhesion and invasion of

pathogens, which ultimately contributes to the improvement of the epithelium resistance, strengthening its barrier functions[15].

Given this, the search for new probiotics that have a high biological value, as well as the development of effective schemes for their practical use are important today.

Indicators for microbial strains of the same species may greatly differ from each other. Therefore, for the production of probiotic foods it is necessary to select strains with high technological properties [12].

A fundamental condition for the creation of a new probiotic product is its high level of usefulness and safety [1, 2, 3, 4, 8, 9, 10].

2. Methods

The aim of the research was the selection in the various regions of North Ossetia of pure cultures of industrial microorganisms, including lactic bacteria, their identification, establishment of biological and technological properties, as well as production testing of the most active strains.

The basic material was the samples taken from cultivated and wild plants, pickled and fresh vegetables, from kefir grains, as well as from the intestinal contents of animals and infants.

Speciesidentity for the selected bacteria strains was determined according to L.A. Bannikova(1975).

The initial culture medium for the primary selection of bacteria strains was 0.5% cow's milk. To obtain pure cultures of lactic acid bacteria MRS - agar was used.

3. Results and their discussion

3.1 Identification and microorganisms properties

In order to obtain objective information, any new strain of bacteria is subjected to a study of its properties needed for the identification and preparation of a passport.

Primary identification of the test microbial strains was performed in the laboratories of the Biotechnology Research Institute of Gorsky State Agrarian University by the investigation of cultural, tinctorial and physiological and biochemical properties.

The conclusive identification of strains was held in the Russian Collection of Industrial Microorganisms of the Federal State Unitary Enterprise GosNIIGenetika bycarrying out of the analysis of 16S RNA.

63 promising strains of 25 species of microorganisms were identified and deposited in the Russian Collection of Industrial Microorganisms of the State Research Institute of Genetics Russian National Collection of Industrial

Microorganisms of the Federal State Unitary Enterprise of State Research Institute of Genetics of Microorganisms (FSUE GosNIIGenetika) (Table 1). All strains are protected by patents of the Russian Federation (RF).

Table 1 contains names of microorganisms' species, strains names and sources of selection. 63 strains of lactic acid bacteria and yeast of 25 species were deposited in the Russian Collection of Industrial Microorganisms.

Table 1. Microorganisms collection selected by the Biotechnology Research Institute of FSBEI of Higher Education.

#	Microorganism species	Strain name	Source of strain selection
1.	*Metschnikowiapulcherrima*:	M.V.	Raspberry
		A.	From the surface of quince
		D.G.	From the surface of wild pear
		V.R.	Grape berries, Vostorg variety
		M.R.	Raspberry
		3-k (14)	Grape berries, Interes variety
2.	*Cryptococcus flavescens*	V.S.	Grape berries, Moldova variety
3.	*Hanseniaspporauvarum*:	E	Blackberry
		7-i	Grape berries, Burgund variety
4.	*Torulasporadelbrueskii*	B-1	Soil of absorption field of molasses distillery stillage
5.	*Rhodotorulamucilaginosa*	5-e	Grape berries, Dekabrskiy variety
6.	*Lactococcuscasei*	S5	Feces of young pigs
7.	*Enterococcus durans*:	R10	Surface of cucumber
		I-26	Kefir grains
		S-45	Feces of young pigs
		K-45	Feces of calves
		K37 VIII (1)	Intestinal content of Caucasian tur
		kos 37 bN19	Intestinal content of European roe deer
8.	*Enterococcus hirae*:	M 25bN23	Intestinal content of brown bear
		MK 25 37(1)10	Intestinal content of European roe deer
		O-45	Feces of lambs
		1-yak	Yacon leaves
		BK-37	Medic inflorescence
		I-27	Kefir grains
		I-29	Kefir grains
		turR37 XV(3)	Intestinal content of Caucasian tur
		M-37a N13	Intestinal content of brown bear

		F-45	White clover from high mountain region over Coban village in RNO-Alania
		F-f (45)	White clover from high mountain region over Coban village in RNO-Alania
		F-f (37)	White clover from high mountain region over Coban village in RNO-Alania
9.	*Enterococcus faecium*:	2-bat	Batata leaves
		30 gs	Sakhalin buckwheat leaves
10.	*Sacharomyces cerevisiae*:	DM-1	Fermented milk whey
		DM-2	Fermented milk whey
		A-1	Homemade ayran
		Den-4	Wild hop cone
		Bag-1	Wild hop cone
11.	*Sacharomycesunisporis*	I-2	Kefir grains
12.	*Streptococcus thermophiles*:	K-45	Sand pink leaves
		Vo4-1	Aconite leaves
13.	*Lactobacillus paracasei*	Z-37	Sauerkraut
14.	*Lactobacillus gallinarum*:	I-2.3	Kefir grains
		G-37	Sand pink inflorescence
		I-37	High mallow leaves
		I-12	Kefir grains
15.	*Lactobacillus fermentum*	V1-1	Large-flowered betony inflorescence
16.	*Lactobacillus plantarum*	V2-II	Large-flowered betony inflorescence
17.	*Streptococcus salivarius*:	M-9	Unpasteurized fermented milk
		M-11	Unpasteurized fermented milk
18.	*Lactobacillus helveticus*:	M-14	Unpasteurized fermented milk
		M-16	Unpasteurized fermented milk
19.	*Enterococcus thailandicus*:	K 45 III	Intestinal content of Caucasian tur
		ır K-25 XIV(I)	Intestinal content of Caucasian tur
		M 25a N15	Intestinal content of brown bear
20.	*Enterococcus mundtii*	turS37 III	Intestinal content of Caucasian tur
		K-f (37)	White clover from high mountain region over Coban village in RNO-Alania
		F-b (37)	Red clover, growing in high mountain region over Coban village in RNO-Alania
		K-b (37)	Red clover, growing in high mountain region over Coban

			village in RNO-Alania
21.	*Enterococcus faecalis*	M-37a N5	Intestinal content of brown bear
22.	*Kluyveromyceslactis*	A-2	Homemade ayran
23.	*Rhodotorulaglutinis*	KR-1	Hop cone
24.	*Aureobasidium pullulans*	PD-3	Hop cone
25.	*Staphylococcushominus*	K-f (45-l)	White clover from high mountain region over Coban village in RNO-Alania

During the study of lactic acid bacteria strains it was found that different strains of the same microorganism species differed significantly from each other. The speed of milk ripening is from 5 to 9.5 hours, and the maximum acid forming activity is from 95 to 357°T. CFU / ml of fermented milk is 10^9 -o 10^{10}. It is also found that different lactic acid bacteria strains of the same species differed significantly from each other in antagonistic activity. Multiple studies conducted have confirmed the fact that the lactic acid bacteria are generally widespread in the environment, and the data obtained restated the individual properties of microbial strains.

As a result of the researches conducted it was found that local strains of lactic acid bacteria could be widely used in the preparation of starter cultures for the production of various dairy products. All bacteria strains identified by the members of the Biotechnology Research Institute of Gorsky State Agrarian University are covered by Russian Federation patent for invention.

3.2. Practical use of microorganisms selected by Biotechnology Research Institute of FSBEI of Higher Education "Gorsky SAU"

Members of the Biotechnology Research Institute of Gorsky State Agrarian University, using their own selections of strains of different microorganisms, developed production technologies for: sour cream"Lakomka" (RF patent), soured milk made of buttermilk (RF patent), pickled cheese with addition of aqueous extract of spiced aromatic plants (12 RF patents), symbiotic sour cream product (RF patent), symbiotic functional cultured milk products (RF patent), yoghurts"Dieticheskiy"and"Diabeticheskiy", functional cultured milk productsenriched with dietary fibers, iodine, spirulina plantex, selenium, lactulose, carbonated fermented milk drink made of buttermilk and cheese whey, mead «Aspirantskaya», fermented milk pastas, enriched with pectin, inulin, fig, cultured milk products of soymilk. All the developed products have probiotic properties.

3.3. Results obtained by using local strains of lactic acid microorganisms in animal and poultry production

In order to establish the feasibility of using the studied strains during breeding of farm animals and poultry,experimental studies on the inspection of lactic acid microorganisms strains have been conducted in 90 pigs

and 2,200 broiler chickensby the introduction of living cultures of lactic acid bacteria inthe feed rations. As a result ofthe studies conducted it has beenfound that the introduction of probiotic lactic acid bacteria in the feed rations can increase the average daily live weight gains of pigs by 17.9%, and for broiler chickens thisindex was 18.5%. Population preservation was 5-10% higher in comparison with analogues of the control groups. The use of probiotics for growing broiler chickens improves digestibility of diet nutritional substances, improves blood hematological parameters within the physiological norm, and increases clean content that is beneficial to meat chemical composition.

4. Conclusion

The collection of the microorganisms with high biological activity selected by the Biotechnology Research Institute of Gorsky State Agrarian University offers great opportunities for the production of new functional fermented milk products and the creation of starter cultures for the production of probiotic fermented milk products and preparations.

References

1. Alkhamova, G.K., 2014. Produktyfunktsionalnogonaznacheniya [Functional Products]. Young Scientist, 12: 62-65.
2. Belokamenskaya, A.M., 2013. Issledovaniepishchevyhproduktoviprodovolstvennogosyriyanasoderzhanie rtutiatomno-absorbtsionnymmetodom [Research of Food Products and Food Ingredients for Mercury Concentration by Atomic Absorption Method]. Young Scientist, 10: 98–101.
3. Belokamenskaya, A.M., 2013. Primeneniephisiko-khimicheskihmetodovissledovaniy v laboratoriyah Chelyabinskoyoblasti [Application of Physicochemical Research Methods in the Laboratories of Chelyabinsk Oblast]. Young Scientist, 4: 48–53.
4. Bogan, V.I., 2013. Sovershenstvovaniemeto dovkontrolyakachestva prodovolstvennogosyriy aipishchevoyproduktsii [Improvements of Methods for Control of Food Ingredients and Food Products]. Young Scientist, 10: 101–105.
5. Botina, S.G., 2008. Vidovayaidentifikatsiyaipaspottizatsiyamolochnokislyhbacteriymetodamimolekulyarno-geneticheskogotipirovaniya [Species Identification and Certification of Lactic Acid Bacteria by the Method of Molecular Genetic Typing]. Dairy Industry, 3: 52.
6. Panin, A.N., N.I. Malik and E.V., 1993. Malik Probioticheskiepreparaty v veterinarii [Probiotics in Veterinary Science]. Vetinform, 2: 9-10.

7. Karycheva, O.V., 2007. Novyekultury dlyakislomolochnyhproductov v assortimentekompanii "KhristianKhansen" [New Cultures for Cultured Milk Products in the Assortment of Christian Hansen Company]. Dairy Industry, 11: 28.
8. Popova, M.A., M.B. Rebezov, A.O. Gayazova and S. V. Lukinyh, 2014. Otsenkaka chestvaibezo pasnostirazra botannogoyogurta [Quality and Safety Assessment of Developed Yoghurt]. Young Scientist, 10 (69): 199–202.
9. Rebezov, M.B., G.K. Alkhamova and N.L. Naumova, 2011. Srokikhraneniyanat sionalnogotvorozhnogoprodukta [Storage Life for Domestic Curd Product]. Dairy Industry, 11: 66–67.
10. Rebezov, M.B., 2012. Kontrol Kachestvarezultatovissledovaniyprodovol stvennogosyriyaipish chevyhproductovnasoderzhaniesvintsa. [Research Quality Control of Food Ingredients and Food Products for Lead Content]. News of Higher Educational Institutions. Applied Chemistry and Biotechnology, 2(1): 157–162.
11. Solovieva, I.V., 2010. Izucheniebiologicheskihsvoystvnovyhshtammovroda Lactobacillus [Investigation of Biological Properties of New Strains of Lactobacillus Genus]. General Biology. Bulletin of Lobachevsky State University of Nizhni Novgorod, 2(2): 462–468.
12. Frolova, M.D., 2008. Osobennostirazrabotkimezo filnyhzakvasok [Features for Development of Mesophilic Cultures]. Dairy Industry, 6: 70.
13. Fumiaki, A., 2010. Kriteriivyboraprobiotika [Probiotic Selection Criteria]. Dairy Industry, 5: 21.
14. Tsugkiev, B.G., R.G. Kabisov, E.V. Ramonova and A.G. Petrukovich, 2015. Vidovoeraznoobra ziemikroor ganizmov, sbrazhivayushchihlaktozu, v Respublike Severnaya Osetiya-Alaniyaiihprakticheskoeispolzovanie. [Diversity of Lactose-Fermenting Microorganisms in the Republic of North Ossetia – Alania and their Application]. Vladikavkaz: Publishing House of FSBEI of Higher Education "Gorsky State Agrarian University", pp: 240.
15. Bezkorovainy, A., 2001. Probiotics: Determinants of Survival and Growth in the Gut. American Journal of Clinical Nutrition, 73(2): 399S-405S.
16. Kailasapathy, K., 2000. Survival and Therapeutic Potential of Probiotic Organisms with Reference to Lactobacillus Acidophilus and Bifidobacterium spp. Immunological Cell Biology, 78(1): 80-88.
17. Pochapin, M., 2000. The Effect of Probiotics on Clostridium Difficile Diarrhea. American Journal of Gastroenterology, 95(1): 11-13.