International Journal of Medicine and Medical Research
2017, Volume 3, Issue 1, p. 50–52

DOI 10.11603/IJMMR.2413-6077.2017.1.6955

INFLUENCE OF *LACTOBACILLUS* SPP. ON COLONIZATION AND ANTI-INFECTIOUS RESISTANCE OF THE MUCOUS MEMBRANES OF THE UPPER RESPIRATORY TRACT

S. V. Kalinichenko[1], O. O. Korotkykh[1], S. I. Pokhil[1], M. G. Bakumenko[2]
[1]NATIONAL ACADEMY OF MEDICAL SCIENCES OF UKRAINE,
[1]MECHNIKOV INSTITUTE OF MICROBIOLOGY AND IMMUNOLOGY, KHARKIV, UKRAINE
[2]KHARKIV CITY STUDENT HOSPITAL, KHARKIV, UKRAINE

Background. *Lactobacilli are very important for the formation of colonization resistance and have pronounced antagonistic effect against a wide range of microorganisms. That is why the lactobacilli have extensive use as a component of classic probiotic agents that are widely used to prevent and treat dysbiotic conditions of digestive and genital systems of people.*

Objective. *The aim of the research was to study the effect of lactobacilli on anti-infectious resistance of mucous membranes of upper respiratory tract.*

Methods. *The colonization degree (lg CFU / g) of nasal mucosal membranes by Lactobacillus spp. and S. aureus was determined in all carriers before the experiment. Also, the level of lysozyme and secretory immunoglobulin A (sIgA) in nasal secretions cavities was identified.*

Results. *It was established a clear dysfunction of anti-infectious resistance in carriers of Staphylococcus aureus - a decrease of colonization resistance and local immunity of mucous membranes of upper respiratory tract. As for the anti-infectious resistance of nasal mucosal of S. aureus carriers, the level of lysozyme and secretory immunoglobulin A gradually increased after the application of probiotic strain L. rhamnosus GG, and in 21 days it reached rates of healthy individuals.*

Conclusions. *It was found out that probiotics for nasal passages sanitation in Staphylococcus aureus carriers lead to gradual eradication of the pathogen (S. aureus) with restoration of colonization and anti-infectious resistance, mucous membranes and upper respiratory tract.*

KEY WORDS: ***Lactobacillus* spp.; lysozyme; sIgA; upper respiratory tract.**

Introduction

Lactobacilli are widely distributed in the environment and have a high biological activity. They are an important part of normal microflora of digestive and genital human tracts and they belong to the resident microflora of nasopharynx [1]. Lactobacilli are actively involved in the formation of colonization resistance of mucous membranes and have significant antagonistic action against a wide range of bacteria [2-4]. That is why the lactobacilli are widely used as a component of classic probiotic agents [3-5]. Probiotics based on lactobacilli are widely used to prevent and treat dysbiotic conditions of digestive and genital human system [6-8]. Nowadays, research on the use of lactobacilli for prevention or treatment of infections and recovery of anti-infectious resistance of mucous membranes of upper respiratory tract (URT) are actual [9].

The aim of the research was to study the effect of *Lactobacillus* spp. on colonization and anti-infectious resistance of mucosal membranes of upper respiratory tract in *Staphylococcus aureus* carriers. The objects of the study were the carriers of Staphylococcus aureus (n=29) among the medical staff of a hospital, Kharkiv (Ukraine).

Methods

The colonization degree (lg CFU / g) of nasal mucosal membranes by *Lactobacillus spp*. and *S. aureus* was determined in all the carriers before the experiment. Also, the level of lysozyme and secretory immunoglobulin A

Corresponding author: Svitlana Kalinichenko, Laboratory of Viral Infections, National Academy of Medical Sciences of Ukraine Mechnikov Institute of Microbiology and Immunology, 14 Pushkinska Street, Kharkiv, Ukraine, 61057.
Phone number: +380577313151
E-mail: imidir@ukr.net

(sIgA) in nasal secretions cavities was identified. Then the carriers were divided into groups: I (n=7, control) – the persons, who received saline as the sanitation agent (0.85% NaCl, pH 7.2) during 30 days, 2–3 drops twice per day in each nostril; II (n=11) – the persons, who received as the sanitation agent a suspension of probiotic strain *L. rhamnosus* GG (in saline) at a concentration of 5×10^9 CFU/ml (according to the group I scheme). The degree of colonization of nasal mucous membranes by *Lactobacillus spp.* and *S. aure*us, lysozyme and sIgA levels in nasal cavities secretions were determined in the individuals taking part in the experiments repeatedly in 7, 14 and 21 days of sanitation. The material was selected as follows: in each nostril, alternatively, with special replicator sterile foam tape was injected and mucus impregnation was held for 5 minutes, after it the tape was immersed in a sterile tube containing 1.0 ml of phosphate-buffered saline (PBS, pH 7.2) for 30 minutes and then separated from the foam carrier mucus by insulin syringe, forcing it into the PBS.

In the comparisons group there were persons, who did not have upper respiratory tract infection within 6 months (almost healthy, n=15).

Lysozyme levels were determined by means of nephelometric method [10]. The concentration of lysozyme was rated in micrograms/ml (μg/ml). To determine sIgA, RIDASCREEN®sIgA ELISA kits of R-Biopharm AG Company (Germany) were used. The results of calculations were carried out on Stat Fax enzyme immunoassay analyser. sIgA concentration was rated in mg/ml.

Results

It was established that in the healthy individuals (n=15) the degree of colonization of the nasal mucous membranes *Lactobacillus* spp. was 6.64±0.9 lg CFU/g, the amount of lysozyme and sIgA in nasal secretions cavities were respectively at 17.45±2.9 μg/ml and 129.3±21.7 mg/ml.

The degree of colonization of nasal mucosa Lactobacillus spp. was 2.2±0.7 lg CFU/g, the amount of lysozyme and sIgA in nasal secretions cavities were respectively 7.45±2.1 μg/ml and 63.9±19.8 mg/ml in the *S. aureus* carriers.

Discussion

Consequently, the degree of colonization of nasal mucosa Lactobacillus spp. in the carriers of Staphylococcus aureus was in 3.8–5.0 ($p<0.01$) times lower compared to the healthy individuals. The amount of lysozyme and sIgA in nasal secretions cavities were also lower in comparison with the healthy individuals, respectively in 1.5–3.8 ($p<0.05$), and 1.2–3.4 ($p<0.05$) times.

Thus, it was established a clear dysfunction of anti-infectious resistance in the carriers of *Staphylococcus aureus:* the decrease of colonization resistance and local immunity of mucous membranes of upper respiratory tract.

According to the literature, application of probiotics or synbiotic treatment regimens in respiratory infections reduces relapse respiratory tract infections and bronchial obstruction in children [9]. But the authors did not conduct any studies to determine the number of lactobacilli in secrets of URT and their influence on anti-infectious resistance of mucous membranes. In connection with the foregoing, we conducted a study on the possibility of recovery colonization and anti-infectious resistance of mucous membranes of upper respiratory tract in the carriers of *S. aureus* using the probiotic strain *L. rhamnosus* GG.

The data in the Table 1 prove that the probiotic strain *L. rhamnosus* GG for sanitation the carriers decreased the degree of colonization by Staphylococcus aureus mucosal in 7 and 14 days after the sanitation of lactobacilli suspension, and in 21 days there was a complete eradication of *S. aureus* from nasal mucous membranes. Moreover, the decrease of *S. aureus* degree colonization on mucosal of the carriers occurred against the background of increase in the number of *Lactobacillus spp*. on mucous membranes.

As for the anti-infectious resistance of nasal mucosal of *S. aureus* carriers, the level of lysozyme and secretory immunoglobulin A after the application of the probiotic strain *L. rham-*

Table 1. Average indexes of mucosal colonization resistance after sanitation, (M±m)

Groups	The degree of mucosal colonization URT, lg CFU / g					
	Lactobacillus spp.			*S. aureus*		
	in 7 days	in 14 days	in 21 days	in 7 days	in 14 days	in 21 days
I	2.3±0.7	2.2±0.6	2.1±0.7	4.7±0.7	4.9±0.6	4.8±0.6
II	3.8±0.4*	4.5±0.4*	5.3±0.4*	1.8±0.6*	0.6±0.6*	0±0*

Note: * – significant difference between these indexes of I and II groups ($p<0.05$).

Table 2. Average indexes of mucosal anti-infectious resistance after sanitation, (M±m)

Group	Indicator of local immunity					
	level of lysozyme, μg/ml			level of sIgA, mg/ml		
	in 7 days	in 14 days	in 21 days	in 7 days	in 14 days	in 21 days
I	7.3±2.4	7.4±2.2	7.4±2.3	61.3±21.3	62.1±18.8	61.9±19.3
II	11.6±3.3*	13.3±2.9*	15.4±2.7*	84.8±19.8*	102.9±21.1*	112.2±19.1*

Note: * – significant difference between these indexes of I and II groups ($p<0.05$).

nosus GG significantly increased, and in 21 days it reached rates of healthy individuals (Table 2).

To be precise, after the sanitation of probiotic nasal of the carriers Staphylococcus aureus, the level of lysozyme and sIgA rose: 7 days, an average of 1.6 ($p<0.05$) and 1.4 ($p<0.05$) times, respectively; at 14 days of 1.8 ($p<0.05$) and 1.6 ($p<0.05$) times, respectively; after 21 days – 2.1 ($p<0.05$) and 1.8 ($p<0.05$) times, respectively, compared with the group the carriers, that sanitation was conducted with saline.

Conclusions

Thus, it was found that the application of probiotics for nasal passages sanitation in the carriers of *S. aureus* leads to the gradual eradication of the pathogen (*S. aureus*) with colonization restoration (increased degree of mucosal lactobacilli settlement), and anti-infectious resistance (increased level of sIgA and lysozyme) in mucous membranes and upper respiratory tract.

This study demonstrated the positive effect of *Lactobacillus spp*. on the formation of colonization and anti-infectious resistance of upper respiratory tract mucous membranes and provided an opportunity to consider probiotics as a useful option for immune exposure to chronic infections of staphylococcal origin.

References

1. Ryzhkova TA. The peculiarities of antagonistic activity of *Lactobacillus* strains (isolated from different econiches and probiotics) under cultivation conditions different by gas composition. Annals of Mechnikov Institute. 2014;2:64–69.
2. Bondarenko VM, Rybalchenko OV. Analysis of preventive and therapeutic action of probiotic drugs from the standpoint of new scientific technologies. Microbiology journal. 2015;2:90–104.
3. Sishel LM. Immunomodulatory activity of probiotic strains lakto- and bifidobacteria in vitro and in vivo. Abstracts XIII Congress of the Society of Microbiologists of Ukraine. 2013;278.
4. Sishel LM. Imunobiotics – a group of new biotechnology drugs with antimicrobial and immunomodulatory properties Abstracts XIII Congress of the Society of Microbiologists of Ukraine. 2013;279.
5. Kalinichenko SV. Microecological disorders and their modern correction. Ukrainian biopharmaceutical journal. 2016;2(43):6–12.
6. Babenko LP. Effect of probiotic strains of lactobacilli and bifidobacteria in range of microbiota and immune reactivity urogenital tract with staphylococca infection. IMV NASU. 2015:28.
7. Bobyr VV. Intestinal virus and human normal microflora: features of interaction. Annals of Mechnikov Institute. 2015;2:25–29.
8. Osolodchenko TP. Role of distal intestinal tract microflora in support of oxalate homeostasis. Annals of Mechnikov Institute. 2015;2:42–46.
9. Stojkovic A, Simovic A, Bogdanovic Z, Bankovic D, Poskurica M. Clinical trial or experimental study (consort compliant): Optimal time period to achieve the effects on synbiotic-controlled wheezing and respiratory infections in young children. Srp Arh Celok Lek. 2016;144(1–2):38–45.
10. Dorofeychuk VG. Determination of lysozyme activity by nephelometric method. Laboratory work. 1968;1:28–30.

Received: 2016-11-18