Application of probiotics for acute respiratory tract infections

Oxana Zolnikova,1 Inna Komkova,2 Nino Potskherashvili,2 Alexander Trukhmanov,1 Vladimir Ivashkin1

1Internal Diseases Propedeutics, I.M. Sechenov First Moscow State Medical University, Moscow; 2Vasilenko Clinic of Internal Disease Propedeutics, Gastroenterology, and Hepatology, I.M. Sechenov First Moscow State Medical University, Moscow, Russian Federation

ABSTRACT

We have reviewed the currently published results on a role of the gut microflora in a prevention of acute respiratory infections. The main biological properties of probiotic bacteria are presented in a context of their modulating activity on an inflammatory immune response. Available data on the reduction of a possible risk, duration, and severity of respiratory infection symptoms during a probiotic medication intake were analyzed. Potential antiviral probiotic mechanisms have been reviewed and discussed.

Introduction

It is recognized, acute respiratory infections (ARIs) are dominating in a global structure of a disease distribution and statistics show the ARIs have surpassing other contagions. Among the currently known respiratory infectants, most of them are viruses (over 200 pathogens). Viral ARI pathogens belong to various virus families, so RNA-containing viruses are more significant: Picornaviridae - rhinoviruses, enteroviruses; Orthomyxoviridae - influenza viruses; Paramyxoviridae - parainfluenza viruses, respiratory syncytial viruses, metapneumoviruses; Reoviridae - rotaviruses; Coronaviridae - coronaviruses, DNA-containing viruses; Adenoviridae - adenoviruses; Rhabdoviridae - bocaviruses. There are both well-known and new virus strains among those. The strains were identified thanks to a development of molecular biology research methods in recent years. Viruses have had a very important ability to change their own antigenic structure. So it leads to a forming of highly toxic strains, which are become resistance to the etiotropic medications.

Acute control of pulmonary inflammation

The respiratory tract epithelium is an entry gate for viruses. The human organism had been improving its own protective mechanisms for centuries. The non-specific factors are the first level of protection: tenacious mucus, a continuous motion of the columnar epithelium’s cilia, non-specific virus replication inhibitors, secretory immunoglobulin A (IgA) have been contained in the respiratory tract secretion. Other protective factors, such as C-type lectins (conglutinin, mannan-binding protein, surfactant proteins A and D) are forming the anti-infective barrier, the lectins have been bound to virus carbohydrates to cause its aggregation and to provide their better imbibition by phagocytes. To have an infection, a virus must overcome these non-specific respiratory resistance factors. The main target consists in the ciliated columnar epithelial cells for that. Upon infection, the viral RNA/DNA will be identified by Toll-like receptors (TLR-3) and a gene (RIG-I), which launch a release of early inflammatory mediators: interferons (IFN) type I, so the proapoptotic factors have been enhanced in epithelial cells; tumor necrosis factor-α (TNF-α) and chemokines (CXCL8 and CXCL11) are activating natural killers (NK) and polymorphonucleocytes. As a result, one...
Probiotics for acute respiratory tract infections causes a destruction of viral nucleic acid and limits a virus replication.\textsuperscript{2,3} The influence of viral infection on the dendritic cells (DC) leads to a cascade of reactions with an activation of CD4+ and CD8+ cells and subsequent development of specific T- and B-cell-mediated immunity (Figure 1).

Selective role of commensal microbiota in immune responses to influenza virus

The human microbiome is the key resource to form and to support natural immunity and adaptive capabilities of the organism. The impact of normal flora on immunoregulation outside of digestive system, particularly in lungs, has been established recently and it is a subject for active research now. Signals have been transmitted by synanthropic bacteria and their metabolites are interacting with TLRs, inducing effector’s functions have been bound to an expression of the nuclear transcription factor (NFkB), DCs, T regulatory lymphocytes, chemokines, and cytokines. Bacterial metabolites, particularly short-chain fatty acids (SCFAs) influence the epithelial and immune cells directly, so the immune response is enhancing significantly. It was demonstrated that SCFAs influenced pattern recognition receptors (PRR) by an activating of NFkB, TNF-α and it reduced a stimulation of PRR\textsuperscript{3-6} (Figure 2). Models of experiments on animals confirm the intestinal microbiota has been involved in a supporting of antiviral respiratory immunity. Specifically, as it was demonstrated by Ichiohe et al., long (for 3 weeks) antibiotic intake reduced a resistance to intranasal influenza A virus infection dramatically. This was accompanied by an increase in viral titers and by a decrease in IgA and IgG levels, and CD8+ and CD4+ T cell deactivation versus the animals without antibiotics. Administering of TLR ligands (commensal bacterial peptidoglycans) restored an antibiotic-suppressed antiviral immune response.\textsuperscript{4} This fact confirms the received signals in the lower gastrointestinal tract, once have been transmitted to the mucosae of other biotopes including the respira-

Figure 1. A model of lung immunology response to a virus infection. A) Lung epithelium. The viral RNA/DNA is recognized by TLR-3 and RIG-I receptors, one launches a release of interferons (IFNs), tumor necrosis factor (TNF)-α, and chemokines. IFNs type I enhances proapoptotic factors in epithelial cells, whilst TNF-α and chemokines are activating Nks and polymorphonucleocytes (PMNs). The viral RNA/DNA interacts with dendritic cells (DCs). The viral RNA/DNA interacts with dendritic cells (DCs). B) Regional lymph nodes. DCs facilitate activation of CD4+ and CD8+ T cells, which migrate back to the infected epithelium with subsequent release of mediators and inflammatory cells including immunoglobulin A (IgA).
tory tract to enhance a protection against infection. So, it provides a basis to research any capabilities of the probiotic application as for the ARI complex treatment, as for a prevention of the diseases. However, a question has arisen, have all probiotic bacteria got an immunotropic activity? Most of researches is conducted to that and significant intergeneric, interspecific and interstrain differences have been detected in the immunomodulatory activity of symbiotic bacteria now. It had been found, that existing differences were not only in the evidence but also in a matter of the immunotropic effects.7,8

Health effects of probiotics in acute respiratory infections

Animal experiments

A clinical potential for the probiotic medications was demonstrated by the experiments on animals. It was shown in many articles, an oral or intranasal administration of the strains of Lactobacillus pentosus,9,10 L. casei Shirota,11,12 L. plantarum,13,14 L. delbrueckii ssp. Bulgaricus OLL1073R1,15 L. rhamnosus GG,16,17 L. gasseri TMC0356,18 Lactococcus lactis ssp. cremoris FC,19 L. brevis KB20 or B. breve YIT406421 has helped for a suppression of the infection symptoms for the influenza-virus-infected mice. Simultaneously it induces a decrease of the viral load titers, an amplification of the mucosal immunity (an augmentation of IgA level in the saliva and serum), a more intensive generation of IFNs type I, tumor necrosis factor and an increase of T helpers in the lung parenchyma. Administration of L. plantarum NCIMB 8826 and L. reuteri F275 also reduced an inflammation severity and the mortality of the mice that were infected with the pneumovirus.22 Besides, L. rhamnosus CRL1505 and L. rhamnosus CRL1506 protected the mice from a respiratory syncytial virus infection.23 Simultaneous administration of L. rhamnosus and B. lactis contributed to an increase of IFN-γ, interleukin (IL)-4, IL-10, and IL-6 in the bronchoalveolar lavage, as well as to the quantity and activity of phagocytic cells and NKs.24,25

Figure 2. A model of regulatory influence of the gastrointestinal microflora on lung immunology. Intestinal bacteria interact with Toll-like receptors (TLRs) and activate nuclear transcription factor (NFκB) and dendritic cells (DCs) functions as from the bowel lumen directly as after translocation. DCs modulate the activity of T regulatory lymphocytes (Treg) and various regulatory cytokines (interleukin (IL)-10, tumor necrosis factor (TGF)-α, interferon (INF)-γ, IL-6). Being the bacterial metabolites, short-chain fatty acids (SCFAs) directly influence on epithelial and immune cells, thereby exerting a significant influence on immune response.
Clinical trials

The experimental findings generally fit into the clinical impression results. Series of clinical studies for preventive probiotic effects on a viral respiratory infection (across age groups) confirmed the antiviral probiotic efficacy in children (Table 1), in adults (Table 2).\textsuperscript{26-41} Mono-preparations with various content of lactobacillus species/strains and complicated medications with lacto- and bifidobacterium were studied. There are a lot of results related just to an evaluation of the quantity and severity of respiratory infection episodes in the presented studies, whilst only few studies informed about the viral load dynamics.\textsuperscript{26-41} The dosage and the schedule of a probiotic administration used in some trials are presented in the Table 3.

An analysis of the obtained results is allowed to suggest that effect of probiotic is undoubtedly highly strain-specific and predetermines their clinical efficacy. Interesting information was also obtained on the impact of lactobacilli on postvaccinal immunity. Davidson \textit{et al.} have demonstrated that LGG administration for 28 days after vaccination enhanced immune response with a titer augmentation in protective antibodies in comparison with the group to whom influenza vaccine only was administered.\textsuperscript{42} Similar findings were also presented by other researchers.\textsuperscript{43,44} The systematic Cochrane Review was published in 2015. The results of 12 randomized controlled studies were included, they involved 3720 people (across age groups) under probiotic medications for 3 winter months on the average. The probiotics have an advantage over placebos in accordance with the following criteria, as it was found: i) the number of participants after an ARI in the probiotic group, there were fewer [after at least 1 ARI episode: odd ratio (OR) 0.53, 95% confidence interval (CI) 0.37-0.76, P<0.001; subjects after ≥3 episodes: OR 0.53, 95% CI 0.36-0.80, P=0.002]; ii) it was observed a decrease in the average ARI episode duration [mean difference (MD) –1.89; 95% CI –2.03 to –1.75, P <0.001] and subjects were

<table>
<thead>
<tr>
<th>Author</th>
<th>Population</th>
<th>Probiotic</th>
<th>Episodes, symptom severity, and duration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hatakka\textsuperscript{26}</td>
<td>571 children</td>
<td>\textit{L. rhamnosus GG}</td>
<td>Incident count (below 4 times a year)</td>
</tr>
<tr>
<td>Kumpu\textsuperscript{28}</td>
<td>523 children</td>
<td>Randomized, double blind, placebo-controlled study</td>
<td>No duration and antibiotic demand changes</td>
</tr>
<tr>
<td>Lin\textsuperscript{36}</td>
<td>1062 children</td>
<td>\textit{L. casei rhamnosus}</td>
<td>Less antibiotic demand and bacterial complications</td>
</tr>
<tr>
<td>Nocerino\textsuperscript{37}</td>
<td>377 children</td>
<td>\textit{L. paracasei CBA L74}</td>
<td>Incident count (below 4 times a year)</td>
</tr>
<tr>
<td>Hojsak\textsuperscript{39}</td>
<td>210 children</td>
<td>\textit{B. animalis}ssp. Lactis</td>
<td>No duration and incidence changes</td>
</tr>
<tr>
<td>Caraiova\textsuperscript{40}</td>
<td>57 children</td>
<td>\textit{L. acidophilus CUL21} and CUL60; \textit{B. bifidum CUL20}; \textit{B. animalis} subspp.\textit{ lactis}</td>
<td>Duration (under 4 days)Incident count (below 3 times a year)Less school absence</td>
</tr>
<tr>
<td>Luoto\textsuperscript{29}</td>
<td>94 premature infants (32-36 weeks)</td>
<td>\textit{L. rhamnosus GG}</td>
<td>No duration, severity changes</td>
</tr>
<tr>
<td>Kukkonen\textsuperscript{30}</td>
<td>925 pregnant</td>
<td>Randomized, placebo-controlled trial</td>
<td>Incident count (below 4 times a year)</td>
</tr>
</tbody>
</table>

Table 1. Probiotic effect on acute respiratory infections incidence, severity, and symptom duration for children.

<table>
<thead>
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</tr>
</thead>
<tbody>
<tr>
<td>Turchet\textsuperscript{33}</td>
<td>360 adults</td>
<td>\textit{L. casei D114001}</td>
<td>Duration (under 4 days)</td>
</tr>
<tr>
<td>Guillemard\textsuperscript{34}</td>
<td>1072 adults</td>
<td>Randomized, double blind, placebo-controlled trial</td>
<td>Incidence count (below 3 times a year)</td>
</tr>
<tr>
<td>De Verse\textsuperscript{41}</td>
<td>479 adults</td>
<td>\textit{L. gasseri PA16/8, B. longum SP07/3, B. bifidum MF 20/5}</td>
<td>Duration (under 3 days)</td>
</tr>
</tbody>
</table>

Table 2. Probiotic effect on acute respiratory infections incidence, severity, and symptom duration for adults.
significantly less likely to resort to antibiotic treatment [risk ratio (RR) 0.65; 95% CI 0.45-0.94].

Probiotic prescription to children is a separate matter. As it was proved in the published systematic review and meta-analysis in 2016, the probiotic prescription to children (6269 people from infants to 18-year-olds) reduced morbidity. The number of patients with 1 ARI episode (RR 0.89, 95% CI 0.82-0.96, P=0.004) was much lower, with a total decrease of sick days (MD –0.16; 95% CI –0.29 to –0.02, P=0.03). Children with a probiotic administration were absentened in school or needed in a day-patient treatment for fewer days (MD –0.94, 95% CI –1.72 to –0.15, P=0.02).46 Thereby, a point of view of evidential medicine is that a probiotic administration significantly benefits ARI prevention both in the adult and infant population.

With regard to pneumonia, probably, the probiotics place and role in treatment of community-acquired pneumonia will be long discussed. Basically, the probiotic application is being considered from the point of view of a microbiotic correction after antibiotic treatment. Nosocomial infection prevention and treatment with probiotic administration are very interesting for scientists. A meta-analysis carried out on 2972 patients during that period, after an intake of probiotics [risk ratio (RR) 0.65; 95% CI 0.45-0.94].

Possible mechanism of actions of symbiotic bacteria in respiratory virus infections

To date, key molecular mechanisms have been disclosed in accordance with the symbiotic bacteria enhancing virus response. Perhaps, the antiviral probiotic effects could be realized by various ways. First of all, there is evolutionary antagonism between bacteria and viruses. It was established that the symbiotic bacterial flora continually produces nucleolytic enzymes (nucleases), which are circulating in the blood and lymph. These enzymes could be responsible for a proteolysis of virion capsids.42 Besides, peptidoglycans and muramyl peptides, as a part of the bacterial wall, significantly enhance the antiviral bodily protection. Interacting with intracellular NOD-receptors, they have additionally induced a signal cascade of reactions, which results into a synthesis for inflammatory cytokines by immune competent cells and activation of bodily immunoprotect mechanisms.48,49 Probiotics can block a virus attachment also by a competition process for certain receptors. Mucosa regeneration is augmented by mucin ability to prevent the virus attachment to epithelial cells and suppress the virus replication. The antiviral effect of probiotics is also related to their ability to produce antimicrobial peptides, dehydrogenases and NOs. Probiotics have also shown an ability to modulate the functions of epithelial cells, dendritic cells, CD4+ CD8+ T lymphocytes, NK cells, and stimulate synthesis of secretory immunoglobulins helping neutralize a virus.50,51

Conclusions

Recent researches on the human microbiome composition and functions have aroused a great interest for a target of a probiotic application and development to prevent ARIs. However, the question about probiotic microorganisms properties, which has to be underlined for a prevention or treatment choice against this particular pathology, remains open. Based on tax-

Table 3. The dosage and the schedule of a probiotic administration.

<table>
<thead>
<tr>
<th>Population</th>
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</tr>
</thead>
<tbody>
<tr>
<td>571 children forming Hatakka26</td>
<td>An average volume of milk (with <em>L. rhamnosus GG ATCC 53103</em>) was 260 mL (1% fat and 5·10^9 colony units/mL (c.f.u.) of strain LGG) for a daily consumption for over seven months</td>
</tr>
<tr>
<td>94 premature 2×10^9 infants (birth weight, &gt;1500 g) gestational age 32-36 weeks Luoto29</td>
<td><em>L. rhamnosus GG ATCC 53103</em> for the 1st and 60 days of lifeAt a dose of 1×10^8 c.f.u./day for 1 to 30 days and c.f.u./day for 31 to 60 daysThe products with approximately 10 mL of breast milk were mixed right before an administration to the infants</td>
</tr>
<tr>
<td>925 pregnant 2-year follow-up Kukkanen38</td>
<td>During the 2 to 4 weeks before delivery, mothers twice daily took 1 capsule containing <em>L. rhamnosus GG (ATCC 53103), 5×10^8 c.f.u.; L rhamnosus LC705(DSM 7061), 5×10^8 c.f.u.; B bifidobacterium breve BB09 (DSM 13692), 2×10^8 c.f.u.; Propionibacterium freudenreichii ssp. shermanii JS (DSM 7076), 2×10^8 c.f.u.</em> Their newborn infants received 1 opened capsule containing the same probiotics mixed with 20 drops of sugar syrup containing 0.8 g of galacto-oligosaccharides once daily for 6 months after birth</td>
</tr>
<tr>
<td>494 children Agustina32</td>
<td>*L. casei D114001; L. reuteri DSM 17938 180 mL of milk twice daily 6 month100 mL of milk with 5×10^8 c.f.u. per day of *L. casei CRL 431, or one with 5×10^8 c.f.u. per day of Lactobacillus reuteri DSM 17938</td>
</tr>
<tr>
<td>479 adults De Verse41</td>
<td>*L. gasseri P416/8; B. longum SP07/3; B. bifidum MF 20/5 5×10^8 c.f.u./tablet during 3 months, at least</td>
</tr>
</tbody>
</table>
onomic, most of the commonly used probiotic bacteria are the *Lactobacillus* and *Bifidobacterium* species. However, their probiotic effects have had a strain specificity, which is obviously affecting their biological activity.

A capability of the probiotic administration for ARI prevention and treatment could be more effective, as it was demonstrated by the presented meta-analysis results.

**References**


**Probiotics for acute respiratory tract infections**


