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Development and clinical assessment of a novel probiotic candy in the prevention of respiratory infections in asthmatic children

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ABSTRACT

Objective: Asthma is the most common chronic disease among children. Upper respiratory infections are often the cause of asthma exacerbation. Studies suggested that spore-forming probiotics can reduce viral infections. This study aimed to determine the effect of spore-forming probiotic *Bacillus* candy on respiratory illnesses in asthmatic children.

Methods & materials: In this randomized trial, 69 children aged 7–11 years with mild intermittent asthma were randomized to receive probiotic candy (containing 10^{10} CFU probiotic *Bacillus* coagulans) or placebo candy, daily for 2 months. The primary outcome was the number of viral respiratory infections. Secondary outcomes included salbutamol metered-dose inhaler (MDI) use, oral corticosteroids, school absenteeism, emergency department visits and hospitalizations, and Pediatric Asthma Control Questionnaire (c-ACT).

Results: The frequency of symptomatic respiratory illnesses was significantly lower in the probiotic candy group compared to placebo in the first month (0.28 ± 0.45 vs. 0.51 ± 0.50, $p = 0.054$), the second month (0.08 ± 0.28 vs. 0.41 ± 0.49, $p = 0.001$) and the total study (0.37 ± 0.54 vs. 0.90 ± 0.73, 0.001). The percent of patients with prednisolone consumption in the probiotic group was lower than the control group (2.9% vs. 14.7%) but did not show a significant difference ($p = 0.081$) and no difference was seen in the rate of emergency department visits and hospitalization between the 2 groups (both $p = 0.254$). The use of salbutamol and school absenteeism in the probiotic group was significantly lower than in the control group ($p = 0.040$ and $p = 0.046$, respectively. There was no significant difference in the evaluated scores for asthma control (c-ACT) in both probiotic and placebo groups. After the intervention, the difference between the 2 groups has become significant ($p < 0.05$).

Conclusion: Adding spore-forming probiotic candy containing *Bacillus* coagulans to standard asthma treatments reduced symptomatic respiratory illnesses over two months. Further studies including longer treatment periods are needed before making recommendations for routine use.

Keywords: Asthma, Spore-forming probiotic candy, Respiratory infections

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INTRODUCTION

Asthma is the most common chronic disease among children and has a significant impact on patients[1] and imposes a huge social and economic burden affecting millions of people worldwide. This condition is reported to affect 1 to 18% of children in different countries across the world, with a mean prevalence of 10.9% in Iran.[2] Viral respiratory infections are the most common cause of asthma attacks in children (80–85%) and, if left uncontrolled, are associated with reduced lung growth and accelerated loss of lung function.

Asthmatic children impose a significant burden on medical resources and morbidity, including hospitalization in Iran.[3] Studies have shown that the airway epithelial cells in asthmatic patients have defective innate immunity and produce less antiviral interferons IFN-β and IFN-λ when infected with human rhinovirus.[4,5] Current drugs to prevent virus-associated asthma exacerbations are ineffective, and new treatments are needed. We have already reviewed strategies to prevent viral infections. A balanced diet, probiotics, and the immune stimulant OM-85 are considered the most effective means of preventing colds in children with asthma.[3]

The increase in asthma prevalence, that started in the developed Western world in the twentieth century,[6] was initially attributed to the "hygiene hypothesis", in which a decrease in family size and reduction of "infections" in early life was posited to increase the risk of allergies, hay fever, and asthma.[7] The major differences between centuries found in the International Study of Asthma and Allergies in Childhood (ISAAC) Phase I are probable to be due to environmental factors.[8]

Emerging evidence suggests that imbalances in the composition, abundance, and diversity of the human microbiota associated with a "Western environment, including diet," promote the maintenance of type 2 T cell responses and allergies in infants.[7] Children with established asthma are likely to have a different lung and intestinal microbiome from healthy children. However, whether these changes, known as symbiosis, are the cause of, or result of, asthma and asthma treatments is not clear. Children at high genetic risk of asthma and allergy have immature responses to bacterial and viral infections in early life.[9] Treatment with immune-modulating agents that act to produce more "balanced" immune responses by inducing and educating regulatory T cells in the gut can prevent recurrent respiratory illnesses in these children.[10,11] The concept of an active gut-lung axis has opened the way for research into advancing maturation of the immune system.[12] Commensal gut bacteria may ferment no digestible oligosaccharides to produce postbiotics including short-chain fatty acids. Infants with higher levels of short-chain fatty acid in feces at 1 year may be at decreased risk for atopic sensitization and asthma between 3 and 6 years.[13]

Intestinal microbiota affects the IFN-α/β receptor surface expression in respiratory epithelial cells, If a respiratory viral infection occurs, these receptors can respond more efficiently to type I IFNs stimulation with enhanced interferon-stimulated gene levels and impede early virus replication.[14,15] Edible probiotics affect the main regulatory and anti-inflammatory cytokine.[1]

Studies have shown that the consumption of probiotics could reduce upper respiratory infection incidence, duration, and disease severity.[16–19] Zhao et al in a 2022 meta-analysis concluded that probiotics can reduce the number of participants diagnosed with at least one event of upper respiratory tract infection (risk ratio RR = 0.76), at least 3 events (RR = 0.59), and shorten the mean duration (MD) of episodes (MD = −1.22 days). Based on the results of 23 randomized controlled trials (RCTs) and 1 cluster RCT, the study concluded that probiotics are superior to no treatment or placebo in preventing acute upper respiratory tract infections.[20] Ahanchian et al showed that a multistrain symbiotic, Lactocare®, reduced episodes of viral respiratory infections in asthmatic children.[4] However, Sadrifar et al in their study confirmed that probiotics could have beneficial effects on the immune system-related.[21] Probiotic supplementation showed a promising beneficial role in the reduction of the incidents and symptoms of influenza-like illness and common cold[22] (Probiotics are summarized in Table 1). A systematic review by Lin neither confirmed nor excluded the beneficial effects of probiotics as

probiotic strain	Dosage	Outcomes
Lactobacillus bulgaricus, Streptococcus thermophilus, and Lactobacillus casei.[34]	100 mL/day for 12 months	Not change the duration of fever in asthmatic children
Lactobacillus rhamnosus[46]	4 capsules/day 5.5 months	Changes in allergic nose, eye, lung, and total symptom scores
Lactobacillus paracasei.for[17]	2 capsules/day 30 days	Change in modified pediatric rhino conjunctivitis and quality of life score
Lactobacillus paracasei-33.200–for[47]	400 mL/day 30 days	Change in modified pediatric rhino conjunctivitis and quality of life score
Lactobacillus acidophilus L-92.[48]	100 mL/day 8 weeks	Change in SMS perennial allergic rhinitis
Lactobacillus salivarius[33]	4 × 109 colony forming units/g/day 12 weeks	The scoring allergic rhinitis index (specific symptoms scores and SMS and blood parameters
Lactobacillus gasseri A5[49]	2 months/twice a day	Reduces rhinitis symptoms and drug usage in children with allergic rhinitis
Lactobacillus casei 3 × 109 CFU/g, Lactobacillus acidophilus 3 × 109 CFU/g, Lactobacillus rhamnosus 7 × 109 CFU/g, Lactobacillus bulgaricus 5 × 108 CFU/g, bifidobacterium breve 2 × 1010 CFU/g, bifidobacterium longum 1 × 109 CFU/g, and Streptococcus thermophilus 3 × 108 CFU/g, and 38.5 mg fructooligosaccharide[21]	1 capsule/day 8 weeks	Pulmonary function tests, IL-4 and IFN-γ levels, and expression of microRNAs were assessed at baseline and after treatment
Lactocare®, a synbiotic containing 1 billion CFU/Capsule of Lactobacillus casei, Lactobacillus rhamnosus, Streptococcus thermophilus, bifidobacterium breve, Lactobacillus acidophilus, bifidobacterium infantis, Lactobacillus bulgaricus, and Fructooligosaccharide[50]	1 capsule/day 8 weeks	Reduce episodes of viral infection in asthmatic children

Table 1. Probiotics as additives on therapy in reduce respiratory infections in asthmatic children

dietary supplements in pediatric asthma. This study suggests that further research should be conducted on the use of probiotics to prevent asthma attacks in children. Functional foods are defined as foods or dietary components that "can provide health benefits beyond the basic diet" and represent an easy, safe, effective, available, and affordable way to prevent respiratory infections in children.[23] This study aimed to measure the effects of probiotic candy sweets as functional foods on clinical symptoms and lung function in asthma patients.

METHOD AND MATERIALS

Study population

Children, aged between 7 and 11 years old, with mild intermittent asthma according to the Global Initiative for Asthma (GINA) invited to

study.[24] The asthma diagnosis was confirmed by a pediatric allergist. To be eligible, the children had no history of cardiac or pulmonary disease including cystic fibrosis and should have no history of recent antibiotic usage (during the past 2 months) or any probiotic consumption for 2 weeks.

Study design

The study was conducted as a double-blind randomized trial, with participants randomized to intervention or placebo groups on a 1:1 basis. Randomization and allocation of placebo and probiotic candy were performed by a researcher who had no contact with the participants. In this study, the block method was used to allocate the sample into 2 groups, and to generate a random sequence from http://www.graphpad.com/quickcalcs/index. Cfm was used.

The sample size was calculated based on the previous study Ahanchian et al.[4] The sample size was calculated by formula difference between 2 independent means (2 groups) and $\alpha = 0.05$, 80% power, and effect size d = 0.58, and it was estimated that at least 30 patients were required for each arm; the final sample size assuming a 20% dropout out was set as 40 individuals in each arm (consort diagram). The block method was used to allocate the sample into 2 groups and to generate a random sequence from http://www.graphpad.com/quickcalcs/index. Randomization and allocation of arm allocation were performed by a researcher who had no contact with the participants, and Cfm was used. Patients and visiting physicians were blinded to the participants' group allocations.

The allocation of each treatment group was kept confidential until final data analysis and all procedures, including randomization, patient enrollment, and allocation of individual patients to interventions, were performed independently of treatment allocation. The interventions were delivered by experienced nurses in the clinic.

Intervention

The probiotic candy had a colony count of 10^{10} CFU of *Bacillus* coagulans provided by the Institute of Food Science and Technology Mashhad, Iran. It is a new candy product supplemented with probiotic spore-forming bacteria that does not require special storage conditions or cold chain. This sweet product was developed using spore-forming probiotics with an edible biopolymer coating as a carrier. The spore-forming probiotics showed good stability during the manufacture and storage of the probiotic candy, a traditional dry sweet product. The candy containing probiotic bacteria was manufactured under dry environmental conditions. The candy were coated with an edible coating containing carboxymethylcellulose, sucrose, glucose syrup, fructose syrup and a probiotic strain. The coating was produced under thermal stress when dissolved in both dry and hot liquids, without any reduction in the viability of the spores used in the coated rock candy. The candies contained in this product had a 90.39% and 89.07% probability of the presence of *Bacillus* coagulans and *Bacillus subtilis* spores, respectively, after 6 months of storage at room temperature.

We aimed to recruit 80 children to be randomized into 2 groups, receiving active intervention (n = 40) or an identical placebo (n = 40). The treatment duration was 2 months from September 10 to November 10, the peak of respiratory infections and school participation in Mashhad. Participants with a respiratory illness were prescribed standard asthma treatment: β2 agonist aerosols and/or oral prednisolone. Participants with a respiratory illness were prescribed standard asthma treatment: β2 agonist aerosols and/or oral prednisolone. This trial was registered at the Iranian Registry of Clinical Trial (IRCT20101020004976N7) available at: https://www.irct.ir. Ethical approval was obtained from the Mashhad University Medical Sciences (IR.MUMS.MEDICAL.REC.1398.925) and informed written consent was obtained from all participants and their parents.

Assessment

The patients had a physical examination, and demographic and anthropometric data were collected. At the first visit, the child's height and weight were measured, and body mass index (BMI) was calculated as BMI = Weight (kg)/height (m)2. BMI-for-age percentile based on CDC growth charts for children and teens ages 2 through 19 years was calculated. The Childhood Asthma Control Test (c-ACT) was completed at the

baseline and the end of the study. The validity and reliability of this questionnaire have previously been reported.[25,26] The patients had scheduled monthly visits plus unscheduled visits at the time of any respiratory infection to assess upper respiratory symptoms including runny nose, sneezing, cough, and fever. The baseline data were compared with the data at the end of the first month and the end of the study.

The primary outcome was the number of respiratory infections during the study time. The secondary outcomes were the use of β2 agonist aerosols, the use of oral corticosteroids, school absenteeism, emergency admission, and the need for hospitalization. In this study, a checklist was given to the patients to check the side effects of probiotics. This checklist included gastrointestinal symptoms, such as vomiting, flatulence, diarrhea, and bowel pain. The patients were asked to contact the researcher if they had the previously listed symptoms. None of the patients reported these symptoms during probiotic and placebo administration.

Data analysis

SPSS software was used for data analysis. The normal distribution of data was assessed using Kolmogorov-Smirnov test. Descriptive data were expressed as mean ± SD or median (interquartile range) and number (percent). The chi-square test was used for comparing qualitative data. Student t-test or Mann-Whitney was used for the comparison of the quantitative data, according to the data distribution. The before-after comparison also was assessed by *t*-test and repeated measures ANOVA. The significance value was 0.05.

RESULTS

Eighty children with mild intermittent asthma were recruited, with 40 randomized to probiotic candy and 40 to placebo candy. Two patients in the probiotic group and 3 patients in the placebo group were excluded from the analysis due to loss of follow-up and 6 Excluded from analysis. Finally, 34 people in the placebo group and 35 people in the probiotic group were included in the statistical analysis (Fig. 1). There have been no full-size variations in baseline traits among the probiotic and placebo groups (Table 2).

The frequency of viral respiratory infections was significantly lower in patients in the probiotic candy group compared to patients in the placebo candy group in the first month (0.28 ± 0.45 vs. 0.51 ± 0.50, $p = 0.054$), the second month (0.08 ± 0.28 vs. 0.41 ± 0.49, $p = 0.001$), and the total study (0.37 ± 0.54 vs 0.90 ± 0.73, 0.001). (Table 3). The percent of patients with prednisolone consumption in the probiotic group was lower than the control group (2.9% vs. 14.7%) but did not show a significant difference ($p = 0.081$) and no difference was seen in the rate of emergency department visits and hospitalization between the two groups (both $p = 0.254$). The use of salbutamol and school absenteeism in the probiotic group was significantly lower than in the control group ($p = 0.040$ and $p = 0.046$, respectively. There was no significant difference in the evaluated scores for asthma control (c-ACT) in both probiotic and placebo groups. After the intervention, the difference between the two groups has become significant ($p < 0.05$). (Table 4). None of the patients reported side effects during probiotic and placebo administration.

DISCUSSION

In this study, the spore-forming probiotic *Bacillus* candy as an intervention or treatment was used for reducing respiratory infections in asthmatic children. Results from the present study show that probiotic candy as a functional food containing *Bacillus* coagulants was associated with fewer acute respiratory illnesses in children with mild intermittent asthma. The use of salbutamol and school absenteeism in the probiotic group were also significantly reduced. However, decreases in the need for oral corticosteroids, emergency department visits, or hospital admissions were not seen. Childhood Asthma Control Test (c-ACT) score reduced.

The microbiome plays an important role in the development of allergic diseases and many other diseases.[27-30] In asthma patients, dysbiosis in the digestive tract and the airways can be observed. Probiotics rebalance the immune response, restore gut microbiota dysbiosis, and reduce airway inflammation.[1] A Cochrane review showed that in children without asthma, probiotics can improve the immune system, especially mucosal immunity

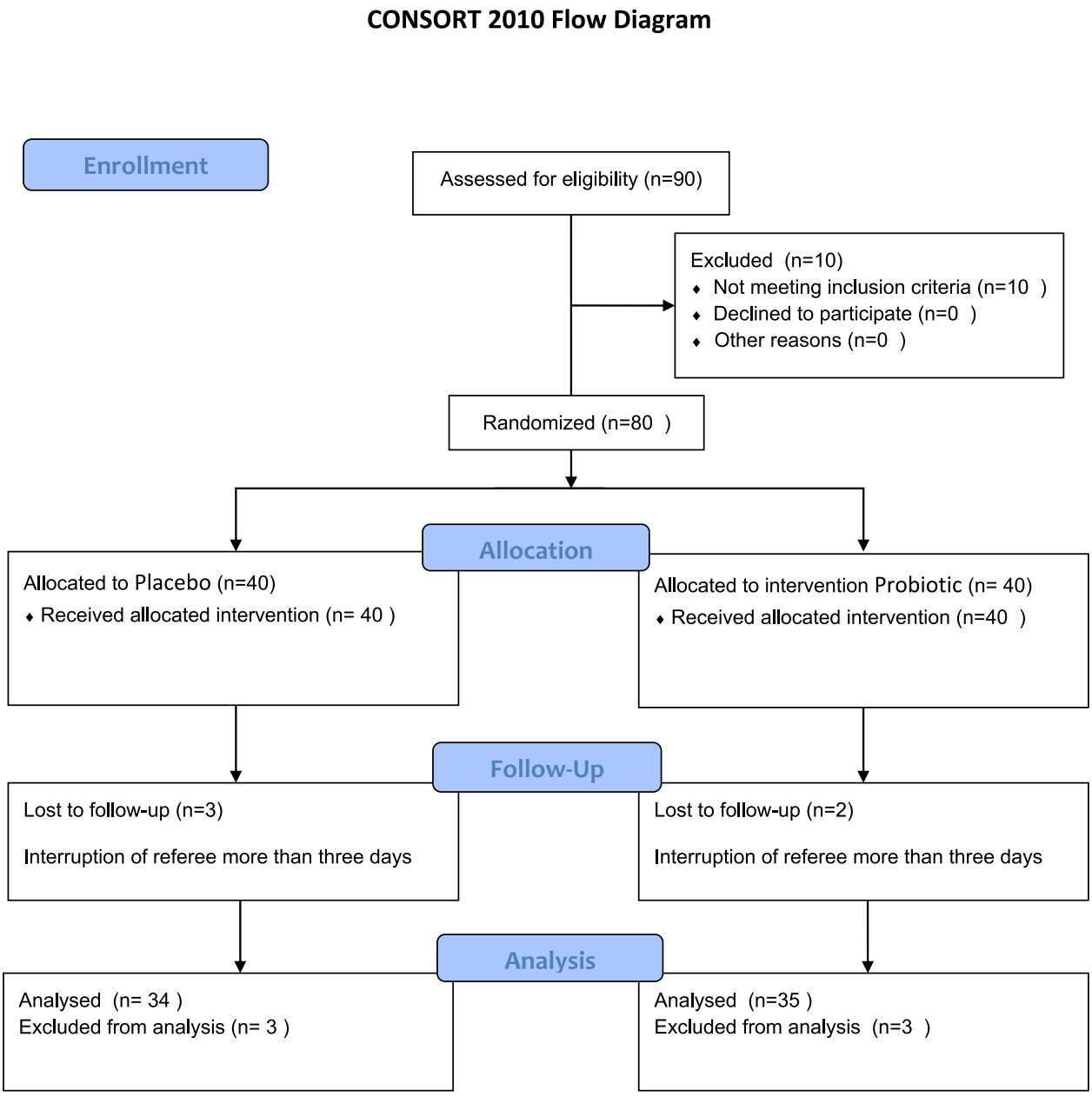

Fig. 1 Flow of participants consented to the each arm follow-up of the clinical trial.

(secretory IgA), reduce the number of colds, pharyngitis, epiglottitis, and laryngotracheitis, reduce the average duration of illness, and reduce school absences due to colds.[18] Based on this evidence and the understanding that viral infections are the main cause of asthma attacks, it has been suggested that the use of probiotics may reduce virus-related asthma attacks.

In 2023, Sadrifar et al showed that probiotics could be beneficial in managing asthma by potentially altering specific microRNA expressions and enhancing pulmonary function.[21] Dietary probiotics affect intestinal epithelial cells and immune cells via Toll-like receptors. This effect stimulates the synthesis of cytokines, mediators, and chemokines, including macrophage chemotactic protein-1, inducing mucosal immune activation. Mucosal immune activation is maintained by an increase in immunoglobulin A-secreting cells in mucosal tissues. Dietary probiotics also activate T cells and stimulate regulatory T cells to release IL-10,

	Variables	Placebo (n = 35)	Probiotic (n = 34)	P -value
Sex	Girl	17 (50.0%)	19(54.3%)	0.722[a]
	Boy	17(50.0%)	16(45.7%)	
Age, year		8.08 ± 1.31	8.20 ± 1.53	0.728[b]
Family history (n, %)		28 (82.4%)	31(88.6%)	0.463[a]
Weight, kg		30.28 ± 8.36	30.11 ± 8.81	0.935[b]
Height, cm		127.42 ± 8.66	127.85 ± 10.12	0.852[b]
BMI-for-age percentile based on CDC growth charts		18.46 ± 3.43	18.19 ± 3.59	0.749[b]
Diagnosis duration (by years)		4.00 ± 2.70	4.05 ± 2.83	0.932[b]

Table 2. Baseline characteristics in probiotic candy Probiotic and placebo groups. [a]*Chi-Square test* [b]t-test

Infection episodes	placebo	Probiotic	p-value[a]
First month	0.51 ± 0.50	0.28 ± 0.45	0.054
Second month	0.41 ± 0.49	0.08 ± 0.28	0.001
Total	0.90 ± 0.73	0.37 ± 0.54	0.001

Table 3. Respiratory infections (primary outcome) in probiotic candy and placebo groups. [a]t-test; *data presented as mean ± SD*

an important regulatory and anti-inflammatory cytokine.[31] Probiotics may enhance type 1 responses, downregulate IgE production, and strengthen immune responses to fight infections.[31,32] The results of a study by Chen et al in 2010 also showed that probiotic supplementation could reduce the production of TNF-α, IFN-γ, IL-12, and IL-13 by PBMCs after probiotic treatment.[33] Another study by Giovannini et al in 2007 showed that consumption of fermented milk containing *Lactobacillus casei* for 12 months did not change the duration of fever in asthmatic children.[34]

Although the results of early interventions were disappointing, subsequent attempts have been more successful. In 2010, Chen et al used probiotics in children with asthma and showed that patients receiving probiotics had significantly improved lung function and reduced clinical symptom scores for asthma and allergic rhinitis compared to the control group. Data reported by Chen et al. The reported results are similar to those of the present study, where probiotic sweets improved c-ACT(33). However, contradictory results have been reported by other studies. The reason for this difference seems to be that in the study Miraglia et al selected children with well-controlled asthma. Also Rose et al the long-term effects of the use of probiotics in young children were investigated and they selected only the children with well-controlled asthma (C-ACT >19).[35,36] Sadrifar in 2023 showed that treatment with probiotic supplementation improved Forced Vital Capacity and Forced Expiratory Volume. The findings of the study showed that probiotics can be used in addition to common asthma treatments.[21] In the present study, probiotic candy did not reduce oral corticosteroid usage, emergency department visits, or hospital admission. Gutkowski et al showed that asthmatic children received Trilac capsules containing *Lactobacillus acidophilus*, *Bifidobacterium bifidum*, and *Lactobacillus delbrueckii* subsp. bulgaricus) had improved pulmonary function tests and

Secondary outcomes		Placebo N = 34	Probiotics N = 35	p-value
Oral prednisolone (n, %)		5(14.7%)	1(2.9%)	0.081[a]
Salbutamol usage(mean ± SD)		0.52 ± 0.61	0.25 ± 0.44	0.040[b]
School absent		0.22 ± 0.42	0.05 ±0 .23	0.046
Emergency department visit		2(5.7%)	0	0.254[a]
Hospital admission (n, %)		2(5.7%)	0(0)	0.254[a]
c-ACT	Before	21.26 ± 1.60	20.77 ± 2.23	0.297
	After	21.82 ± 1.64	23.05 ± 1.78	0.004
	Difference	0.56 ± 1.74	2.28 ± 2.68	0.002

Table 4. Secondary outcome variables. [a]*Fisher's exact test* [b]t-test

reduced the duration of asthma exacerbations. The amount of bronchodilators used was also significantly reduced.[37]

The different results from different studies can be explained by the fact that the effect of probiotics depends heavily on the strain. The results can also depend on the dose, duration, genetics, diet and many other factors. Only specific strains with immunobiological properties and precise doses and durations can reduce the number of episodes of upper respiratory tract infections. In 2016, we tested a multi-strain synbiotic (lactocare®) containing *Lactobacillus casei*, *Lactobacillus rhamnosus*, *Streptococcus thermophilus*, *Bifidobacterium breve*, *Lactobacillus acidophilus*, *bifidobacterium asmais* in *bidobacillus bulgaricus*, and fructooligosaccharides in 72 children (6–12 years old) with mild persistent asthma. Asthmatic children in the probiotic group consistently used bronchodilators less during the study.[4] Italian researchers Miraglia et al in 2017 investigated the effects of a bifidobacterium mixture consisting of *Bifidobacterium longum* BB536, *Bifidobacterium infantis* M − 63, and *Bifidobacterium breve* M − 16V in children with intermittent asthma. These probiotics reduced respiratory symptoms and improved quality of life.[38] As the effect of probiotics is strain-dependent, some strains used in the Italian trial are immunobiotics selected for use in the current study. Consumption of probiotics Lactobacilli and Bifidobacteria are known as 2 effective probiotics for inflammatory factors. These probiotics have anti-inflammatory and immune-stimulating effect.[39] Another important feature is the ability of the strains to colonize the gut microbiota. Preliminary studies have shown that Bifidobacterium breve colonizes the intestinal tract.[40] Huang et al reported that two different probiotic strains (*Lactobacillus fermentum* GM-090 and *L. paracasei* GMNL-133) were associated with reduced asthma severity and asthma control test results after 3 months.[41]

Lin et al in a systematic review and meta-analysis revealed an overall reduction in episodes of asthma in the probiotics group than in the control group (risk ratio = 1.3). In this study, a decrease in IL-4 and an increase in IFN-γ were considerable and statistically significant, respectively after the treatment of probiotics. Hematological and immunological parameters such as the IgE – Total (ECL), T lymphocyte, and human peripheral blood mononuclear cells (PBMCs) derived cytokines were determined before and after the probiotics consumption. However, no effect was seen on the Asthma Control Test scores, asthmatic symptoms, the number of symptom-free days.[42] In another meta-analysis study probiotic consumption reduced the incidence of the common cold and influenza-like illness episodes but had no significant effect on decreasing the influenza episodes. Probiotic supplementation showed a promising beneficial role in the reduction of the duration and symptoms of influenza-like disease and common

cold.[19] More recently, the PROPAM Study used a probiotic mixture containing *Lactobacillus salivarius* LS01 and *Bifidobacterium breve* B632. Results demonstrated that probiotic strains *Ligilactobacillus salivarius* LS01 (DSM 22775) and *Bifidobacterium breve* B632 (DSM 24706) were safe and significantly reduced by more than a third the frequency of asthma exacerbations.[12]

Currently, there is a trend in many countries to incorporate natural ingredients into the diet and to use complementary and alternative medicine, especially for chronic diseases such as asthma. Functional foods, including probiotics, have been part of people's diets for centuries. Ancient physicians in the Middle East prescribed yogurt to cure stomach ailments and stimulate the appetite. A passage in the Old Persian Testament states that "Abraham owed his long life to the consumption of sour milk".[43]

Functional foods and probiotics gained popularity when Nobel Prize-winning microbiologist Ilya Metchnikov noted that people who regularly consumed fermented yogurt lived longer and suggested that this may be related to the beneficial effects of *Lactobacillus*. Fermented foods have been found to contain significant amounts of live, functional microorganisms, at least 10^6-10^9 microbial cells per gram. Among fermented foods, yogurt and kefir are two of the most attractive sources of probiotics, mainly from the genus *Lactobacillus*, *S. thermophilus* and *S. thermophilus*.[44] Fermented foods have been shown to suppress Th2-mediated allergic responses through the induction of CD4, CD25, and regulatory T cells in mice, and to reduce the production of Th2 cytokines.[45] In this study, probiotic candy was associated with a reduction in acute respiratory disease in asthmatic children, and while it is a simple adjunct therapy for children with mild intermittent asthma, probiotic candy prevented more severe asthma attacks and achieved asthma control scores (c-ACT). This study has some limitations. Mechanisms and immunological parameters were not investigated. The follow-up period was also short. It is not known what type of microorganisms are causing the respiratory infections. The advantage of this study is that, to our knowledge, probiotic candies are used for the first time, introducing a product that is available to consumers, easy to use, and capable of reducing mild asthma attacks in children. These candies may be a potential complementary therapy for primary care physicians.

CONCLUSION

The results of this study showed that supplementing standard asthma treatment with probiotic candy containing *Bacillus* coagulans as an alternative complementary medicine may have some benefits, including the reduction of acute respiratory diseases. Evidence regarding the benefits of probiotics is incomplete and contradictory. However, relatively few studies have been conducted and the methodology is inconsistent. Factors such as duration of treatment, study population and children's genetics, background diet, outcome variables, bacterial strains used (multiple or single strains), as well as clinical and functional characteristics of subjects, varied from study to study. Therefore, it is suggested that further studies using different methodologies should be conducted to draw more convincing conclusions about the benefits of probiotics in the treatment of childh asthma.

Abbreviations

MDI: Metered-dose inhalers; c-ACT: Asthma Control Questionnaire; URTIs: Upper respiratory tract infections; GINA: Global Initiative for Asthma; BMI: body mass index.

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Consent for publication

All of authors declared consent for publication in WAO.

Consent

A written informed consent was obtained from each study participant and their parents.

Data availability

The data that support the findings of this study are available from the corresponding author upon request.

Author's contributions

The study was conceptualized and designed by **Rana Tafrishi and Hamid Ahanchian**. **Seyed Ali Jafari, Hamidreza Kianifar** and **Mohammadali Kiani** were

responsible for the execution of the study, including participant recruitment. The data analysis and manuscript writing were carried out by Tahereh Sadeghi. **Nasrin Moazzen and Abolfazl Pahlevanloo** contributed to the study by conducting participant recruitment, preparing test meals, and collecting data, as well as actively engaging in manuscript writing. Additionally, Hamid Ahanchian and **Peter D Sly** played a role in the study's conception and design, manuscript writing, and manuscript revision. All authors thoroughly reviewed and provided their approval for the final manuscript.

Ethics approval

The research ethics committee at Mashhad University of Medical Sciences approved the study protocol (IR.MUMS.MEDICAL.REC.1398.925).

Declaration of competing interest

The authors have no conflicts of interest to report.

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