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Probiotic Supplementation and Respiratory Infection and Immune Function in Athletes: Systematic Review and Meta-Analysis of Randomized Controlled Trials

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Objective: To evaluate the effectiveness of probiotic supplementation on upper respiratory tract infection (URTI) and inflammatory markers in elite athletes.

Data Sources: We searched the PubMed, EBSCOhost, Scopus, and Web of Science databases using the following terms: *probiotic* OR *probiotics* AND *exercise* OR *sport* OR *athletes* AND *URTI* OR *respiratory infection* OR *URTIs* OR *inflammation* OR *inflammatory* OR *cytokines*.

Study Selection: We screened the titles and abstracts of 2498 articles using our inclusion criteria. A total of 14 articles were selected for further analysis.

Data Extraction: For each study, 2 independent reviewers extracted the study design, participant characteristics, inclusion and exclusion criteria, intervention characteristics, outcome measures, and main results.

Data Synthesis: We did not observe an effect of probiotic supplementation on the number of days of illness or the mean number or duration of URTI episodes but did note an effect of probiotic supplementation on the total symptom severity score (–0.65; 95% CI = –1.05, –0.25; P = .02). Lower levels of interleukin 6 (–2.52 pg/mL; 95% CI = –4.39, –0.66 pg/mL; P = .002) and tumor necrosis factor α (–2.31 pg/mL; 95% CI = –4.12, –0.51 pg/mL; P = .01) were also reported after supplementation.

Conclusions: This meta-analysis provides evidence that probiotic supplementation, especially among professional athletes, is an effective way to decrease the total URTI symptom severity score. In addition, probiotic supplementation may decrease interleukin 6 and tumor necrosis factor α levels. More studies involving larger groups are needed to better assess this effect. The optimal timing, duration, composition, and dose of such supplementation need to be determined.

Key Words: upper respiratory tract infection, inflammation, interleukin 6, tumor necrosis factor α

Key Points

- Probiotic supplementation, especially among professional athletes, effectively decreased the total symptom severity score of upper respiratory tract infection, especially when taken as a single-strain probiotic.
- Probiotic supplementation may decrease levels of interleukin 6 and tumor necrosis factor α.
- More studies with larger groups are needed to better evaluate this effect.
- Determination of the optimal timing, duration, composition, and dose of such supplementation is needed.

Among athletes, heavy, acute, and prolonged intense physical exercise; insufficient rest and sleep; emotional stress; and inadequate nutrition may generate serious health problems, such as inflammation or respiratory infections, that may affect their physical performance and sports achievements.[1] Researchers[2] have observed that such factors can lower the resistance of athletes by reducing the number and activity of natural killer cells, decreasing neutrophil activity, impairing proliferation of T lymphocytes, decreasing the level of anti-inflammatory cytokines, and increasing levels of proinflammatory cytokines and salivary immunoglobulin A (IgA), inducing respiratory tract infections. Indeed, many elite athletes reported substantial bouts of infections that interfered with their ability to compete and train.[2] Epidemiologic studies[3–5] also indicated that extensive training was associated with an increased risk of upper respiratory tract infection (URTI). Therefore, dietary strategies to improve the immune function of athletes and reduce their risk of URTI have been sought for many years.

Probiotics are live microorganisms that, when administered in adequate amounts, may confer a health benefit on the host.[4] In a meta-analysis involving general adult populations, Hao et al[5] showed that probiotics were better than a placebo in reducing the number of participants experiencing episodes of acute URTIs, the rate ratio of episodes of acute URTI, and antibiotic use. This suggests that probiotics may be more beneficial than a placebo for preventing acute URTIs.[5] Wang et al[6] also found that probiotic supplementation in children led to fewer days of URTI and fewer days absent from day care or school than did a placebo.

Furthermore, many investigators have demonstrated the beneficial effect of probiotic supplementation on inflammatory markers in the general population. For example, in a systematic review and meta-analysis, Kazemi et al[7] showed that probiotics and synbiotics decreased some inflammatory markers. The intervention was most effective in reducing C-reactive protein (CRP) and tumor necrosis factor (TNF)-α in healthy or ill general populations.[8] The use of probiotics in physically active individuals might serve as

a strategy to further improve respiratory symptoms and inflammatory status and consequently enhance the physical performance of athletes.[8] In a recent review, Sivamaruthi et al[9] indicated that probiotic supplementation improved the immune system and reduced the severity and incidence of URTIs in athletes, but to the best of our knowledge, no authors have confirmed these results via a systematic literature review or meta-analysis. Therefore, the aim of our study was to evaluate the effectiveness of probiotic supplementation on respiratory infection and inflammatory markers in elite athletes based on data from randomized controlled trials and crossover studies.

METHODS

This systematic review was designed according to the Preferred Reporting Items for Systematic Reviews and Meta-analyses (PRISMA) recommendations and registered in the International Prospective Register of Systematic Reviews (No. CRD4202017669).

Search Strategy

We searched for relevant articles published through the end of September 2020 using the PubMed, EBSCOhost, Scopus, and Web of Science databases between February 1 and September 30, 2020. The databases were searched using the following key words and their various combinations: *probiotic* OR *probiotics* AND *exercise* OR *sport* OR *athletes* AND *URTI* OR *respiratory infection* OR *upper respiratory tract infections* OR *inflammation* OR *inflammatory* OR *cytokines* without restrictions. We also reviewed the reference lists of related reviews and original articles. The full search strategy is described in Figure 1.

Study Selection

The inclusion criteria were based on the Population, Intervention, Comparison, Outcome (PICO) framework.[10] Studies were included if they (1) involved healthy adult professional athletes of both sexes ($\geq$18 years); (2) incorporated interventions using probiotics; (3) involved a control or placebo group; (4) examined outcomes not previously defined (as an open question: all outcomes were reported in the studies); (5) focused on inflammatory variables, respiratory infections, or both; and (6) were randomized controlled trials (RCTs) and/or crossover studies.

Data Extraction

We independently performed a literature search and selection of articles. Publications were evaluated according to the titles, abstracts, and full texts in subsequent stages. Each selected publication was studied critically. During the data-abstraction process, we made no attempt to contact the authors for further information beyond what had been published. Discrepancies were resolved through consensus or arbitration. The data extracted from each study were as follows: authors, year of publication, cohort age and sex, exercise, intervention and control groups, main outcome, and results.

Bias Assessment

We used the Cochrane risk of bias assessment tool to judge the methodologic quality of each trial with the aim of evaluating the performance and methods of randomization, the extent of blinding (whether it affected data collectors, data analysis, outcome assessors, or participants), allocation concealment, incomplete outcome data, selective reporting, and other possible sources of bias. In line with the Cochrane handbook[11] criteria for judging bias risk, each study was classified as having a high, low, or unclear risk of bias.

Statistical Analysis

Statistical analysis was carried out using the Statistica (version 13.0; StatSoft) software. The therapeutic effect of probiotic supplementation on URTI and cytokine levels compared with the placebo was estimated using the standardized mean difference with a 95% CI. We examined publication bias through visual inspection of a funnel plot and then using the Egger test[12]; $P < .05$ indicated a significant publication bias. Heterogeneity across studies was measured using the Cochran Q statistic, with $P < .10$ implying a significant difference, and the I^2 statistic, with $I^2 = 0\%$ indicating *no heterogeneity* and $I^2 = 100\%$ indicating *maximal heterogeneity*. A random-effects model was selected when $I^2 \geq 50\%$. All statistical tests were 2 sided, and P values $<.05$ were considered statistically different.

RESULTS

Search Results

From the initial search strategy, we identified 2501 articles. A total of 14 RCTs or crossover trials met the inclusion criteria and were included in the final meta-analysis (Figure 1).

Population and Study Characteristics

Supplements in capsules[13–21] or beverages[19,22–25] were used in most studies. Sachets with probiotic bacteria were used in 1 study.[26] The number of study participants, their characteristics, duration of the intervention, type of supplement, and sport discipline are presented in Table 1. A total of 1309 study participants, 771 study participants from the supplemented group (PRO) and 538 from the placebo group (PLA), were involved in the 14 selected investigations. The mean age of participants ranged from 20.1 $\pm$ 1.5 years[20] to 37 $\pm$ 11 years.[26] Interventions were based on supplementation of probiotic bacteria, such as *Bifidobacterium animalis*,[16,26] *B. bifidum*,[13,16,17,25] *B. lactis*,[17] *B. longum*,[13] *Lactobacillus acidophilus*,[16,17,26] *L. casei*,[17,22] *L. casei* Shirota,[23,24] *L. fermentum*,[17,18] *L. gasseri*,[13] *L. helveticus*,[15] *L. plantarum*,[14,17] *L. rhamnosus*,[17,19] *L. salivarius*,[21] *Saccharomyces boulardii*,[17] *Streptococcus thermophilus*,[17] and *Bacillus subtilis*.[20] Although the publication dates of the selected papers were unrestricted, all articles in this systematic review were published after 2007.

Effect of Probiotic Supplementation on Upper Respiratory Tract Infection

The 8 studies[13,15,18,19,21,23,24,26] included in our meta-analysis measured the effects of probiotic supplementation

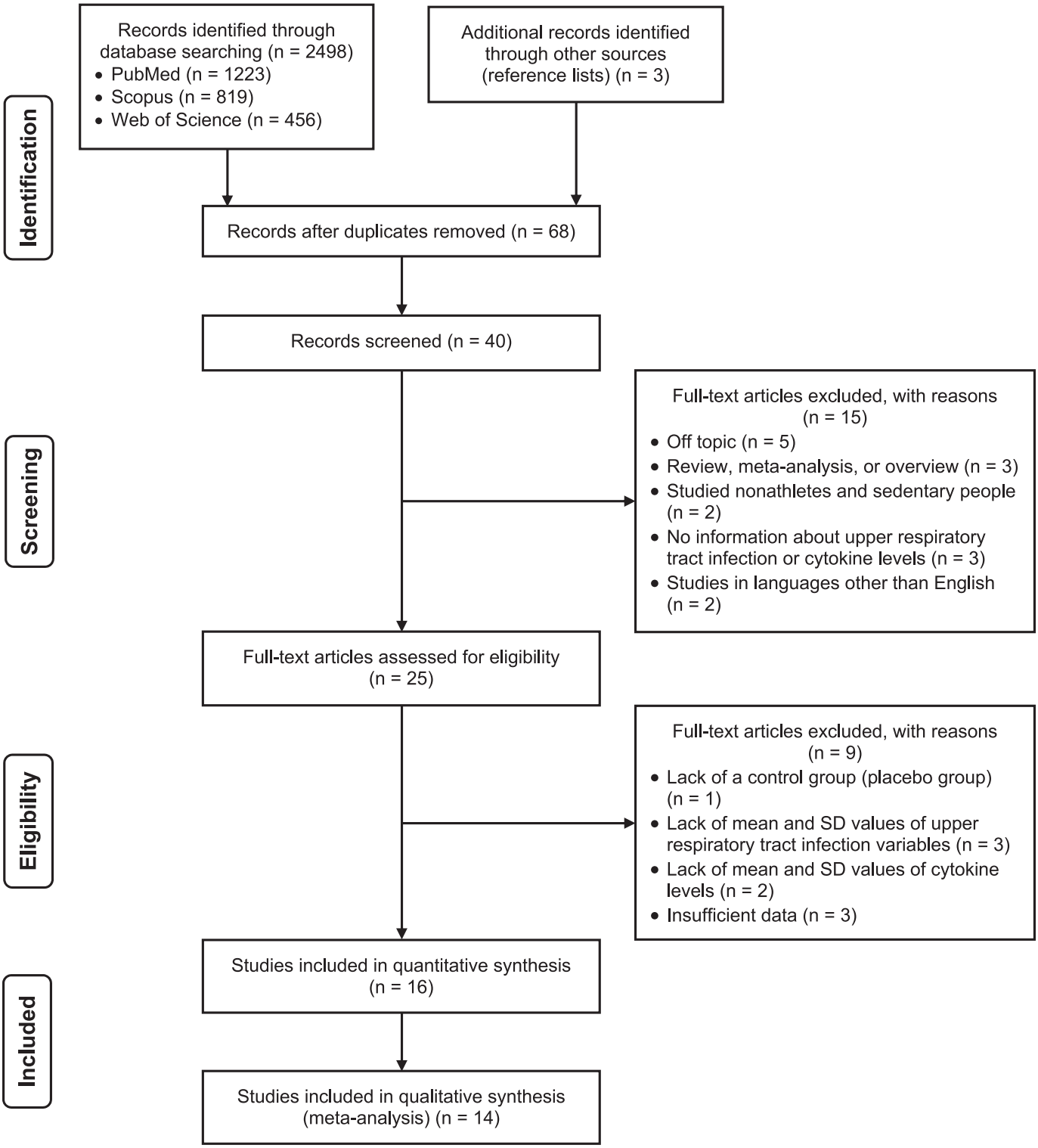

Figure 1. Study selection process.

on URTI. Data from the included articles allowed us to assess the effects of probiotic supplementation on the number of days of illness, the number of URTI episodes, total symptom severity score, and mean duration of URTI symptoms. The Begg rank correlation test (number of days of illness: $P = .99$; number of URTI episodes: $P = .14$; total symptom severity score: $P = .99$; mean duration of URTI symptoms: $P = .29$) and Egger linear regression test (number of days of illness: $P = .66$; number of URTI episodes: $P = .31$; total symptom severity score: $P = .10$; mean duration of URTI symptoms: $P = .84$) suggested that no publication bias was present.

In the groups receiving supplements, the days of illness, number of URTI episodes, total symptom severity score, and duration of URTI symptoms among supplemented groups ranged from 0.37 ± 0.72[18] to 5.3 ± 6.8 days,[19]

Table 1. Summary of Studies of Probiotics for Upper Respiratory Tract Infection and Cytokines Concentrations in Athletes Compared With Control Participants (N = 1309) Continued on Next Page

Study, Type	Population	Intervention	Control	Main Outcome Measure	Intergroup Differences, Mean ± SD		Adverse Effect
					PRO	PLA	
Gill et al[22] (2016), crossover	Male runners (age = 26 ± 6 y) PRO: n = 8 PLA: n = 8	Beverage containing 1×10^{11} *Lactobacillus casei* x 1 wk	Beverage without *Lactobacillus casei*	IgA, mg/L IgA secretion rate, μg/min	527 ± 317 260 ± 183	505 ± 170 264 ± 182	[a]
Gleeson et al[23] (2011), parallel	Male and female runners, cyclists, swimmers, triathletes, racket sports, and team gamers PRO: n = 32, age = 32 ± 14 y PLA: n = 26, age = 25 ± 9 y	Drink containing 6.5×10^9 *Lactobacillus casei* Shirota x 16 wk	Drink without *Lactobacillus casei* Shirota	Episodes of URTI Total symptom severity score Duration of URTI, d	1.2 ± 1.0 49 ± 55 7.9 ± 5.5	2.1 ± 1.2 50 ± 32 7.6 ± 3.2	[a]
Gleeson et al[21] (2012), parallel	Male and female runners, cyclists, swimmers, triathletes, racket sports, and team gamers PRO: n = 27, age = 25 ± 5 y PLA: n = 27, age = 24 ± 4 y	Capsules with 2×10^{10} *Lactobacillus salivarius* CFUs, 1.78 maltodextrin, and 0.01 g magnesium stearate x 16 wk	0.78 maltodextrin and 0.01 g magnesium stearate	Episodes of URTI 8-wk IgA, mg/L 8-wk IgA secretion rate, μg/min 16-wk IgA 16-wk IgA secretion rate, μg/min	1.5 ± 2.3 127 ± 101 63 ± 36 123 ± 62 mg/mL 57 ± 29	1.4 ± 1.5 143 ± 95 63 ± 41 128 ± 58 mg/L 59 ± 32	[a]
Gleeson et al[24] (2016), parallel	Male and female runners, cyclists, swimmers, triathletes, and team gamers PRO: n = 126 (58% men), age = 20.3 ± 0.2 y PLA: n = 117 (59% men), age = 20.6 ± 0.2 y	Drink with 6.5×10^9 *Lactobacillus casei* Shirota x 20 wk	Drink without *Lactobacillus casei* Shirota	Episodes of URTI Total symptom severity score Duration of URTI symptoms, d	0.7 ± 0.1 62 ± 5 5.6 ± 0.4	0.6 ± 0.1 64 ± 6 5.9 ± 0.5	[a]
Haywood et al[13] (2014), parallel	Elite rugby union players (age = 24.7 ± 3.6 y) PRO: n = 30 PLA: n = 30	Capsules with 2.6×10^9 *Lactobacillus gasseri* CFUs, 0.2×10^9 *Bifidobacterium bifidum* organisms, and 0.2×10^9 *Bifidobacterium longum* organisms x 4 wk	Capsules without probiotic bacteria	Days of illness Total symptom severity score Duration of URTI symptoms, d	3.4 ± 4.6 8.7 ± 25.1 4.6 ± 3.1	5.8 ± 6.6 13.33 ± 14.6 7.2 ± 6.9	[a]
Huang et al[14] (2019), parallel	Male triathlon teams Study I PRO: n = 9; age range = 19–24 y PLA: n = 9; age range = 19–21 y Study II PRO: n = 8; age range, 19–21 y PLA: n = 8; age range, 19–26 y	Capsules with 1.5×10^{10} *Lactobacillus plantarum* PS128 CFUs and 100 mg excipient of microcrystalline cellulose x 8 wk	400 mg excipient of microcrystalline cellulose	Study I, pg/mL IL-6 IL-10 TNF-α Study II, pg/mL IL-6 IL-10 TNF-α	 6.6 ± 0.7 9.4 ± 1.9 10.5 ± 0.8 14.1 ± 1.3 131.7 ± 14 15.2 ± 1.9	 9.3 ± 1.2 8.7 ± 1.1 14.4 ± 1.8 19.7 ± 2.2 84.6 ± 1.8 22.1 ± 2.4	[a]

Table 1. Continued From Previous Page

Study, Type	Population	Intervention	Control	Main Outcome Measure	Intergroup Differences, Mean ± SD		Adverse Effect
					PRO	PLA	
Kekkonen et al[19] (2007), parallel	Male and female marathon runners PRO: n = 61, mean age = 40 y (range = 22–58 y) PLA: n = 58, mean age = 40 y (range = 23–69 y)	3×10^8 *Lactobacillus rhamnosus* LGG CFU/milk-based fruit drink or 5×10^9 CFU/capsule x 12 wk	Milk-based fruit drink or capsule without probiotic bacteria	Days of illness Episodes of URTI Duration of URTI symptoms, d	5.3 ± 6.8 0.7 ± 0.9 7.9 ± 7.1	3.9 ± 5.9 0.5 ± 0.7 6.3 ± 4.3	NR
Khani et al[25] (2018), parallel	Male sprint athletes (age = 21 ± 3 y) PRO: n = 53 PLA: n = 3	2×10^9 *Bifidobacterium bifidum* (BIB2) CFUs x 12 wk	Fruit juices without probiotic bacteria	IgA, mg/L	253 ± 83	194 ± 42 mg/L	[a]
Michalickova et al[15] (2016), parallel	Badmintonists, swimmers, cyclists, alpinists, athletes, karateists, kayakers, judoists, triathletes (men and women) PRO: n = 20, age = 23.5 ± 2.7 y PLA: n = 19, age = 22.8 ± 2.5 y	2×10^{10} *Lactobacillus helveticus* Lafti L10 CFUs x 14 wk	1% magnesium stearate and 99% maltodextrin	Episodes of URTI Total symptom severity score Duration of URTI symptoms, d IL-10, pg/mL	4.92 ± 1.96 110.92 ± 96 7.25 ± 2.90 365.2 ± 75.07	6.91 ± 1.22 129.73 ± 40.33 10.64 ± 4.67 434.8 ± 71.51	NR
Pugh et al[16] (2019), parallel	Male and female runners PRO: n = 12; age = 34.8 ± 6.9 y PLA: n = 12; age = 36.1 ± 7.5 y	25×10^9 *Lactobacillus acidophilus*, *Bifidobacterium bifidum*, and *Bifidobacterium animalis* subsp *lactis* CFUs x 4 wk	Cornstarch	IL-6, pg/mL IL-10, pg/mL	10.96 ± 7.86 5.27 ± 3.91	13.58 ± 12.9 5.78 ± 3.26	[a]
Pumpa et al[17] (2019), parallel	Elite rugby union athletes PRO: n = 9; age = 27.0 ± 3.2 y PLA: n = 10; age = 26.6 ± 2.9 y	60×10^9 viable bacteria *Lactobacillus rhamnosus*, *L. casei*, *L. acidophilus*, *L. plantarum*, *L. fermentum*, *Bifidobacterium lactis*, *B. bifidum*, and *Streptococcus thermophilus* and 250 mg of *Saccharomyces boulardii* x 17 wk	Microcrystalline iron oxide yellow, iron oxide red, and gelatin capsule and SB Floractiv[b] (microcrystalline cellulose, lactose, calcium hydrogen phosphate dihydrate, povidone, silica colloidal anhydrous, magnesium stearate, and gelatin capsule)	Stage I: IgA, mg/L Stage II: IgA, mg/L	487.2 ± 173.1 452.4 ± 166.3	523.2 ± 192.3 480.7 ± 200.6	NR
Townsend et al[20] (2018), parallel	Male baseball players (age = 20.1 ± 1.5 y) PRO: n = 13 PLA: n = 12	1.2×10^9 *Bacillus subtilis* CFU/capsule x 12 wk	Maltodextrin	IL-10, mg/L TNF-α, mg/L IgA secretion rate, μg/min	2.89 ± 1.08 2.07 ± 0.76 176.6 ± 86.6	3.27 ± 1.02 2.78 ± 0.95 156.1 ± 98.3	NR

Table 1. Continued From Previous Page

Study, Type	Population	Intervention	Control	Main Outcome Measure	Intergroup Differences, Mean ± SD		Adverse Effect
					PRO	PLA	
West et al[18] (2011), parallel	Male cyclists and triathletes PRO: n = 29; age = 35.2 ± 10.3 y PLA: n = 33; age = 36.4 ± 8.9 y	1×10^{10} *Lactobacillus fermentum* (VRI-003 PCC) CFUs x 11 wk	Microcrystalline cellulose	Days of illness	0.37 ± 0.72	1.26 ± 1.9	[a]
				Episodes of URTI	0.6 ± 0.9	1.0 ± 1.5	
				Total symptom severity score	1.23 ± 0.36	1.65 ± 0.57	
				Duration of URTI symptoms, d	3.5 ± 6.6	7.4 ± 10.3	
				IL-6, pg/mL	0.92 ± 2.14	1.22 ± 2.1	
				IL-10, pg/mL	0.95 ± 1.69	1.16 ± 1.44	
				TNF-α, pg/mL	1.27 ± 2.02	1.66 ± 2.22	
	Female cyclists and triathletes PRO: n= 18; age = 36.5 ± 8.6 y PLA: n = 17; age = 35.6 ± 10.2 y			Days of illness	1.98 ± 2.63	1.08 ± 1.32	
				Episodes of URTI	1.6 ± 2.1	0.5 ± 0.8	
				Total symptom severity score	1.38 ± 0.45	2.07 ± 0.77	
				Duration of URTI symptoms, d	12.2 ± 14.8	5.1 ± 14.7	
				IL-6, pg/mL	0.71 ± 2.93	2.29 ± 4.8	
				IL-10, pg/mL	0.89 ± 1.84	1.45 ± 2.03	
				TNF-α, pg/mL	1.15 ± 2.0	1.71 ± 1.8	
West et al[26] (2014), parallel	Male and female athletes PRO: n = 161; age = 36 ± 12 y PRO1: n = 155; age = 36 ± 11 y PLA: n = 149; age = 37 ± 11 y	PRO: 2×10^9 CFU of *Bifidobacterium animalis* subsp *lactis* BI-04 PRO1: 1×10^{10} *Lactobacillus acidophilus* NCFM and *Bifidobacterium animalis* subsp *lactis* Bi-07 sucrose base CFU x 11 wk	Sucrose base without the probiotic bacteria	Days of illness	PRO: 4.4 ± 1.3 PRO1: 4.3 ± 1.9	4.1 ± 1.6	NR
				Duration of URTI symptoms, d	PRO: 6.1 ± 3.3 PRO1: 6.2 ± 4.8	5.7 ± 3.6	

Abbreviations: CFU, colony-forming unit; IgA, immunoglobulin A; IL, interleukin; NR, not reported; PLA, placebo group; PRO, probiotic group; TNF, tumor necrosis factor; URTI; upper respiratory tract infection.

[a] Adverse effects were not estimated.

[b] FIT-BioCeuticals Ltd.

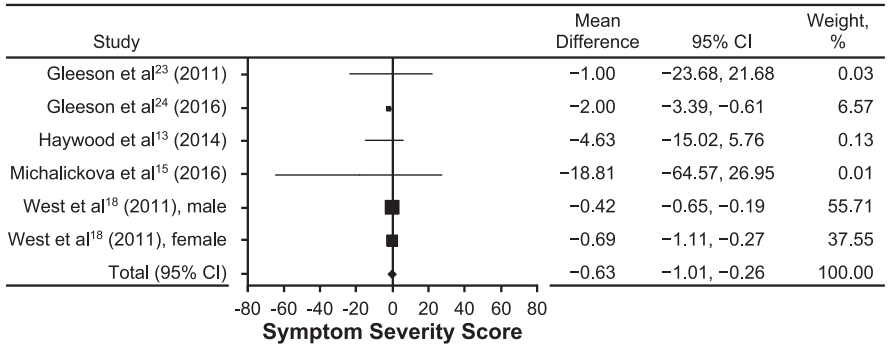

Figure 2. Effects of probiotic supplementation on total symptom severity score in athletes with upper respiratory tract infections.

from 0.6 ± 0.9[18] to 4.92 ± 1.96,[15] from 1.23 ± 0.36[18] to 110.92 ± 96,[15] and from 3.5 ± 6.6[15,18] to 12.2 ± 14.8 days,[18] respectively. In the placebo group, respective ranges were from 1.08 ± 1.32[18] to 5.8 ± 6.6 days[13] for days of illness, from 0.5 ± 0.7[19] to 6.91 ± 1.22[15] for number of URTI episodes, from 1.65 ± 0.57[18] to 129.73 ± 40.33[15] for total symptom severity score, and from 5.1 ± 14.7[18] to 10.64 ± 4.67[15] days for duration of URTI symptoms.

We did not observe any effect of probiotic supplementation on the number of days of illness or on mean number or duration of URTI episodes, but we did note an effect of probiotic supplementation on total symptom severity score (Figure 2; Table 2). The effect remained after the analysis was performed on single-strain supplements only[15,18,23,24] (Figure 3; Table 2).

Effect of Probiotic Supplementation on Inflammatory Measures in Athletes

Nine studies[14–18,20–22,25] in the meta-analysis quantified the effect of probiotic supplementation on inflammatory marker levels. Data from these articles allowed us to assess the effects of probiotic supplementation on interleukin (IL) 6, IL-10, TNF-α blood, and salivary IgA levels. The Egger linear regression test (IL-6: $P = .94$; IL-10: $P = .06$; TNF-α: $P = .16$; IgA: $P = .74$; salivary IgA: $P = .40$) suggested that there was no publication bias.

After intervention in the supplemented group, mean IL-6 ranged from 0.71 ± 2.93[18] to 14.1 ± 1.3[14] pg/mL, IL-10 from 0.89 ± 1.84[18] to 365.2 ± 75.07[15] pg/mL, TNF-α from 1.15 ± 2.0[18] to 15.2 ± 1.9[14] pg/mL, IgA from 123 ± 62[21] to 527 ± 317[22] mg/L, and salivary IgA from 57 ± 29[21] to 260 ± 183[22] μg/min. In the placebo group, mean ranged from 1.22 ± 2.1[18] to 19.7 ± 2.2[14] pg/mL for IL-6, from 1.16 ± 1.44[18] to 434.8 ± 71.51[15] pg/mL for IL-10, from 1.66 ± 2.22[18] to 22.1 ± 2.4[14] pg/mL for TNF-α, from 128 ± 58[21] to 523.2 ± 192.3[17] mg/L for IgA, and from 59 ± 32[21] to 264 ± 182[22] μg/min for salivary IgA (Table 1).

The meta-analysis did not show an effect of probiotic supplementation on IL-10 and IgA levels, but we found an effect of probiotic supplementation on IL-6 ($I^2 = 81.99\%$, Q = 22.21, $P = .002$) and TNF-α ($I^2 = 91.92\%$, Q = 49.49, $P = .01$; Figures 4 and 5, Table 3).

Bias Assessment

Most studies (93%) described the randomization process in detail and were judged as having a low risk of bias for this domain. Only 1 study demonstrated an unclear risk of bias.[25] Full details are presented in the Supplementary Table (see Supplemental Table, available online at http://dx.doi.org/10.4085/1062-6050-0592.20.S1).

DISCUSSION

Modulation of the immune system to increase defenses against URTIs is one of the most extensively researched areas in professional sports. We showed that probiotic supplementation positively affected both IL-6 and TNF-α levels. After probiotic supplementation, especially when single-strain probiotics were used, the total symptom severity score of respiratory infections was lower than in the placebo groups. However, we detected no effect of probiotic supplementation on the days of illness, number of URTI episodes, duration of URTI symptoms, or IL-10 and IgA levels (whether in blood or saliva).

In addition to the investigations cited in this meta-analysis, several other RCTs assessing the effects of probiotic supplementation on URTI and cytokine levels in elite athletes have been published. For example, Cox et al,[27]

Table 2. Effect of Probiotic Supplementation on Upper Respiratory Tract Infections

Variable	Meta-Analysis, Mean Difference (95% CI)	Value, %		P Value	No. of Studies
		I^2	Q		
Total symptom severity score	−0.63 (−1.01, −0.26)	27.21	6.87	.02	5
Single strain	−0.64 (−1.04, −0.24)	36.18	6.27	.02	4
Days of illness	0.04 (−0.49, 0.58)	65.97	14.69	.88	4
Single-strain probiotic	0.25 (−0.17, 0.59)			.75	2
Multistrain probiotic	0.21 (−0.18, 0.60)			.30	2
Upper respiratory tract infection episodes (single strain only)	−0.21 (−0.61, 0.19)	82.48	34.25	.30	5
Duration of upper respiratory tract infection symptoms	−0.20 (−0.95, 0.56)	65.87	23.44	.61	7
Single strain	−0.51 (−2.04, 1.02)			.51	5
Multistrain	0.1 (−0.91, 1.11)			.85	2

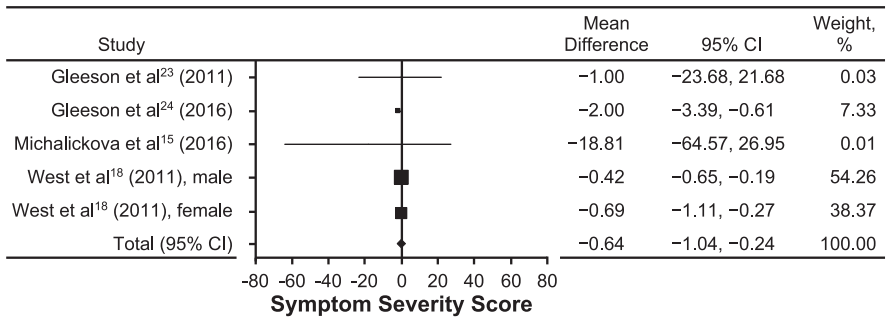

Figure 3. Effects of probiotic supplementation (single-strain probiotics only) on total symptom severity score in athletes with upper respiratory tract infections.

Komano et al,[28] and Strasser et al[29] reported that supplementation was associated with a reduction in the number of days, severity of respiratory illness, and incidence of URTI. Lamprecht et al[30] determined that probiotic supplementation beneficially affected TNF-α but not IL-6; this was in contrast to Jäger et al,[31] who showed that probiotic supplementation resulted in an overall decrease in circulating IL-6. We excluded the IL-10 data of Michalickova et al[15] from analysis because the mean value of this variable was extremely high. Shing[32] identified no effects of probiotic supplementation on plasma concentrations of IL-6, IL-10, and TNF-α. However, given the lack of mean and SD values, we could not use these data in our meta-analysis.

Respiratory infections are known to substantially reduce the sport performance of athletes. We found that one of the basic reasons for the development of infections was weakening of the effectiveness of both specific and nonspecific humoral immune mechanisms related to the mucous membranes. Pyne and Gleeson[33] demonstrated that this manifested predominantly as a decrease in secreted salivary IgA, which has been linked to a higher incidence of URTI. In their 1-year observational study, Fahlman et al[34] suggested that a season of football training could result in decreases in both salivary IgA and the secretion rate of salivary IgA, as well as an increase in the incidence of URTI. Probiotic supplementation has received attention because of the suggestion that it can stimulate T-cell immune responses in vitro, which is important because many researchers[35–37] have shown that athletes have decreased numbers of T cells after intensive anaerobic exercise. In their review, Wosinska et al[35] emphasized that *Bacteroides acidifaciens* induced IgA production in murine models and, as a consequence, elevated the production of IgA+ B cells and B cells. These findings were significant because IgA plays a pivotal role in maintaining intestinal homeostasis, primarily by preventing the adherence of pathogens in the intestine. These results may suggest the mechanism of probiotic supplementation in the prevention of URTI. Unfortunately, our findings were the opposite of these reviews and reflected no increases in salivary or IgA blood levels after probiotic treatments compared with placebo. Given the limited number of publications, we were unable to perform subgroup analysis by sex, age group, design and duration of intervention or dose and type of probiotics; these factors may also have affected our findings.

Whereas recreational and moderate exercise may have anti-inflammatory and immunomodulatory effects, the intense exercise of elite athletes can induce inflammation through the synthesis and release of many cytokines (IL-6, IL-1β, macrophage inflammatory protein-1α, IL-8, TNF-α, IL-10, and IL-1 receptor antagonist).[38] In a recent meta-analysis that included adults with diabetes, Tabrizi et al[39] reported that probiotic supplementation lowered serum CRP and TNF-α and increased nitric oxide levels but did not affect IL-6 levels. However, in a meta-analysis, Milajerdi et al[40] showed that probiotic supplementation reduced serum concentrations of proinflammatory cytokines, including IL-6, IL-12, IL-4, high-sensitivity CRP, and TNF-α, but did not affect IL-1β, IL-8, interferon-γ, or IL-17 concentrations. In their meta-analysis, Nazari et al[41] determined that probiotic consumption resulted in decreased plasma concentrations of IL-6 and TNF-α without an increase in interferon-γ (standardized mean difference = 0.43; 95% CI = 0.09, 0.76; $P = .01$). Milajerdi et al[40] also documented increased serum concentrations of IL-10 as an anti-inflammatory cytokine after probiotic supplementation.

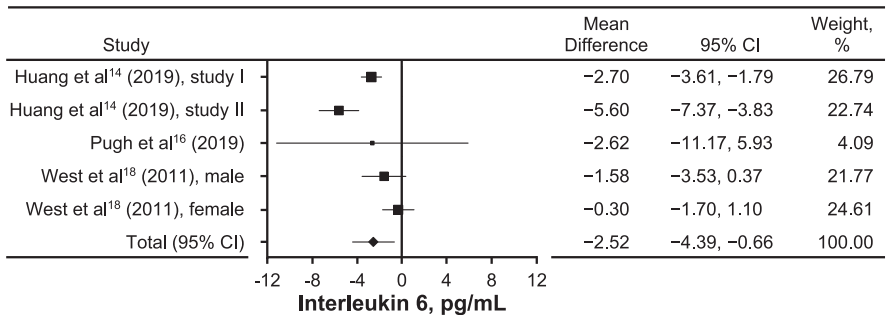

Figure 4. The effect of probiotic supplementation on interleukin-6 (pg/mL).

Table 3. Effect of Probiotic Supplementation on Cytokine Levels

Variable	Meta-Analysis, Mean Difference (95% CI)	Value, %		*P* Value	No. of Studies
		I^2	*Q*		
Tumor necrosis factor α, pg/mL	−2.31 (−4.12, −0.51)	91.92	49.49	.01	3
Interleukin-6, pg/mL	−2.52 (−4.39, −0.66)	81.99	22.21	.002	3
Interleukin-10, pg/mL	2.08 (−0.37, 4.52)	94.57	92.06	.10	4
Immunoglobulin A, mg/L	9.34 (−14.59, 33.28)	1.1	5.056	.44	4
Immunoglobulin A, μg/min	−0.6 (−13.15, 11.95)	0	0.356	.93	3

We observed similar results in our meta-analysis, that elite athletes had lower levels of TNF-α and IL-6 after probiotic supplementation; yet unlike Milajerdi et al,[40] we noted no changes in IL-10 concentration.

The beneficial effects of probiotic supplementation on IL-6 and TNF-α levels are particularly important for athletes, as an increase in IL-6 secretions is one of the causes of inflammation, fatigue, pain, mood changes, and concentration disorders.[31,42,43] Czarkowska-Paczek et al[44] demonstrated that physical exercise induced muscle damage and a complex cascade of nonspecific inflammatory responses. Interleukin-6 mRNA is detectable in skeletal muscle after prolonged, intense exercise, indicating that IL-6 is produced locally in the skeletal muscle.[44] The increases in plasma TNF-α concentration postexercise also suggest a source in damaged muscles.[35] The increased cytokine level may be associated with not only a decrease in the athlete's sport performance but also changes in immune health and an increased incidence of URTIs.[31] Probiotics can help regulate inflammation in a number of ways, improving the structure and function of intestinal epithelial barriers.[41] Probiotics and some of their secreted metabolic products can act as ligands for innate immune system receptors, directly affecting key proinflammatory pathways.[41] Probiotic single-strain and multispecies probiotic supplementation activated the T and B lymphocytes and subsequently increased the production of important regulatory cytokines, including IL-10.[31,45,46]

Our analysis also suggested that probiotic supplementation positively affected the total symptom severity score. The multistrain probiotic formulation could increase the chance of adhesion and colonization of the host by the probiotic strain. If the strains are compatible, they confer synergetic effects. However, in this case, the positive effect of probiotic supplementation on the total symptom severity score was primarily affected by single-strain supplements. We demonstrated no differences in the days of illness, the number of URTI episodes, or the duration of URTI symptoms.

To the best of our knowledge, this is the first meta-analysis to summarize the effects of oral probiotic supplementation on respiratory tract infection and inflammatory cytokines in elite athletes. Nazari et al[41] attempted to assess the effects of probiotic consumption on inflammatory markers, but their research also included sedentary participants and nonprofessional athletes.[47,48] The strengths of our meta-analysis include the comprehensive literature search, the specified inclusion and exclusion criteria for the studies, the explicit methods of data extraction, the measures taken to reduce the influence of bias, and the assessment of heterogeneity.

Our work had certain limitations. First, as mentioned, few publications could be included in the meta-analysis. Second, there were many variations among the studies, including methods, duration of intervention, sport disciplines, type of training and follow-up, probiotic formulation, dose and duration of the intervention, formulation of probiotics, and functions of the strains. Some investigators gave supplements in the form of probiotic capsules, whereas others used beverages or probiotic sachets. Meybodi and Mortazavian[49] suggested that supplements were able to transfer large numbers of viable probiotics into the gastrodigestive tract without much loss during storage. In contrast, the viable number of probiotics in food products, especially fermented items, can considerably decrease after consumption, but foods provide their matrix protection to probiotics. Unfortunately, the best type of probiotic supplement is not yet known.

CONCLUSIONS

This meta-analysis has provided evidence that probiotic supplementation, especially among professional athletes, is an effective way to decrease the total symptom severity score of URTI, especially when this supplementation is taken as a single-strain probiotic. In addition, probiotic

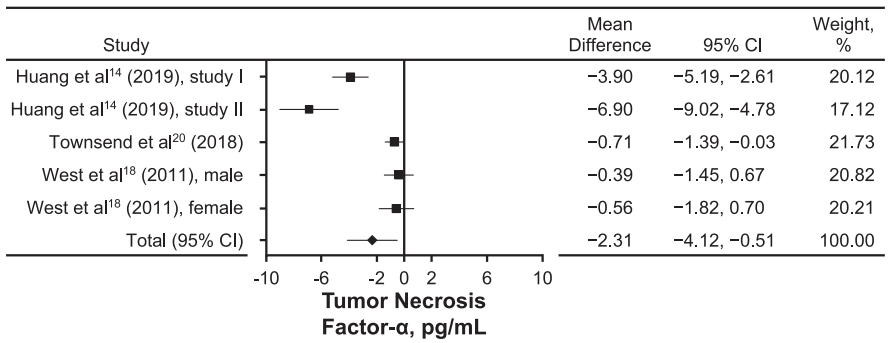

Figure 5. The effect of probiotic supplementation on tumor necrosis factor-α(pg/mL).

supplementation may decrease TNF-α and IL-6 levels. More studies with larger groups are needed to better estimate this effect. Moreover, the optimal timing, duration, composition, and dose of such supplementation need to be estimated.

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SUPPLEMENTAL MATERIAL

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