Immunomodulatory Effects of Probiotic Supplementation in Schizophrenia Patients: A Randomized, Placebo-Controlled Trial

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ABSTRACT: Although peripheral immune system abnormalities have been linked to schizophrenia pathophysiology, standard antipsychotic drugs show limited immunological effects. Thus, more effective treatment approaches are required. Probiotics are microorganisms that modulate the immune response of the host and, therefore, may be beneficial to schizophrenia patients. The aim of this study was to examine the possible immunomodulatory effects of probiotic supplementation in chronic schizophrenia patients. The concentrations of 47 immune-related serum proteins were measured using multiplexed immunoassays in samples collected from patients before and after 14 weeks of adjuvant treatment with probiotics (*Lactobacillus rhamnosus* strain GG and *Bifidobacterium animalis* subsp. *lactis* strain Bb12; $n = 31$) or placebo ($n = 27$). Probiotic add-on treatment significantly reduced levels of von Willebrand factor (vWF) and increased levels of monocyte chemotactic protein-1 (MCP-1), brain-derived neurotrophic factor (BDNF), RANTES, and macrophage inflammatory protein-1 beta (MIP-1) beta with borderline significance ($P \leq 0.08$). *In silico* pathway analysis revealed that probiotic-induced alterations are related to regulation of immune and intestinal epithelial cells through the IL-17 family of cytokines. We hypothesize that supplementation of probiotics to schizophrenia patients may improve control of gastrointestinal leakage.

KEYWORDS: schizophrenia, probiotic, add-on treatment, von Willebrand factor

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Introduction

Abnormal immune responses have been found in many individuals with schizophrenia, regardless of disease stage or medication status.[1] This is also linked to the hypothesis that schizophrenia can originate from early exposure to microbial infections, which may contribute to the etiology through chronic neuroinflammatory and autoimmune processes.[2,3] However, current antipsychotic medications show limited immunomodulatory effects.[4] Recent clinical trials have attempted to target schizophrenia-related immune activation using anti-inflammatory agents. Supplementation with celecoxib, acetylsalicylic acid, and minocycline in addition to standard antipsychotic medication has resulted in overall patient improvement, in particular in a reduction in positive psychotic symptoms.[5–7] However, the number of the immunomodulatory compounds tested to improve symptoms in schizophrenia is small, and further studies are required in this field.

Probiotics are beneficial microorganisms that modulate the immune response of the host by affecting the composition of gut microbiota.[8] Several probiotic species have been tested for health benefits, including the gram-positive anaerobic genres *Lactobacillus* and *Bifidobacterium*. These have shown beneficial effects on systemic inflammatory cytokine levels,[9] neurotransmitter and neurotrophic factor production,[10] intestinal permeability,[11] and oxidative stress in animal models.[12] In humans, oral administration of probiotics is known to restore normal inflammatory status,[13] increase systemic antioxidant capacity,[14] change the activity of brain regions responsible for processing of emotion and sensation,[15] and reduce anxiety.[16] Improved brain function after probiotic supplementation has been attributed to the gut–brain axis, ie, multiple ways of communication between bacteria inhabiting the human intestine and the central nervous system.[17,18] For example, probiotic microorganisms interact with the innate immune system, affecting secretion of pro- and anti-inflammatory

cytokines which, in turn, can regulate brain development and function.[19] In addition, bacteria of the species *Lactobacillus* and *Bifidobacterium* are capable of producing neurotransmitters such as gamma-aminobutyric acid (GABA) and acetylcholine, which directly target receptors in the central nervous system.[20] Therefore, probiotics have been suggested as a potential novel therapeutic approach for a range of neurodevelopmental disorders.[21]

We recently carried out a clinical trial to assess whether supplementation of probiotic strains *Lactobacillus rhamnosus* GG and *Bifidobacterium lactis* Bb12 can reduce symptom severity in schizophrenia patients remaining on long-term antipsychotic treatment.[22] The present follow-up study was undertaken to examine the systemic immunomodulatory effects of probiotic supplementation in the same patient population. Using multiplexed immunoassays, we measured the levels of 47 immune molecules in patient sera collected before and after treatment with adjunctive probiotics or placebo. Group comparisons revealed probiotic-specific changes in levels of molecules involved in innate and adaptive immune responses and intestinal epithelial cell function. These alterations may be related to improved function of the intestinal tract in the probiotic arm of the trial reported before.[22]

Materials and Methods

Participants and study procedures. The patient population and probiotic compound investigated in this study have been described in detail previously.[22] Briefly, 65 outpatients from psychiatric rehabilitation programs in the Baltimore area (MD, USA) diagnosed with schizophrenia or schizoaffective disorder according to DSM-IV criteria, with at least moderately severe psychotic symptoms [Positive and Negative Syndrome Scale (PANSS) positive score ≥1, PANSS negative symptom score ≥4, or total PANSS score ≥50, containing at least three positive or negative items with scores ≥3 at screening] were enrolled in the study between December 2010 and August 2012. Participants were randomized into a double-blind 14-week treatment protocol with adjunctive probiotic ($n = 33$) or placebo ($n = 32$), with initial 2-week placebo run-in (Fig. 1). All patients received antipsychotic treatment for at least eight weeks prior to starting the trial and did not change the medication within the previous 21 days. Patients suffering from any clinically significant or unstable medical condition, including congestive heart failure, celiac disease, or immunodeficiency syndromes, as well as those receiving antibiotics within the previous 14 days were excluded from the study.

The active study compound consisted of one tablet containing approximately 10^9 colony forming units of the probiotic organism *L. rhamnosus* GG and 10^9 colony forming units of the probiotic organism *Bifidobacterium animalis* subsp. *lactis* BB12 (Ferrosan) or placebo. The probiotic microorganisms were grown in media that do not contain casein, lactose, other milk products, or gluten, to reduce the risk of allergic reactions to these ingredients.

In total, 58 participants completed the trial, comprising 31 in the probiotic arm and 27 in the placebo arm. Blood samples were collected from all subjects at the beginning and at the end of the trial. Serum was prepared by allowing clot

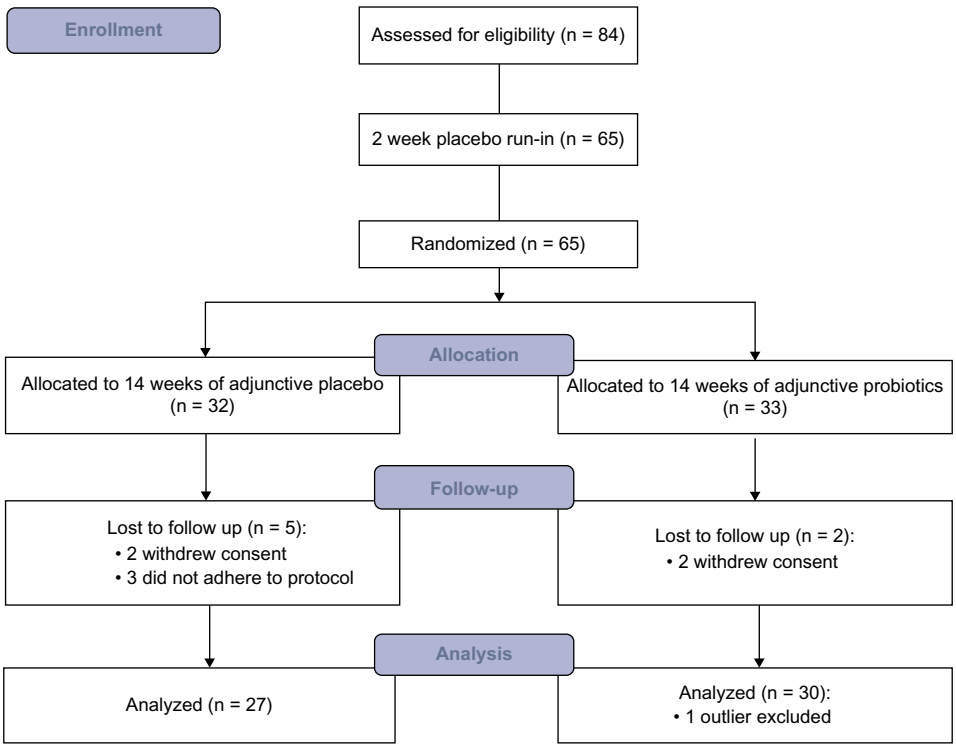

Figure 1. CONSORT (Consolidated Standards of Reporting Trials) flow diagram of the trial.

formation for two hours at room temperature and subsequent centrifugation at 4000*g* for five minutes. The resulting serum supernatants were stored at -80 °C until analysis.

Multiplexed immunoassays. Serum samples were analyzed using the Human InflammationMAP panel in a Clinical Laboratory Improvement Amendments (CLIA)-certified laboratory (Myriad RBM). The panel consisted of 47 multiplexed immunoassays targeting selected inflammatory markers, including cytokines, chemokines, and acute-phase reactants (Table 1A). Analyte levels were estimated in each sample from the 8-point standard curves, and assay performance was validated using three control samples. The same multiplex immunoassay platform has been applied successfully for serum biomarker profiling in a range of high-impact studies.[23–25]

Data analysis. For those participants who completed the double-blind phase, immune marker data acquired from multiplex immunoassay analyses were filtered separately for each treatment group and time point. Principal component analysis was applied to identify artifactual effects on the overall variance. One extreme outlier was detected outside of the Hotelling's T^2 ellipse showing 95% confidence intervals[26] in the probiotic-supplemented group and was removed from the analysis. Molecules with more than 60% low values were excluded from further analysis to allow a minimum of 10 measurements per comparison group. This equated to 20 analytes (Table 1A). For the 27 analytes remaining in the dataset, missing values were replaced with half the minimum value for that specific assay. Shapiro–Wilk tests showed that the majority of analyte levels were non-normally distributed and a large proportion (30%) remained non-normally distributed after $\log_{10}$-transformation. Therefore, a non-parametric Wilcoxon signed-rank test was applied to compare analyte levels before and after treatment. Resulting *P*-values were controlled for false discovery rate with a conservative Benjamini–Hochberg approach.[27]

Pathway analysis was performed using the ingenuity pathways knowledge database (IPKB; Ingenuity® Systems). Only molecules from the datasets that met the *P*-value cutoff of 0.10 and were associated with the biological functions and/or canonical pathways in the IPKB were considered for the analysis. A right-tailed Fisher's exact test was used to calculate *P*-values associated with the identified pathways. The significance of the association between the dataset and canonical pathways was measured by the ratio between the number of molecules from the dataset divided by the total number of known molecules in that pathway and by the *P*-value (Fisher's exact test).

Ethical considerations. The study was approved by Institutional Review Board of the Sheppard Pratt Health System and the Johns Hopkins School of Medicine. The trial was registered at ClinicalTrials.gov corresponding to NCT01242371 and monitored by a data safety monitoring board. Written informed consent was obtained from all study participants. The research complied with the principles of the Declaration of Helsinki.

Results

A total of 65 patients were enrolled in the study and randomized, 33 to the adjunctive probiotics arm and 32 to the adjunctive placebo arm. A total of 58 participants (89%) completed the study (Fig. 1). The clinical characteristics of the completers are shown in Table 1. There were no significant differences in age, gender, race, duration of education, PANSS scores, and proportion of patients receiving clozapine between the groups at the beginning of the study.[22] PANSS psychiatric symptom scores did not change over the course of the trial, but patients receiving probiotic supplement were less likely to report severe bowel difficulties ($P = 0.003$).[22]

In terms of the mechanism of action, treatment with probiotics significantly decreased the levels of the acute-phase reactant von Willebrand factor (vWF; Table 2). Uploading the accession numbers of all analytes, which showed a change at a significance level of <0.10 [vWF, monocyte chemotactic protein-1 (MCP-1), brain-derived neurotrophic factor (BDNF), T-cell-specific protein RANTES, and macrophage inflammatory protein-1 beta (MIP-1 beta)] into the IPKB (www.ingenuity.com), indicated that supplementation with probiotics most significantly affected the regulation of cytokine production in macrophages, T helper cells and intestinal epithelial cells by IL-17A and IL-17F pathways (top canonical pathways, $P = 9.34E-06$ and $1.54E-05$, ratio 0.111 and 0.087; Table 3). Missing values for the proteins included in pathway analysis ranged between 0 and 31%.

Changes identified in the placebo group [vascular cell adhesion molecule 1 (VCAM-1) and intercellular adhesion molecule 1 (ICAM-1); Table 2] may have resulted from abnormally high levels of these proteins at baseline and therefore cannot be assigned as true placebo effects. Initial levels of VCAM-1 in the placebo group were 21% higher than in the group supplemented with probiotics ($P = 0.021$, Mann–Whitney test). After treatment, VCAM-1 returned to similar levels (3% difference, $P = 0.581$), with the greatest change

Table 1. Demographical and clinical data of study completers. The table shows mean values ± standard deviation. Detailed description of this patient population can be found in Ref. 22.

	PROBIOTIC SUPPLEMENT	PLACEBO SUPPLEMENT	*P*-VALUE
N	31	27	–
Age	44.8 ± 11.2	48.1 ± 9.4	0.236[a]
Gender (male/female)	22/9	16/11	0.413[b]
Race (white/other)	16/15	20/7	0.106[b]
PANSS[c] total start	67.3 ± 11.9	70.2 ± 11.6	0.258[a]
PANSS[c] total end	66.8 ± 11.6	67.3 ± 11.9	0.773[a]
CRP[d] start (µg/ml)	6.7 ± 8.2[e]	6.3 ± 7.6	0.829[a]
CRP[d] end (µg/ml)	7.1 ± 7.9	7.3 ± 10.1[e]	0.841[a]

Notes: [a]Mann–Whitney *U* test. [b]Fisher's exact test. [c]Positive and Negative Syndrome Scale. [d]C-reactive protein. [e]One outlier excluded.

Table 2. Changes in immune marker levels following probiotic and placebo supplementation with *P*-value <0.10. The table shows mean values before and after treatment ± standard deviation. The full list of changes is available in Table 2A.

ANALYTE	UNIPROTKD ID	UNITS	PROBIOTIC SUPPLEMENT				PLACEBO SUPPLEMENT			
			BEFORE	AFTER	*P*-VALUE	Q-VALUE[a)]	BEFORE	AFTER	*P*-VALUE	Q-VALUE[a)]
von Willebrand Factor	P04275	µg/ml	30.5 ± 21.0	25.5 ± 18.4	0.047	0.431				
Monocyte Chemotactic Protein 1	P13500	pg/ml	184.9 ± 125.7	226.1 ± 135.5	0.054	0.431				
Brain Derived Neurotrophic Factor	P23560	ng/ml	3.2 ± 1.5	4.1 ± 2.6	0.063	0.431				
T Cell Specific Protein RANTES	P13501	ng/ml	5.7 ± 2.6	7.3 ± 4.7	0.069	0.431				
Macrophage Inflammatory Protein 1 beta	P13236	pg/ml	185.9 ± 110.9	205.0 ± 109.9	0.080	0.431				
Vascular Cell Adhesion Molecule 1	P19320	ng/ml					505.0 ± 160.8	442.1 ± 134.4	0.016	0.313
Intercellular Adhesion Molecule 1	P05362	ng/ml					200.1 ± 108.4	166.9 ± 66.6	0.023	0.313
Tumor necrosis factor receptor 2	P20333	ng/ml					5.8 ± 2.9	5.0 ± 1.9	0.072	0.450
Ferritin	P02794 P02792	ng/ml					142.1 ± 106.2	121.3 ± 85.7	0.073	0.450
Matrix Metalloproteinase 3	P08254	ng/ml					19.0 ± 12.9	16.8 ± 11.0	0.099	0.450

Note: [a]q-value – *P*-value controlled for false discovery rate with Benjamini–Hochberg procedure.

Table 3. A list of canonical pathways most significant to the probiotic and placebo groups revealed by Ingenuity Pathway Analysis. Only proteins that met the *P*-value cut-off of 0.10 were considered for the analysis. Significance of the association was measured by *P*-value and by the ratio of the involved molecules. Probiotic supplementation showed enrichment in IL-17A- and IL-17F-related pathways.

PROBIOTIC SUPPLEMENT	PLACEBO SUPPLEMENT
Differential Regulation of Cytokine Production in Macrophages and T Helper Cells by IL-17A and IL-17F (*P* = 9.34E-06, ratio = 1.11E-01)	Atherosclerosis Signaling (*P* = 2.54E-08, ratio = 3.1E-02)
Differential Regulation of Cytokine Production in Intestinal Epithelial Cells by IL-17A and IL-17F (*P* = 1.54E-05, ratio = 8.7E-02)	Leukocyte Extravasation Signaling (*P* = 2.37E-05, ratio = 1.6E-02)
Role of Hypercytokinemia/ hyperchemokinemia in the Pathogenesis of Influenza (*P* = 4.99E-05, ratio = 4.9E-02)	Role of Macrophages, Fibroblasts and Endothelial Cells in Rheumatoid Arthritis (*P* = 8.33E-05, ratio = 1.0E-02)
Role of IL-17 A in Arthritis (*P* = 8.7E-05, ratio = 3.3E-02)	HMGB1 Signaling (*P* = 4.3E-04, ratio = 2.1E-02)
Role of MAPK Signaling in the Pathogenesis of Influenza (*P* = 1.3E-04, ratio = 3.0E-02)	Hepatic Fibrosis/Hepatic Stellate Cell Activation (*P* = 9.31E-04, ratio = 1.4E-02)

observed for patients with the highest initial VCAM-1 levels (Spearman's rho = −0.61, $P = 0.0007$). This suggests that changes in the placebo group may be because of the regression to the mean effect.[28,29] In contrast, the levels of vWF at the end of the trial were 11% lower in the probiotic arm, suggesting a true change in vWF levels by probiotics.

Discussion

This is the first study to investigate the immunomodulatory effects of probiotic supplementation in schizophrenia. In this investigation, 65 patients undergoing long-term antipsychotic treatment were randomized to 14 weeks of either probiotic or placebo add-on supplementation and 58 participants completed the trial (Fig. 1). Using multiplexed immunoassays, we measured levels of 47 immune markers before and after add-on treatment, of which 27 satisfied the strict criteria for analysis. We found that probiotic supplementation significantly reduced levels of vWF, and levels of MCP-1, BDNF, T-cell-specific protein RANTES, and MIP-1 beta were found to be increased with borderline significance ($P \leq 0.08$). These changes were related mostly to the regulation of cytokine production in macrophages, T helper cells, and intestinal epithelial cells as shown by the effects on IL-17A and IL-17F pathways. Although IL-17 levels were not detectable in our study, we identified a trend toward increased levels of other cytokines related to IL-17, namely MCP-1 and RANTES. The decreased levels of VCAM-1 and ICAM-1 in the placebo group were interpreted as regression to the mean effect,[28,29] ie, they resulted from abnormally high initial levels in the placebo

group and did not differ between the groups at the end of the study. This is consistent with the fact that the placebo was provided in the context of long-term stable antipsychotic treatment, and therefore, no changes were expected in this group.

The only molecule that changed when applying conventional significance criteria ($P < 0.05$) after probiotic supplementation was vWF (FC = 0.84). vWF is a positive acute-phase reactant produced by endothelial cells in response to injury and plays an important role in blood coagulation. This protein has been found to be positively correlated with cardiovascular risk factors in schizophrenia and bipolar disorder, and does not change with second-generation antipsychotic treatment.[30] In our study, probiotic supplementation decreased the levels of vWF. Probiotics are known to decrease levels of certain cardiovascular risk parameters.[31] However, their effect on cardiovascular risk and antipsychotic-related thromboembolism[32] should be assessed in future longitudinal studies by measuring more specific cardiovascular and coagulation markers, such as D-dimer, triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL).

Pathway analysis suggested that probiotic add-on treatment, but not standard antipsychotic therapy, modulates immune function via type 17 immune responses. This pathway involves the IL-17 family of cytokines, which are regulatory proteins produced by T helper 17 (Th17) cells involved in cellular responses to extracellular bacteria such as those colonizing the intestinal lumen.[33] Therefore, we hypothesize that the observed cytokine changes are related to improved bowel functioning, which we reported in this group of patients previously.[22] These cytokines are also critical mediators of autoimmune reactions. Deregulation of the type 17 response has been observed in schizophrenia. Studies have shown that this pathway was blunted in psychotic episodes.[34] Also, autoimmune processes against central nervous tissue components, which are regulated by type 17 cytokines, are a known phenomenon that may contribute to schizophrenia etiology and/or pathology.[35] Here, we showed that molecules associated with the type 17 response, in particular MCP-1 and RANTES, increased with borderline significance in the group of patients treated with probiotic supplement. Pathway analysis suggested that these changes may be associated with improved control of gastrointestinal leakage mediated through the innate and adaptive immune systems. This is consistent with the observed decrease in vWF levels after probiotic supplementation, which might be a secondary effect of improved intestinal epithelium integrity. IL-17 levels were not detectable in any of the samples (<5 pg/mL). Therefore, further studies using assays with improved sensitivity are necessary to identify any direct effects on T helper type 17 pathways.

The finding that BDNF was increased with borderline significance by probiotic add-on treatment is interesting as it is a neurotrophin involved in neuronal survival and plasticity and has been associated previously with schizophrenia pathophysiology.[36] However, an increase in BDNF levels did not translate into improved symptoms in the probiotic arm and requires further validation to show statistical significance.

Several limitations of the molecular profiling results should be considered when interpreting the results of this study. First, we were not able to detect all targeted cytokines in our clinical samples. This included molecules that have previously been shown to be altered in schizophrenia and modulated by probiotics, such as IL-1 beta, IL-6, TNF-alpha, and IFN-gamma. Owing to relatively high P- and q-values ($q = 0.431$ for $P < 0.1$ in the probiotic arm), the identified changes require further validation studies. Repeating these experiments using more sensitive and diverse multiplex immunoassays is essential to provide a more complete picture of the effects. Second, although we investigated only patients who remained on stable, long-term antipsychotic treatment during the trial period, it is still possible that the antipsychotic compound exhibited certain immunomodulatory effects. This relates in particular to the changes identified in VCAM-1 and ICAM-1 levels in the placebo group. Furthermore, because of the lack of a control group, it was not possible to determine whether baseline levels of the analytes were altered. In addition, the small number of analytes identified as significantly or borderline different between the two groups was a limiting factor for the pathway analysis, and more complex immune networks may be affected by probiotic supplementation. For example, although pathway analysis revealed that probiotic supplementation may affect the function of intestinal cells, further studies with more specific markers are required to assess whether probiotics can attenuate gastrointestinal inflammation.

We conclude that probiotics have immunomodulatory effects in schizophrenia patients, affecting molecules that do not respond to standard antipsychotic therapy. These changes may be associated with the improvement in bowel functioning reported previously in the same group of patients through IL-17-related immune responses, which control the intestinal microbiome–host interaction. However, supplementation of probiotics does not reduce psychotic symptoms. We suggest that future studies should be carried out that test the exact biological and neurobiological mechanisms of probiotic supplementation.

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Author Contributions

Conceived and designed the experiments: RHY, FBD. Analyzed the data: JT. Wrote the first draft of the manuscript: JT, FBD. Contributed to the writing of the manuscript, jointly developed the structure and arguments for the paper, made critical revisions and agree with the manuscript results and conclusions: JT, RHY, SB, FBD. All authors reviewed and approved of the final manuscript.

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Appendix

Table 1A. Inflammatory markers measured using Human InflammationMAP multiplexed immunoassay platform. Analytes excluded from the analysis because of high proportion of missing values are shown in gray.

PROTEIN	UNIPROTKB ID	PROTEIN	UNIPROTKB ID
Alpha-1-Antitrypsin (AAT)	P01009	Interleukin-10 (IL-10)	P22301
Alpha-2-Macroglobulin (A2Macro)	P01023	Interleukin-12 Subunit p40 (IL-12p40)	P29460
Beta-2-Microglobulin (B2M)	P61769	Interleukin-12 Subunit p70 (IL-12p70)	P29459
Brain-Derived Neurotrophic Factor (BDNF)	P23560	Interleukin-15 (IL-15)	P40933
C-Reactive Protein (CRP)	P02741	Interleukin-17 (IL-17)	Q16552
Complement C3 (C3)	P01024	Interleukin-18 (IL-18)	Q14116
Eotaxin-1	P51671	Interleukin-23 (IL-23)	Q9NPF7
Factor VII	P08709	Macrophage Inflammatory Protein-1 alpha (MIP-1 alpha)	P10147
Ferritin (FRTN)	P02794, P02792	Macrophage Inflammatory Protein-1 beta (MIP-1 beta)	P13236
Fibrinogen	P02671, P02675, P02679	Matrix Metalloproteinase-2 (MMP-2)	P08253
Granulocyte-Macrophage Colony-Stimulating Factor (GM-CSF)	P04141	Matrix Metalloproteinase-3 (MMP-3)	P08254
Haptoglobin	P00738	Matrix Metalloproteinase-9 (MMP-9)	P14780
Intercellular Adhesion Molecule 1 (ICAM-1)	P05362	Monocyte Chemotactic Protein 1 (MCP-1)	P13500
Interferon gamma (IFN-gamma)	P01579	Stem Cell Factor (SCF)	P21583
Interleukin-1 alpha (IL-1 alpha)	P01583	T-Cell-Specific Protein RANTES (RANTES)	P13501
Interleukin-1 beta (IL-1 beta)	P01584	Tissue Inhibitor of Metalloproteinases 1 (TIMP-1)	P01033
Interleukin-1 receptor antagonist (IL-1ra)	P18510	Tumor Necrosis Factor alpha (TNF-alpha)	P01375
Interleukin-2 (IL-2)	P60568	Tumor Necrosis Factor beta (TNF-beta)	P01374
Interleukin-3 (IL-3)	P08700	Tumor necrosis factor receptor 2 (TNFR2)	P20333
Interleukin-4 (IL-4)	P05112	Vascular Cell Adhesion Molecule-1 (VCAM-1)	P19320
Interleukin-5 (IL-5)	P05113	Vascular Endothelial Growth Factor (VEGF)	P15692
Interleukin-6 (IL-6)	P05231	Vitamin D-Binding Protein (VDBP)	P02774
Interleukin-7 (IL-7)	P13232	von Willebrand Factor (vWF)	P04275
Interleukin-8 (IL-8)	P10145		

Table 2A. Changes in immune marker levels following probiotic and placebo supplementation. FC – average fold change; *P*-value <0.05 was considered significant (in bold); *q*-value – *P*-value controlled for false discovery rate with Benjamini–Hochberg procedure.

ANALYTE	FUNCTION[a)]	PROBIOTIC SUPPLEMENT				PLACEBO SUPPLEMENT			
			FC	*P* VALUE	*q*-VALUE		FC	*P*-VALUE	*q*-VALUE
von Willebrand Factor	AP	↓	**0.84**	**0.047**	**0.431**	↑	1.15	0.751	0.881
Monocyte Chemotactic Protein 1	C	↑	1.22	0.054	0.431	↓	0.93	0.525	0.782
Brain Derived Neurotrophic Factor	GF	↑	1.28	0.063	0.431	↑	1.06	0.895	0.966
T Cell Specific Protein RANTES	C	↑	1.28	0.069	0.431	↓	0.96	0.551	0.782
Macrophage Inflammatory Protein 1 beta	C	↑	1.1	0.080	0.431	↓	0.97	0.62	0.821
Factor VII	O	↑	1.05	0.221	0.759	↓	0.92	0.117	0.450
Ferritin	AP	↓	0.91	0.225	0.759	↓	0.85	0.073	0.450
Vascular Endothelial Growth Factor	GF	↑	1.08	0.225	0.759	↑	1.01	0.957	0.994
Stem Cell Factor	GF	↑	1.06	0.285	0.855	↓	0.91	0.243	0.513
Haptoglobin	AP	↓	0.93	0.365	0.876	↑	1.04	0.4	0.720

(*Continued*)

Table 2A. (*Continued*)

ANALYTE	FUNCTION[a)]	PROBIOTIC SUPPLEMENT				PLACEBO SUPPLEMENT			
			FC	*P*-VALUE	*q*-VALUE		FC	*P*-VALUE	*q*-VALUE
Fibrinogen	AP	↓	0.95	0.375	0.876	↑	1.02	0.511	0.782
Complement C3	AP	↓	0.96	0.421	0.876	↓	0.93	0.247	0.513
Interleukin 8	C	↓	0.95	0.449	0.876	↑	1.03	0.713	0.875
Interleukin 1 receptor antagonist	C	↑	1.04	0.559	0.876	↓	0.83	0.104	0.450
Alpha 2 Macroglobulin	AP	↑	1.02	0.581	0.876	↑	1.02	1.000	1.000
Vascular Cell Adhesion Molecule 1	O	↑	1.02	0.584	0.876	**↓**	**0.88**	**0.016**	**0.313**
Beta 2 Microglobulin	TR	↓	0.99	0.589	0.876	↓	0.93	0.139	0.461
Alpha 1 Antitrypsin	AP	↑	1.03	0.613	0.876	↓	0.98	0.638	0.821
Interleukin 18	C	↑	1.03	0.65	0.876	↓	0.9	0.343	0.661
Vitamin D Binding Protein	O	↑	1.02	0.713	0.876	↓	0.95	0.234	0.513
Interleukin 23	C	↑	1.02	0.727	0.876	↓	0.88	0.154	0.461
Eotaxin 1	C	↑	1.02	0.757	0.876	↓	0.96	0.431	0.727
Tumor necrosis factor receptor 2	TR	↑	1.01	0.758	0.876	↓	0.87	0.072	0.450
Intercellular Adhesion Molecule 1	TR	↑	1.03	0.805	0.876	**↓**	**0.83**	**0.023**	**0.313**
Matrix Metalloproteinase 3	O	↓	0.98	0.811	0.876	↓	0.88	0.099	0.450
Tissue Inhibitor of Metalloproteinases 1	O	↑	1.02	0.914	0.949	↓	0.93	0.242	0.513
C Reactive Protein	AP	↓	0.44	0.992	0.992	↑	1.54	0.882	0.966

[a]**Abbreviations:** AP, acute-phase protein; C, cytokine; GF, growth factor; TR, transmembrane receptor; O, other.