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Probiotics Impact on Cognitive Ability and Emotions in Adolescents: A Brain-Gut Axis Study

Boran Cui[1,a,†], Zhenxiong Wen[1,b,†], Yuhan Zhang[2,c], Weibo Zhang[1,d,*]

[1]*Dalton Academy, The Affiliated High School of Peking University, Beijing, China*
[2]*Prince of Wales Secondary School, Vancouver, Canada*
[a]*cuiboran2025@i.pkuschool.edu.cn,* [b]*wenzhenxiong2024@i.pkuschool.edu.cn,*
[c]*elinor716@hotmail.com,* [d]*zhangweibo@i.pkuschool.edu.cn*
[*]*Corresponding author*
[†]*Equal contribution*

Abstract: *The Gut-Brain Axis (GBA) is a bidirectional communication system between the central nervous system (CNS) and the enteric nervous system (ENS), with gut microbiota playing a crucial role in regulating its functions. This study investigates probiotics' effects on adolescents' cognition, memory, and emotions. A double-blind, randomized controlled trial involved 15 healthy adolescents aged 15–18, divided into a treatment group receiving probiotics and a control group given a placebo. Assessments included baseline and post-intervention evaluations after a four-week period using the Positive and Negative Affect Schedule (PANAS) and the Montreal Cognitive Assessment (MoCA). Results showed significant cognitive improvement and increased positive emotions in the treatment group, while the control group exhibited no notable changes. These findings suggest probiotics positively influence cognitive function and emotional state by modulating gut microbiota. Despite the small sample size, this study provides preliminary evidence for probiotics' therapeutic potential in adolescents. Larger, longer-term studies are needed to confirm these results.*

Keywords: *Gut-Brain Axis, Probiotics, Cognitive Function, Emotional State, Adolescents, Lp-G18*

1. Background

The gut-brain axis (GBA) is a bidirectional communication network between the central nervous system (CNS) and the enteric nervous system (ENS), involving brain regions responsible for emotion and cognitive functions, as well as peripheral mechanisms within the gut [1]. Previous studies have emphasized the crucial role of the gut microbiota in modulating GBA function. This microbial community communicates with the brain via multiple signaling pathways—including neural, endocrine, immune, and humoral routes—forming a complex bidirectional regulatory network [2].

A substantial body of recent research has demonstrated that the GBA significantly impacts neurodegenerative diseases and individual cognitive functions, influencing conditions such as Alzheimer's disease (AD) [3] and Parkinson's disease (PD). A multi-level analysis revealed that molecular and microbial characteristics associated with PD are related to the functional structure of the GBA, and these features correlate with the heterogeneity of PD phenotypes [4]. Moreover, metabolic products of the gut microbiota, such as short-chain fatty acids (SCFAs), have been shown to cross the blood-brain barrier and affect brain function, thereby influencing mood and cognition [5]. These findings provide robust evidence for the role of the gut microbiota in regulating the GBA.

Notably, dietary patterns are considered an important factor in exploring the influence of the gut microbiota on cognitive function. Studies have demonstrated that adherence to the Mediterranean diet is associated with a reduced risk of PD and is linked to a delayed onset of PD in aging populations [6]. Furthermore, specific dietary components—such as polyunsaturated fatty acids, antioxidants, vitamins, and minerals—are associated with PD risk [7]. These findings further support the evidence that the gut microbiota influences cognitive function via the GBA by metabolizing dietary components.

Furthermore, studies have indicated that gut inflammation is related to the pathological processes of PD [8] and is also associated with emotional disorders such as depression [9]. However, studies focusing on adolescent populations remain relatively scarce. Therefore, this study aims to explore the impact of the gut microbiota on the memory abilities of adolescents—a key component of cognitive function—

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highlighting the importance and necessity of this research.

2. Research Design

2.1. Objectives

This study aims to explore the effects of probiotic supplementation on the cognitive abilities, memory, and emotions of adolescents aged 15 to 18, based on the gut-brain axis (GBA) theory. The goal is to provide scientific evidence for interventions targeting adolescent mental health and cognitive development (see *Figure 1*).

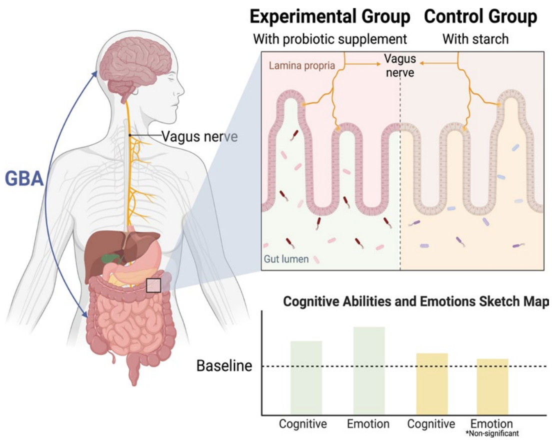

Figure 1: Illustration of GBA and Experimental Design

2.2. Hypothesis

We hypothesize that the treatment group receiving probiotic supplements will exhibit significant improvements in cognitive ability, memory, and emotional state compared to the control group receiving a placebo.

2.3. Subjects

The study planned to recruit 15 healthy adolescent volunteers aged between 15 and 18 years. Subjects were randomly assigned to comprising 8 participants. The control group received a placebo (starch), while the treatment group received probiotic supplements.

2.4. Experimental Design

2.4.1. Pre-Treatment Phase

All participants underwent baseline assessments, including the Positive and Negative Affect Schedule (PANAS) to evaluate emotional states [10] and the Montreal Cognitive Assessment (MoCA) to assess cognitive abilities [11].

2.4.2. Treatment Phase (4 Weeks)

Participants received a four-week treatment according to their group assignment. The control group received a placebo, whereas the treatment group received probiotic supplements.

2.4.3. Post-Treatment Phase (4 Weeks)

After the four-week intervention, participants completed the PANAS and MoCA assessments again to evaluate the effects of the intervention.

2.5. Probiotic Supplements

The probiotic supplement used in this study is a composite preparation of active strains, including the following nationally recognized food-grade strains: Lp-G18 (*Lactobacillus plantarum*), Lr-G14 (*Lactobacillus rhamnosus*), CP-9 (*Bifidobacterium animalis*), LR-G100 (*Lactobacillus reuteri*), LF-G89

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(*Lactobacillus fermentum*), LA-G80 (*Lactobacillus acidophilus*), LPc- G110 (*Lactobacillus paracasei*), LG-G12 (*Lactobacillus gasseri*), LC-G11 (*Lactobacillus casei*), HN019 (*Bifidobacterium lactis*), HN001 (*Lactobacillus rhamnosus*), BB-G90 (*Bifidobacterium bifidum*), BL-G301 (*Bifidobacterium longum* subsp.), BB-G95 (*Bifidobacterium breve*), LJ-G55 (*Lactobacillus johnsonii*), LS-G60 (*Lactobacillus salivarius*), ST-G30 (*Streptococcus thermophilus*), LB-G40 (*Lactobacillus delbrueckii* subsp. *bulgaricus*), BI-G201 (*Bifidobacterium longum* subsp. *infantis*), and BL-G101 (*Bifidobacterium animalis* subsp. *lactis*).

During the experiment, the probiotic supplement was provided to participants in the treatment group in standardized dosage forms to ensure consistency and reproducibility. The control group received an equal amount of placebo to eliminate non-specific effects on the study results.

2.6. Probiotic Supplements

- **Positive and Negative Affect Schedule (PANAS):** A self-report questionnaire used to assess participants' positive and negative emotions;
- **Montreal Cognitive Assessment (MoCA):** A cognitive test designed to evaluate various domains such as memory, attention, language, and executive functions.

2.7. Data Analysis

Statistical software will be used for data analysis, including but not limited to repeated measures ANOVA and independent samples T-tests, to determine whether there are significant differences before and after treatment in PANAS and MoCA assessments between the treatment and control groups.

2.8. Expected Results

We anticipate that the treatment group receiving probiotic supplements will show significant improvements in cognitive abilities, memory, and emotional states on the PANAS and MoCA assessments compared to the control group receiving a placebo. These expected results will provide empirical support for the role of probiotics in regulating cognitive and emotional health through the GBA and offer scientific evidence for the potential therapeutic application of probiotics in adolescent populations.

2.9. Research Methods

This study employs a double-blind experimental design, where both participants and researchers are unaware of group assignments (probiotic supplements or placebo).

3. Results

In the analysis of pre- and post-test results from the Montreal Cognitive Assessment (MoCA) and the Positive and Negative Affect Schedule (PANAS) conducted on 15 participants, we observed some significant trends.

3.1. MoCA Data

The MoCA Data paired t-test results for the control group and experimental group are shown below [12] in *Figure 2*.

Through paired sample t-tests, we found a statistically significant difference in the MoCA scores of the control group before and after the experiment. Specifically, the control group's MoCA scores slightly increased from an initial average of 25.50 (± 1.51) to an average of 26.50 (± 1.41) at the end of the experiment. This change is marginally significant at the 0.05 level (t = -2.366, p = 0.050). This result suggests that the improvement in cognitive abilities in the control group may be due to the familiarity effect caused by repeated MoCA testing, but this effect is not pronounced.

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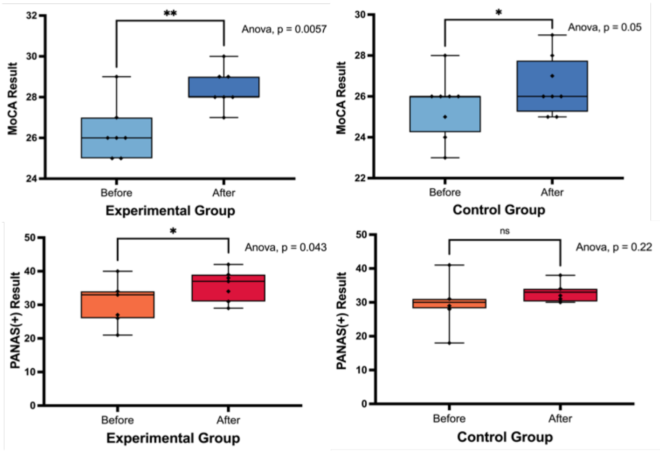

Figure 2: Box Plot Analysis of Cognitive and Emotional Test Results

For the experimental group receiving probiotic supplements, the paired sample t-test revealed a highly significantly improvement in cognitive abilities. The MoCA scores of the experimental group significantly increased from an initial average of 26.29 (± 1.38) to an average of 28.43 (± 0.98) at the end of the experiment. This improvement is statistically significant at the 0.01 level (t = -3.873, p = 0.008). Compared to the control group, the experimental group showed a higher level of significance, indicating that probiotic intake may have a significant positive effect on the cognitive abilities of adolescents. This finding is consistent with the current scientific consensus on the interaction between probiotics and the gut-brain axis in other age groups. We speculate that the increase in MoCA scores for participants in both the control and experimental groups after repeated testing may be due to familiarity with the test content. However, the experimental group outperformed the control group in both statistical significance and the magnitude of score improvement. Therefore, we believe this phenomenon strongly suggests that the intake of probiotics has a significant positive impact on the cognitive abilities of adolescents aged 15 to 18.

3.2. PANAS Data

The pre- and post-experiment data for all subjects are shown in *Figure 3*, and the PANAS data [12] for the control group and the experimental group are also shown in *Figure 2*.

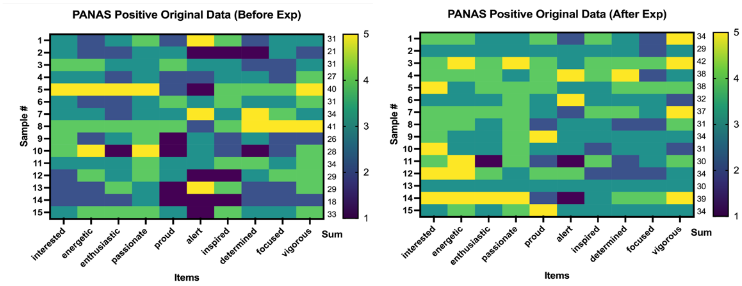

Figure 3: Heat Maps of PANAS Positive Data

In comparison, although the control group showed an increase in PANAS positive affect scores, the change was not statistically significant (t = -1.984, p = 0.088). This may indicate that the emotional state of the control group did not undergo a significant positive change over time.

Experimental data showed that the experimental group had a significant increase in the positive dimension of emotional state from the beginning to the end of the test. Specifically, the positive affect scores increased from an average of 30.71 (± 6.37) to an average of 35.71 (± 4.61). This change is statistically significant (t = -2.562, p = 0.043), indicating the potential effect of probiotic intake on enhancing the positive emotions of adolescents [13].

Integrating the analysis results of the PANAS data, we observe that compared with the control group, the experimental group showed significant improvement in the positive emotion dimension after receiving probiotic supplements. This finding further supports the hypothesis that probiotics may have a positive impact on emotional health by regulating the gut microbiota. Moreover, since the participants in the experimental and control groups were randomly assigned, we can reasonably infer that the impact of recent individual events on emotions should be randomly distributed between the two groups and is unlikely to systematically bias one group. Additionally, although the sample size of 8 participants per group is small, it is sufficient to detect differences in emotional changes between the two groups, especially when the experimental group shows significant emotional improvement, and the control group does not. Therefore, we can reasonably exclude the possibility that individual recent events affecting emotions are the cause of inaccurate experimental results. The significant improvement observed in the experimental group is more likely due to the effect of probiotic intervention rather than the influence of random events or non-specific factors. Future research can further validate these preliminary findings by increasing the sample size and implementing longer follow-up periods.

4. Overview of Research and Bioinformatics Analysis of Lp-G18

4.1. Research Background of Lp-G18

In studies exploring the effects of probiotics on adolescents' cognitive abilities and emotions, the strain Lp-G18 (*Lactobacillus plantarum*) occupies a significant proportion in our composite probiotic supplement; therefore, we focus on its research. Based on existing scientific literature and our research results, we have conducted a bioinformatics-based summary and analysis of this strain. We found that Lp-G18 exhibits significant effects in regulating the immune system, improving gut health, and combating pathogenic microorganisms [14-15]. Additionally, Lp-G18 shows great potential in treating skin infections and in ophthalmology [16-17]. The following discussion will focus on its impacts on the gut.

Lp-G18 can help restore and maintain the balance of the gut microbiota by competitively excluding pathogens, thereby reducing the number of harmful bacteria such as pathogenic *Escherichia coli* and *Staphylococcus aureus*. Existing studies have also pointed out that the competitive exclusion effect impacts the competition between probiotics and pathogens for nutrients and adhesion sites and promotes the production of potent antimicrobial substances that contribute to competitive exclusion [14-15]. Furthermore, recent studies have found that Lp-G18 can enhance the expression of specific proteins in intestinal epithelial cells, such as tight junction proteins claudins and occludin. This enhancement strengthens the tight junctions of intestinal epithelial cells, reduces intestinal permeability, and prevents pathogens and harmful substances from crossing the intestinal barrier into the bloodstream [16]. Moreover, Lp-G18 can produce short-chain fatty acids (SCFAs) like acetate, propionate, and butyrate in the intestine, which can cross the blood-brain barrier and affect brain functions, including emotional and cognitive processes [16].

Lp-G18 can also influence the host's immune response through interactions with intestinal epithelial cells and immune cells like dendritic cells and macrophages. It can stimulate beneficial immune responses, such as increasing the number and activity of regulatory T cells (Tregs), while reducing the production of pro-inflammatory cytokines like tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6) [14-15].

4.2. Bioinformatics Analysis

We performed an in-depth bioinformatics analysis using the nucleotide data of Lp-G18 (BioProject ID: PRJNA715285) from the NCBI BioProject database [18]. Our aim was to conduct a preliminary theoretical study on Lp-G18 to reveal its basic genomic characteristics and to compare it with other lactic acid bacteria strains, providing a foundation for subsequent research.

Our phylogenetic tree analysis (see *Figure 4*) illustrated the relationship between Lp-G18 and other *Lactobacillus plantarum* strains. By constructing a phylogenetic tree based on 16S rRNA gene sequences, we observed that Lp-G18 has a high degree of homology with known *L. plantarum* strains. This finding indicates its stability and conservation within the *Lactobacillus* genus. Our genomic circular map (see *Figure 5*) displays its basic genomic characteristics [19].

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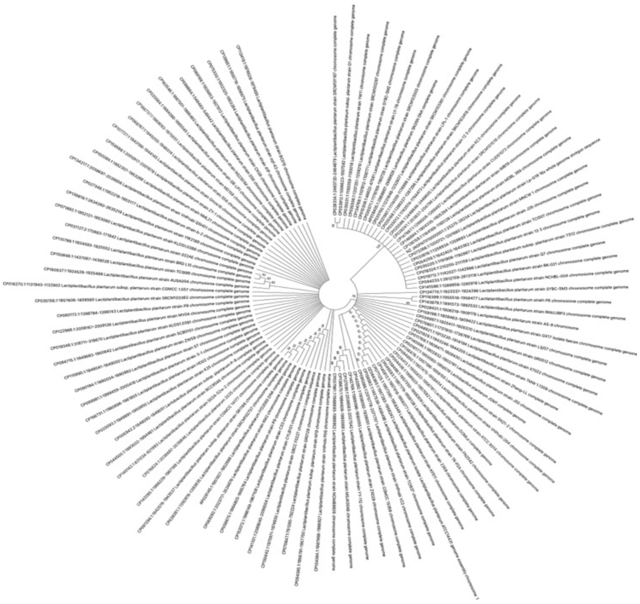

Figure 4: Phylogenetic Tree Analysis of Lp-G18

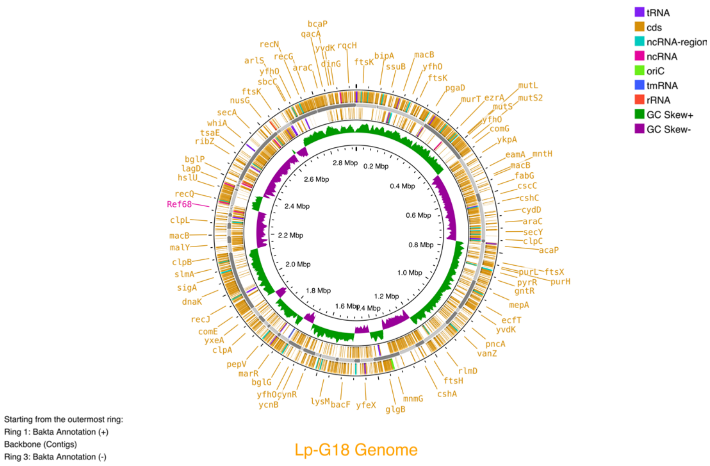

Figure 5: Genomic Circular Map of LP-G18

5. Discussion

This study aimed to explore the effects of probiotic supplementation on adolescents' cognitive abilities, memory, and emotional states, based on the gut-brain axis theory. Through PANAS and MoCA assessments conducted on adolescent subjects, we observed the potential impacts of probiotics on the cognitive function and emotional health of the target population.

In terms of cognitive function, the paired-sample t-test analysis of MoCA scores revealed significant differences between the control and experimental groups. While the control group showed a slight increase in MoCA scores during the study, this change was not statistically significant. In contrast, the experimental group exhibited a significant improvement in cognitive performance, evidenced by a notable increase in MoCA scores from baseline to the end of the study. This suggests that probiotic intake may have a positive effect on the cognitive abilities of adolescents aged 15 to 18.

Furthermore, the paired-sample t-test analysis of PANAS data provided evidence for the potential impact of probiotics on emotional states. The experimental group showed a significant increase in positive emotions during the study, reflecting an improvement in emotional health after receiving the supplement. In comparison, the control group did not exhibit significant changes in positive emotions. These results indicate that probiotics may have a positive regulatory effect on the emotional states of adolescents.

Of particular interest is the significant role played by the strain Lp-G18 (*Lactobacillus plantarum*) in

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the composite probiotic supplement used in this study. Previous research has shown that Lp-G18 can enhance intestinal barrier function by competitively excluding pathogenic bacteria and can directly influence brain functions—including cognition and emotion—through the production of metabolic products like short-chain fatty acids [14-16]. These mechanisms may explain the significant improvements in cognitive function and emotional state observed in the experimental group. The presence of Lp-G18 may enhance the bidirectional regulatory function of the gut-brain axis by modulating the gut microbiota, thereby promoting emotional health and cognitive abilities.

Our findings resonate with existing literature, emphasizing the importance of bidirectional communication via the gut-brain axis mediated by the gut microbiota [20]. Probiotics show potential in improving cognitive function and emotional states by modulating the gut microbiota. The improvements observed in the experimental group regarding cognitive abilities and positive emotions support the positive regulatory role of probiotics on the gut-brain axis and may have a beneficial impact on enhancing cognitive performance and emotional health.

However, this study has some limitations. The sample size was small, with a total of 15 participants divided into control and experimental groups of 7 and 8 each. A larger sample size would enhance the generalizability and statistical power of the findings. Additionally, the intervention period was only four weeks, which may not have been sufficient to capture the long-term effects of probiotics on cognitive function and emotional states. Future studies should consider extending the intervention duration to evaluate the long-term effects of probiotics on adolescent cognition and emotion.

This study employed a double-blind design to reduce bias and ensure research rigor. Both participants and researchers were unaware of group assignments, enhancing the credibility of the results and providing more reliable evidence for the effects of probiotics on cognitive and emotional outcomes.

In conclusion, this study provides new insights into the potential benefits of probiotics in promoting cognitive abilities and positive emotions in adolescents. The findings support the bidirectional communication role of the gut microbiota in the gut-brain axis and suggest that interventions targeting the gut microbiota—such as probiotic supplementation—may have positive effects on cognitive function and emotional health. Future research should consider increasing the sample size and extending the intervention period to validate and expand upon these findings, providing scientific evidence for the potential therapeutic applications of probiotics in adolescent populations.

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Author Contributions Statement

B.C., Z.W., Y.Z., and W.Z. conceived the experiment(s), B.C. wrote the manuscript, B.C., Z.W. and Y.Z. conducted the experiment(s), B.C. analysed the results. All authors reviewed the manuscript.

Data Availability Statement

The authors confirm that the data supporting the findings of this study are available within the article and its Supplementary material. Raw data that support the findings of this study are available from the corresponding author, upon reasonable request.

References

[1] Martin C R, Osadchiy V, Kalani A, et al. The Brain-Gut-Microbiome Axis[J]. Cellular and Molecular Gastroenterology and Hepatology, 2018, 6(2): 133–148.
[2] Guo C, He P. Brain-gut Axis and Its Research Progress[J]. World Latest Medicine Information, 2017, 17(95): 89–91.
[3] Akbari E, Asemi Z, Daneshvar Kakhaki R, et al. Effect of Probiotic Supplementation on Cognitive

ISSN 2616-5791 Vol. 6, Issue 1: 38-45, DOI: 10.25236/AJMHS.2025.060106

Function and Metabolic Status in Alzheimer's Disease: a Randomized, Double-Blind and Controlled Trial[J]. Frontiers in Aging Neuroscience, 2016, 8(256).

[4] Morton J T, Jin D-M, Mills R H, et al. Multi-level analysis of the gut–brain axis shows autism spectrum disorder-associated molecular and microbial profiles[J]. Nature Neuroscience, 2023, 26(7): 1208–1217.

[5] Cryan J F, Dinan T G. Mind-altering microorganisms: the Impact of the Gut Microbiota on Brain and Behaviour [J]. Nature Reviews Neuroscience, 2012, 13(10): 701–712.

[6] Tosti V, Bertozzi B, Fontana L. Health Benefits of the Mediterranean Diet: Metabolic and Molecular Mechanisms [J]. The Journals of gerontology. Series A, Biological Sciences and Medical Sciences, United States: 2017, 73(3): 318–326.

[7] Wang A, Lin Y, Wu Y, et al. Macronutrients intake and risk of Parkinson's disease: A meta-analysis[J]. Geriatrics & Gerontology International, 2014, 15(5): 606–616.

[8] Houser M C, Chang J, Factor S A, et al. Stool Immune Profiles Evince Gastrointestinal Inflammation in Parkinson's Disease[J]. Movement Disorders, 2018, 33(5): 793–804.

[9] Wallace C J K, Milev R. The effects of probiotics on depressive symptoms in humans: A systematic review [J]. Annals of General Psychiatry, 2017, 16(1).

[10] Watson D, Clark L A, Tellegen A. Development and validation of brief measures of positive and negative affect: The PANAS scales.[J]. Journal of Personality and Social Psychology, 1988, 54(6): 1063–1070.

[11] Nasreddine Z S, Phillips N A, Bedirian V, et al. The Montreal Cognitive Assessment, MoCA: A brief screening tool for mild cognitive impairment[J]. Journal of the American Geriatrics Society, 2005, 53(4): 695–699.

[12] Gao Y, Zhang G, Jiang S, et al. Wekemo Bioincloud: A user-friendly platform for meta-omics data analyses [J]. iMeta, Wiley, 2024, 3(e175).

[13] Zhang H, Xu J. Modern Psychology and Educational Statistics[M]. Beijing Normal University Press, 2007.

[14] Fortes P M, Vieira R, Henrique L, et al. Inflammatory cytokines and lipid profile in children and adolescents with nephrotic syndrome receiving L. Plantarum: a randomized, controlled feasibility trial [J]. Journal of the Brazilian Medical Association, Brazilian Medical Association, 2020, 66(11): 1487–1492.

[15] Anca-Ioana (Amzăr) Scărlătescu, Bruno Ștefan Velescu, Miruna-Maria Apetroaei, et al. Practices and Trends in the Prescription of Probiotics: A Detailed Analysis in an Eastern European Region[J]. Processes, Multidisciplinary Digital Publishing Institute, 2024, 12(9): 1856–1856.

[16] Almeida M, Alexia Figueiredo Ferreira, Caetano C, et al. Development and Characterization of Hydroxyethyl Cellulose-Based Gels Containing Lactobacilli Strains: Evaluation of Antimicrobial Effects in In Vitro and Ex Vivo Models[J]. Pharmaceuticals, 2023, 16(3): 468–468.

[17] Joshi N. Objective Assessment of Retinal Ganglion Cell Function in Glaucoma[D]. SUNY College of Optometry: 2017.

[18] Research and Development Center, Biogrowing Co., Ltd. Lactiplantibacillus plantarum strain:Lp-G18 (ID 715285) - BioProject - NCBI[EB/OL]. Nih.gov. 2021-03-27/2024-09-29. https://www.ncbi.nlm. nih.gov/ bioproject/715285.

[19] Grant J R, Enns E, Marinier E, et al. Proksee: in-depth characterization and visualization of bacterial genomes[J]. Nucleic Acids Research, Oxford University Press, 2023, 51(W1): W484–W492.

[20] Xie S. A Psychosomatic Study of Test Anxiety and Gastrointestinal Discomfort in students: Based on the Theory of brain-gut Axis[J]. Examination Weekly, 2018, 60: 1–3.