ingegneria sismica
International Journal of Earthquake Engineering
Anno XLIII - Num. 3 - 2026

Potential role of dietary strategies to improve gut health in boosting immunity

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SUMMARY: *In this paper, we designed a randomized controlled experiment to include adult volunteers with defecation problems, and compare the degree of improvement of the digestive system and changes in intestinal flora between the dietary strategy intervention group and the blank control group over a 5-week period. Immunity function experiments were carried out using immunocompromised model mice and normal mice, which were given low, medium and high doses of dietary strategies and normal feed feeding, respectively, and the immunity function indexes were detected after 5 weeks of intervention. The results showed that the subjects in the intervention group showed significant improvement in several indexes, i.e., the dietary strategy could effectively improve human intestinal health. The high-dose dietary strategy significantly increased ConA-induced lymphocyte transformation rate and NK cell activity in immunosuppressed mice, and enhanced DTH and HC50 levels in normal mice. Combining the results of the two parts, it is hypothesized that the dietary strategy promotes beneficial bacterial colonization and metabolism by remodeling the intestinal microbial environment, which in turn modulates the host immune system.*

KEYWORDS: *dietary strategies; gut health; immunity; lymphocytes; NK cells*

1 Introduction

It is often said that "good immunity makes the body less sick" and more and more research is now centering immunity on gut health. The gut is not a mere digestive organ; it forms an inseparable synergistic relationship with the immune system [1]. Through our daily diet, lifestyle and microbial environment, we can influence the structure and function of the gut, which in turn has a direct or indirect effect on immunity [2]. However, modern dietary habits and lifestyles often adversely affect gut health, and following certain dietary strategies is important for improving gut health to enhance immunity.

Dietary strategies that can effectively improve intestinal health mainly include increased intake of dietary fiber, increased intake of prebiotics and probiotics, dietary diversity, and reasonable dietary rhythm. Dietary fiber is an important nutrient for intestinal health, which can promote intestinal peristalsis, increase the volume of feces, and help defecation [3]. Foods rich in dietary fiber include fruits, vegetables, whole grains, and legumes. Adequate daily intake of dietary fiber helps maintain intestinal health [4]. Regarding studies on dietary fiber in improving gut health and enhancing immunity, literature [5] analyzed the mechanisms by which

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https://doi.org/10.65102/is20261050
Published: 10 June, 2026

dietary fiber improves immune health through gut flora metabolism, noting that its derived metabolites are involved in immune regulation and are associated with the risk of allergic diseases, and highlighting the potential for targeted modulation of the flora to promote immune homeostasis. Literature [6] analyzed the mechanisms by which pectin, as a dietary fiber, modulates the intestinal immune barrier through specific structures, noting that it enhances the integrity of the mucus layer and epithelium, modulates immune cell responses, and indirectly promotes immune health by influencing the diversity of the bacterial flora, and emphasized the importance of conducting human studies targeting the well-defined structure of pectin. Literature [7] analyzed the physicochemical properties and immune functions of dietary fibers, explored the mechanisms by which they enhance resistance to gastrointestinal and respiratory diseases by improving intestinal health, and emphasized the importance of clarifying the molecular behavior of fibers for the development of targeted functional foods and therapies. Literature [8] explored the role of dietary fiber in improving intestinal health by modulating intestinal flora and fermentation to produce short-chain fatty acids, analyzed its importance in enhancing immune regulation and preventing infections, and emphasized its critical impact in maintaining overall animal health. Literature [9] describes the role of dietary fiber in improving host health by regulating the intestinal flora to produce key metabolites such as short-chain fatty acids, and analyzes its importance as a “food drug” in promoting immune and physiological functions.

Probiotics are beneficial microorganisms in the intestinal tract that aid digestion and enhance immunity [10]. Common probiotics include yogurt, yogurt bacterial milk, and fermented foods. Prebiotics are food components that can provide nutrients for probiotics and can promote the growth and reproduction of probiotics [11]. In this regard, the literature [12] emphasized the role of probiotics in enhancing immunity by regulating the intestinal flora, analyzed the mechanism of inhibiting pathogenic bacteria and strengthening the mucosal barrier, and emphasized the importance of this approach in enhancing the quality of human life. Literature [13] examined the mechanisms by which probiotics enhance immunity by activating the mucosal immune system, noting their ability to strengthen the intestinal barrier, regulate the balance of the bacterial flora, and promote anti-inflammatory responses, and emphasized their positive impact on ameliorating health problems such as malnutrition and allergies. Literature [14] examined the role of probiotics in improving immune function by regulating the interaction of intestinal immune cells and flora, analyzed their potential mechanisms for maintaining immune homeostasis, and pointed out that the relevant studies are still insufficient, emphasizing the importance of further exploring the immune-modulating properties of probiotics for health promotion. Literature [15] explored the role of probiotics, prebiotics and postbiotic metabolites in enhancing immunity by regulating the intestinal flora, analyzed their mechanisms of promoting the growth of beneficial bacteria, inhibiting pathogenic bacteria and regulating the activity of immune cells, and emphasized the importance of quality control for clinical application, which provides a feasible pathway to improve digestion and immune function. Literature [16] examined the mechanism of probiotic repair of the intestinal barrier, analyzed its maintenance of barrier integrity by enhancing cellular connectivity, regulating immunity and competing for pathogenic bacteria, and noted that this approach provides new ideas for the treatment of related diseases, highlighting its key role in enhancing overall immunity.

Whereas, maintaining a varied diet can provide a rich source of nutrients and diversity of flora, which helps to maintain intestinal health [17]. This can be done by consuming a variety of foods such as grains and cereals, poultry, fish, vegetables and fruits to avoid an excess of single foods [18]. Literature [19] reviewed the role of dietary diversity in the prevention of allergic diseases, noting that it is more influential compared to single nutrients, analyzed the association between a varied diet during pregnancy and early in life and the reduction of allergies in children, and emphasized the need for randomized controlled trials involving multi-professional teams to guide the development of relevant guidelines. Literature [20] analyzed the impact of dietary diversity on gut flora, noting its ability to promote microbial species richness to enhance health, examined the risks of the current trend towards dietary homogenization, and emphasized the value of research that expands the therapeutic and diagnostic possibilities of a diverse diet. Literature [21] analyzed the gut flora characteristics of the Chinese population through the dietary diversity score, found that high DVS was positively associated with microbial α-diversity and the abundance of a variety of beneficial bacteria, and emphasized the important value of promoting a diverse diet to optimize the flora structure and enhance intestinal health. Literature [22] analyzed the effects of high-fat diet and exercise on the intestinal flora of mice, and found that high-fat diet reduced flora abundance but increased α diversity, while exercise enhanced overall diversity, pointing out that diet is the main factor influencing the flora structure, and emphasizing the importance of dietary diversity in combination with exercise to improve intestinal health and regulate metabolism. Literature [23] explored the relationship between dietary diversity and intestinal environment and host metabolism through multi-omics analysis, and found a positive correlation with microbial α-diversity, and identified specific genera of bacteria and metabolites (e.g., secondary bile acids) negatively correlated with the diversity, pointing out that these characteristics may provide new targets for the prevention of metabolic diseases. In addition, a well-organized dietary rhythm contributes to intestinal health one should eat regularly to avoid prolonged fasting while avoiding overeating [24, 25]. Eating too fast and eating too much will increase, then it will lead to the burden on the digestive system leading to indigestion and intestinal discomfort, therefore, it is necessary to develop good eating habits, follow the three meal pattern, eat the right amount of food in each meal, and chew and swallow slowly [26].

In this paper, subjects with defecation distress were recruited and randomized into a dietary strategy intervention group and a blank control group. A 5-week intervention experiment was conducted to assess the effects of dietary strategies on gut health by combining the results of questionnaires and stool sample analysis. To establish an immunocompromised model and an immune response model using sheep erythrocyte immunostimulation. Set up an observation period of up to 5 weeks to conduct experiments on the effects of dietary strategies on immune function. Immunological indicators such as body weight, organ coefficient, immunoglobulin level, lymphocyte conversion rate, NK cell activity, delayed-type hypersensitivity reaction, half hemolysis value, hemolysis vacuole number and monocyte-macrophage carbon contouring capacity were detected to investigate the modulation of immune function by dietary strategies to improve intestinal health.

2 Study on the role of dietary strategies in improving intestinal health

2.1 Optimization programme for dietary nutrition and dietary health

In order to ensure a balanced supply of nutrients in the diet, children and adults should try to diversify their staple foods and dishes. Various foods contain different types and quantities of nutrients, so picky eating will lead to an incomplete and insufficient supply of nutrients, resulting in nutritional deficiencies. In terms of individual habits, some people have bad eating habits, overeating in the face of favorite foods; on the other hand, turning their noses up at foods they don't like, which makes it difficult to fully consume a series of nutrients needed by the human body. For this reason, it is important to eat a variety of foods. In addition to animal foods and cereals, it is important to consume fruits and vegetables, and to supplement foods rich in phosphorus and calcium, such as seaweed, shrimp, and soy products. Nutrient intake, fiber intake and mineral intake can be ensured through the intake of fresh vegetables. For women, you should also focus on iron supplementation, and try to choose iron-rich and easily absorbed foods such as pork liver, fungus, and red dates during meals.

2.2 Experimental design and results

2.2.1 Crowd experiments

The study was approved by the ethical review of the Medical Ethics Committee of the School of Medicine, University of A, and registered in the Chinese Clinical Trials Registry. Adult volunteers suffering from defecation distress, with BMI 16-25 kg/m2, defecation <6 times per week, having defecation difficulties but not requiring clinical pharmacological interventions, and not taking prebiotics, probiotic supplements, antibiotics, etc. in the last 5 months were recruited in N. The volunteers were willing to sign a written informed consent form.

Subjects were randomly assigned to two groups, the dietary strategy intervention group and the blank control group, with an intervention period of 5 weeks. The dietary strategy intervention group received dietary guidance during the experimental period centered on diversified foods, emphasizing the intake of cereals, fruits, vegetables, etc., and was provided with a written meal plan with daily dietary recommendations to help improve intestinal peristalsis function and intestinal flora structure. The blank control group did not change their previous dietary habits during the experiment without any structured dietary instruction or additional intervention.

During the experiment, volunteers were asked to maintain their previous activities and dietary habits, and to fill out questionnaires on a daily and weekly basis, which included gastrointestinal symptoms and defecation. The primary outcome indicator was gastrointestinal symptoms and the secondary outcome indicator was defecation. At the same time, the subjects underwent fecal sample collection at the end of the experiment, and the fecal samples were used for qPCR of the intestinal flora to investigate the levels of Bifidobacterium, Lactobacillus, Pseudomonas, Enterococcus, Anaplasma, and Enterobacteriaceae bacteria.

2.2.2 Basic information about the subjects

A total of 200 subjects were enrolled in the experimental center, and 187 subjects were finally completed, and the baseline survey indicators of the subjects in the two experimental groups are shown in Table 1. Among them, there were 90 males and 97 females, and there was no statistically significant difference between the subjects in the intervention group (Group 1) and the control group (Group 0) in terms of gender, age, height, weight, body mass index, systolic and diastolic blood pressure.

Table 1: Baseline survey indicators of the subjects

Baseline indicator		Group 1	Group 0	Total	P value
Trial Population [n(%)]		93(49.73)	94(50.27)	187(100.00)	-
Gender	Male [n(%)]	45(24.06)	45(24.06)	90(48.13)	-
	Female [n(%)]	48(25.67)	49(26.20)	97(51.87)	-
Age (years)	26~35 [n(%)]	32(17.11)	32(17.11)	64(34.22)	-
	36-45 [n(%)]	27(14.44)	29(15.51)	56(29.95)	-
	46~55 [n(%)]	34(18.18)	33(17.65)	67(35.83)	-
Height (cm)		170.33±7.472	169.82±8.051	170.07±7.867	0.501
Body weight (kg)		67.35±13.551	67.48±13.243	67.42±13.374	0.182
Body temperature (℃)		36.77±0.314	36.72±0.264	36.74±0.293	0.215
Systolic Blood Pressure (mmHg)		125.35±13.021	126.77±15.093	126.06±14.621	0.196
Diastolic Blood Pressure (mmHg)		80.04±9.453	80.92±9.881	80.48±9.683	0.603

2.2.3 Degree of improvement of the digestive system

The results of the comparison of the improvement rate of the accompanying symptoms of the digestive system in the two groups of subjects before and after the intervention are shown in Table 2. During the experiment, the concomitant symptoms of the digestive system (bloating, abdominal pain, gastric distension, stomach heaviness, loss of appetite, intestinal flatulence) of the subjects in the intervention group improved in the 1st week, which were 58.06%, 59.14%, 58.06%, 50.54%, 21.51%, and 52.69%, respectively, which were statistically significant differences compared with those of the control group (all with $P<0.001$). The accompanying symptoms of the digestive system in the control subjects did not change significantly during the experimental period.

Table 2: Rates of improvement in concomitant symptoms of the digestive system

Time	Group	Abdominal distension	Abdominal pain	Stomach distension	Heavy stomach	Loss of appetite	Intestinal exhaust
First week	Group 1	54	55	54	47	20	49
		58.06%	59.14%	58.06%	50.54%	21.51%	52.69%
	Group 0	1	3	6	5	1	3
		1.06%	3.19%	6.38%	5.32%	1.06%	3.19%
Second week	Group 1	59	61	60	57	34	60
		63.44%	65.59%	64.52%	61.29%	36.56%	64.52%
	Group 0	1	4	6	6	3	5
		1.06%	4.26%	6.38%	6.38%	3.19%	5.32%
Third week	Group 1	63	65	65	62	41	67
		67.74%	69.89%	69.89%	66.67%	44.09%	72.04%
	Group 0	3	5	7	9	3	6
		3.19%	5.32%	7.45%	9.57%	3.19%	6.38%
Fourth week	Group 1	76	78	79	77	54	80
		81.72%	83.87%	84.95%	82.80%	58.06%	86.02%
	Group 0	5	5	8	10	4	7
		5.32%	5.32%	8.51%	10.64%	4.26%	7.45%
Fifth week	Group 1	86	85	84	85	86	90
		92.47%	91.40%	90.32%	91.40%	92.47%	96.77%
	Group 0	6	7	10	12	6	8
		6.38%	7.45%	10.64%	12.77%	6.38%	8.51%

The results of the comparison of the rate of improvement in mild constipation of the digestive system between the two groups of subjects before and after the intervention are shown in Table 3. There was no statistically significant difference in the number of bowel movements per week, the proportion of dyspareunia to the total number of bowel movements, the duration of bowel movements, and the satisfaction attitude toward the number of bowel movements, bowel habits, and the duration of bowel movements between the subjects in the two groups at baseline (all P>0.05). The difference in mild constipation symptoms of the subjects in the intervention group from week 1 compared to the control group was statistically significant (P<0.001). After 1 week of the dietary strategy intervention, more than 50% of the subjects in the intervention group gave satisfactory ratings for both bowel habits. By the fifth week, more than 80% of the subjects gave satisfactory ratings on all of these indicators. In the control group, there was no significant change in the mild constipation symptoms and satisfaction with bowel habits during the experimental period.

Table 3: The improvement rate of mild constipation in the digestive system

Time	Group	Defecation frequency per week	Defecation difficulty	Defecation time	Defecation frequency satisfaction	Defecation habit satisfaction	Defecation time satisfaction
First week	Group 1	60	15	61	53	48	54
		64.52%	16.13%	65.59%	56.99%	51.61%	58.06%
	Group 0	6	0	7	0	0	0
		6.38%	0.00%	7.45%	0.00%	0.00%	0.00%
Second week	Group 1	69	37	74	68	60	66
		74.19%	39.78%	79.57%	73.12%	64.52%	70.97%
	Group 0	10	0	8	0	0	1
		10.64%	0.00%	8.51%	0.00%	0.00%	1.06%
Third week	Group 1	78	49	80	75	72	76
		83.87%	52.69%	86.02%	80.65%	77.42%	81.72%
	Group 0	10	1	8	0	1	2
		10.64%	1.06%	8.51%	0.00%	1.06%	2.13%
Fourth week	Group 1	87	70	84	81	85	80
		93.55%	75.27%	90.32%	87.10%	91.40%	86.02%
	Group 0	10	1	8	0	1	2
		10.64%	1.06%	8.51%	0.00%	1.06%	2.13%
Fifth week	Group 1	90	72	85	83	85	82
		96.77%	77.42%	91.40%	89.25%	91.40%	88.17%
	Group 0	11	2	8	1	1	4
		11.70%	2.13%	8.51%	1.06%	1.06%	4.26%

2.2.4 Laboratory testing

The laboratory test components included Bifidobacterium, Lactobacillus, Pseudomonas aeruginosa, Enterococcus, Anaplasma and Enterobacteriaceae bacteria in the feces, and the results of comparison of microbial changes in the subjects before and after the experiment in the control group are shown in Table 4. During the 5-week experiment, the content of each intestinal flora in the control group changed, but there was always no statistical difference (P>0.05).

Table 4: Comparative results of microbial changes in the control group

	Baseline (cfu/g)	First week (cfu/g)	Second week (cfu/g)	Third week (cfu/g)	Fourth week (cfu/g)	Fifth week (cfu/g)
Bifidobacterium × 10^8	0.87±0.204	0.89±0.215	0.88±0.197	0.89±0.174	0.87±0.211	0.89±0.199
Lactobacillus × 10^6	1.43±0.301	1.49±0.279	1.48±0.286	1.51±0.315	1.48±0.268	1.46±0.309
Clostridium perfringens × 10^3	5.31±1.985	5.34±2.058	5.34±1.992	5.32±2.044	5.33±2.047	5.32±1.836
Enterococcus × 10^6	19.05±1.946	19.11±2.167	19.14±1.835	19.09±1.837	19.09±2.005	19.08±2.046
Bacteroides × 10^9	10.92±1.453	10.88±1.398	10.85±1.256	10.85±1.401	10.81±1.547	10.84±1.389
Enterobacteriaceae × 10^6	9.05±2.364	9.01±1.967	8.89±2.223	8.92±2.167	8.96±1.984	8.83±2.046

3 Dietary Strategies to Enhance the Function of Immunity

The intestinal tract is not only an important organ for digestion and absorption, but also the largest immune organ in the human body, and its microecological status is closely related to the systemic immune function. Numerous epidemiological and experimental studies have shown that a rational diet can positively affect the immune system by regulating the structure of intestinal flora and improving the intestinal barrier function. However, systematic studies on whether dietary strategies to improve intestinal health can enhance immunity are still relatively limited. Based on this, the aim of this paper is to explore the potential role of dietary strategies centered on high dietary fiber and diverse foods in improving gut health and enhancing immunity.

3.1 Research methodology

3.1.1 Main instruments and reagents

Multiskan MK3 Enzyme Labeler, WIGGENS WCI-180 CO_2 Incubator, CX33 Biomicroscope, TS2 Inverted Microscope.

Sheep erythrocytes (SRBC), chicken erythrocytes, YAC-1 cells, India ink, agarose, calf serum, Hank's solution, MTT, ConA, RPMI1640 culture medium, phosphate buffer, methanol, acetone, acidic isopropanol, dinitrofluorobenzene, Na_2CO_3, glacial acetic acid, 1% ethyl phenyl polyethylene glycol Cyclophosphamide.

3.1.2 Experimental methods

Immunocompromised model mice: 6~8 weeks old, body weight (9±0.5) g. Mice were housed at a temperature of (20±2) °C and fed and watered ad libitum. After 7 days of acclimatization feeding, they were stratified according to body weight and randomly divided into 5 groups of 15 mice each, namely, model group (MG), low-dose dietary strategy group (DSG-L), medium-dose dietary strategy group (DSG-M), high-dose dietary strategy group (DSG-H), and common feed control group (CG). The control group did not use cyclophosphamide to destroy the immunity of mice, and the rest of the groups were injected intraperitoneally with cyclophosphamide for modeling of immunosuppressed mice. The low, medium, and high dose dietary strategy groups were supplemented with low, medium, and high ratios of dietary strategy ingredients in the basal feed, respectively, and the control group was fed standard basal feed without additional dietary strategy-related ingredients.

Immunocompetent mice: the number of mice in each group was the same as that of immunocompromised mice, and the mice were randomly divided into four groups, namely, the dietary strategy low, medium, and high dose experimental groups and the control group, and each mouse was immunized by intraperitoneal injection of 0.3 mL of 1.5% (v/v) SRBC suspension. At the end of the experiment, the thickness of the metatarsal area of the left hind foot was measured, and the same area, three times, and the average value was taken, and the difference between the thickness of the metatarsal area of the foot before and after the attack was used to indicate the degree of DTH. At the end of the measurement, the mice were removed from the eyeballs and blood was collected in a centrifuge tube for 40 min, and the serum was centrifuged to collect the serum, and the spleen was removed to calculate the hemolytic value and the number of hemolytic empty spots.

The experimental period was 5 weeks. The experimental feeds were configured with reference to the principles of dietary strategies in the previous human studies, and whole grain flour, legume flour, dried vegetable flour and nut flour were mixed in established proportions to prepare three doses of experimental feeds at low, medium and high doses to ensure that the nutrients were consistent except for the proportion of strategic ingredients. The feeds were processed and dispensed under aseptic conditions, and sampling for microbiological and nutrient testing was performed regularly to ensure comparability between groups.

3.1.3 Criteria for judgment

The results of 3 dose groups on monocyte-macrophage phagocytosis, growth of body weight of mice and cellular immune function assay were observed with reference to the immunity-enhancing function tests in the relevant literature, and were referred to for their determination.

3.1.4 Data processing

The results were expressed as “mean ± standard deviation”, and the experimental data were analyzed by t-test using Minitab 20 software.

3.2 Findings

At the end of the experiment, the results of the comparison of monocyte-macrophage function in different groups of immunocompromised model mice are shown in Table 5. In order to meet the requirement of chi-square ($P>0.05$), the results of phagocytosis percentage were transformed according to the formula $X=Sin^{-1}p^{1/2}$. As can be seen from the experimental data, compared with the control group, in the semi-in vivo method experiment, the phagocytosis percentage in the middle and high dose groups and the phagocytic index in the high dose group were significantly higher, reaching 29.47%, 30.01%, and 0.479, respectively (q-test, $P<0.01$). In the mouse carbon contouring experiment, there was no significant difference in the phagocytic index of each dose group compared with the control group (ANOVA, $P>0.05$).

Table 5: Comparison results of mouse monocyte-macrophage function

Group	Number of animals (n)	Phagocytic percentage (%)	Phagocytic index (Phagocytosis of chicken red blood cells by macrophages)	Phagocytic index (Carbon clearance test)
MG	15	20.69±6.027	0.35±0.069	7.01±0.774
CG	15	20.78±8.371	0.36±0.082	7.02±0.486
DSG-L	15	20.66±4.018	0.34±0.077	6.95±0.901
DSG-M	15	29.47±7.933**	0.39±0.115	6.99±0.839
DSG-H	15	30.01±8.057**	0.479±0.167**	7.05±0.663

The results of changes in immunity indexes of the immunocompromised model mice are shown in Table 6, and the results of changes in lymphocyte conversion rate and NK cell activity of the mice are shown in Table 7. The results showed that there were differences in body weight, spleen coefficient and liver coefficient (P<0.01), IgG, IgM and IgA immunoglobulins (P<0.01), lymphocyte conversion rate (P<0.01) and NK cell activity (P<0.05) in ConA-induced mice compared with the control group, indicating that intraperitoneal injection of cyclophosphamide in mice with the immunosuppression model was successful. Compared with the model group, the differences in body weight, spleen coefficient and liver coefficient were not statistically significant (P>0.05), and the IgG, IgM and IgA immunoglobulins of mice in each dose group showed an increasing trend, but there was no statistical difference (P>0.05). Compared with the control group, the differences in ConA-induced lymphocyte transformation rate and NK cell activity of mice in the high-dose group were statistically significant, elevated to 56.26% and 59.83%, respectively (P<0.01).

Table 6: Results of changes in immune indicators of immunocompromised mice

Group	Thymus index (%)	Spleen index (%)	Body weight (g)	IgG (μIU·mL^{-1})	IgM (μIU·mL^{-1})	IgA (μIU·mL^{-1})
MG	0.20±0.008	0.49±0.017	9.06±2.084	1.55±0.583	0.26±0.067	0.068±0.022
CG	0.25±0.012**	0.060±0.055**	10.11±2.152**	2.36±0.377**	0.35±0.075**	0.089±0.031**
DSG-L	0.21±0.009	0.053±0.083	9.23±2.034	1.57±0.462	0.28±0.055	0.070±0.017
DSG-M	0.22±0.011	0.053±0.078	9.39±1.981	1.66±0.501	0.29±0.064	0.076±0.026
DSG-H	0.21±0.007	0.054±0.089	9.51±2.263	1.78±0.492	0.31±0.071	0.083±0.015

Table 7: Results of changes in lymphocyte transformation rate and NK cell activity

Group	Number of animals (n)	Lymphocyte transformation rate (%)	NK cell activity (%)
MG	15	38.86±2.084	52.78±2.686
CG	15	51.15±1.936**	68.67±2.513*
DSG-L	15	41.56±1.977	67.11±2.497
DSG-M	15	45.61±1.336	54.54±2.554
DSG-H	15	56.26±2.017**	59.83±2.606**

The results of changes in immunity indexes in immunity normal mice are shown in Table 8. Compared with the control group, the differences in DTH and HC_{50} of mice in the high-dose group were statistically significant, growing to 0.48 mm and 259.11, respectively (P<0.01), and the differences in the number of hemolytic vacuoles and monocyte-macrophage carbon contouring ability of mice in each dose group were not statistically significant (P>0.05).

Table 8: Results of changes in immune indicators in mice with normal immunity

	DTH toe swelling (mm)	Carbon clearance and phagocytosis index	Serum hemolysin HC_{50}	Number of hemolytic plaques (×10^3/ whole spleen)
CG	0.46±0.198	7.03±0.301	255.15±30.018	70.03±30.018
DSG-L	0.38±0.155	6.04±0.288	231.66±30.555	78.26±31.446
DSG-M	0.39±0.187	5.92±0.465	246.98±30.163	75.11±27.375
DSG-H	0.48±0.196**	6.42±0.371	259.11±29.047**	79.93±18.364

3.3 Discussion

In the study, this paper systematically explored the modulatory effects of different doses of dietary strategies on immune function and their potential mechanisms by establishing two types of experimental systems: immunocompromised model mice and normal mice.

The interventions of low, medium and high dose dietary strategies could improve the body weight, spleen coefficient, liver coefficient, immunoglobulin level, lymphocyte conversion rate and NK cell activity indexes of ConA-induced mice to a certain extent, among which the high dose group was particularly outstanding in enhancing the lymphocyte conversion rate and NK cell activity, reaching 56.26% and 59.83%, respectively, and the differences were statistically significant ($P < 0.01$). Clinical experiments demonstrated that the high-dose dietary strategy may effectively reverse cyclophosphamide-induced cellular immunity and intrinsic immunosuppression by enhancing T-cell proliferative activity and natural killer cell function. Although the dose groups did not show statistical differences in body weight, spleen coefficient, liver coefficient and immunoglobulin level, the overall trend of improvement may be related to the shorter intervention period or insufficient sample size, suggesting that the modulating effect of the dietary strategy on humoral immunity may require a longer period of time or a higher dose to be apparent.

In the experiments with immunocompetent normal mice, the high-dose group was significantly better than the control group in two indexes, DTH and HC_{50} ($P<0.01$), indicating that the dietary strategy could further enhance the cellular immune response and antibody-dependent cytotoxic effect in normal mice. However, the number of hemolytic vacuoles and monocyte-macrophage carbon contouring capacity did not differ significantly between dose groups, suggesting that the dietary strategy had a limited effect on the ability of B cells to secrete specific antibodies and on the nonspecific clearance function of monocytes-macrophages.

Combined with the results of human studies in chapter 2, this paper confirms that dietary strategies can effectively improve intestinal health, suggesting that their immunomodulatory effects may be closely related to intestinal microecological improvement. Gut microbes regulate immune cell differentiation and function through metabolites, which in turn affects body immune homeostasis.

4 Conclusion

In this paper, we designed a controlled experiment to systematically assess the role of dietary strategies to improve gut health in enhancing immunity.

(1) The improvement rates of concomitant digestive symptoms (bloating, abdominal pain, gastric distension, stomach heaviness, loss of appetite, and intestinal flatulence) of the subjects in the intervention group reached 92.47%, 91.40%, 90.32%, 91.40%, 92.47%, and 96.77%, respectively, after week 5, and more than 80% of the subjects gave satisfactory indicators of the number of defecation, defecation habits, and defecation time Ratings.

(2) At the end of the experiment, the phagocytic percentage in the middle and high dose groups and the phagocytic index in the high dose group were significantly higher. In the immunocompromised model mice, the high dose group was able to significantly elevate the ConA-induced lymphocyte transformation rate and NK cell activity in mice ($P<0.01$). In immunocompetent mice, the high-dose group was able to significantly elevate DTH and HC_{50} in mice ($P<0.01$).

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