



Development and Bioactivity Evaluation of a “Medicine and Food Homology” Composite Schisandra Chinensis Functional Beverage

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Abstract: Schisandra chinensis, a “medicine and food homology” herb, is highly regarded for its medicinal and nutritional value. Composite beverages that combine Schisandra chinensis with other medicinal and edible plants are gaining popularity due to their traditional health benefits. The orthogonal experiment results showed that the optimal formulation consisted of 30% Composite Schisandra chinensis extract (Schisandra chinensis: Lycium barbarum: Ophiopogon japonicus=4:1:1), 8% xylitol, and 0.010% citric acid. In vivo studies that all dose groups reduced malondialdehyde (MDA) and hydrogen peroxide (H₂O₂) levels to varying degrees (P<0.05), mitigating the oxidative damage induced by alcohol consumption. The anti-fatigue effect was more pronounced with higher concentrations of the Composite Schisandra chinensis beverage extract (P<0.05). No significant toxic side effects were observed in the beverage formulated with xylitol and citric acid as excipients. Additionally, all dosage groups significantly increased ear swelling difference (P<0.05) and modulated cyclophosphamide-induced organ index alterations in varying extents (P<0.05), indicating enhanced immune function in the experimental animals.

Keywords: medicine and food homology; composite Schisandra chinensis beverage; antioxidant activity; anti-fatigue effects; acute toxicity; immune enhancement

1. Introduction

Schisandra chinensis, a “medicine and food homology” herb, is highly regarded for its medicinal and nutritional value. As recorded in the Newly-Revised Materia Medica, it is described as “the skin tastes sweet, the flesh tastes sour, the seed tastes spicy and bitter, and all three have a salty flavor” which is reflected in its name, Schisandra chinensis.[1,2] In daily life, Schisandra chinensis is typically consumed in the form of wine or fruit, with products on the market including Schisandra chinensis tea bags, creams, and juices. With increasing research into its nutritional composition and functional benefits, Schisandra chinensis is gaining attention beyond its traditional use as a dried fruit in pharmacological studies.[3] The berries have attracted the interest of health product manufacturers, functional food producers, and other related industries. Among the emerging products, Composite beverages that combine Schisandra chinensis with other medicinal and edible plants are gaining popularity due to their traditional health benefits. These functional beverages are now entering the natural fruit juice market.[4] This paper presents a novel approach to developing and utilizing a complex functional beverage made from Schisandra chinensis, Lycium barbarum, and Ophiopogon japonicus, focusing on its antioxidant, anti-fatigue, acute toxicity, and immune-enhancing effects.

2. Methods

2.1 Materials

Schisandra chinensis and Lycium barbarum were sourced from the Chinese herbal medicine cultivation base in Zhenping County, Ankang City, Shanxi Province, China, while Ophiopogon japonicus was obtained from a similar cultivation base in Cixi City, Zhejiang Province, China.

2.2 Instruments and equipment

Model HA-HH-S1 constant temperature water bath, Beijing Heng odd Instrumentation Co., Ltd., Beijing, China; Model CAX-370 centrifuge, Tomy Seiko, Osaka, Japan; L15-P919 wall-breaker, Joyoung Co., Ltd., Shandong, China.

2.3 Sensory evaluation

Ten experienced personnel were selected to form a sensory evaluation team to assess the developed Composite Schisandra chinensis functional beverage in terms of color, odor, taste, and texture (Table 1).[5] In accordance with the pre-experimental herb ratio (Schisandra chinensis: Lycium barbarum: Ophiopogon japonicus = 4:1:1), the samples were first

homogenized, crushed, and precisely weighed. The extract was concentrated under reduced pressure to obtain the Composite Schisandra chinensis extract. To prepare the Composite Schisandra chinensis beverage, the extract was blended with xylitol and citric acid. The basic formulation included 25% Composite Schisandra chinensis extract, 6% xylitol, and 0.012% citric acid. Finally, based on the results from the one-way experiments, an L9(33) orthogonal experimental design was conducted (Table 2).[6]

Table 1. Criteria for sensory evaluation (out of 100)

Color (20 points)	Odor (30 points)	Taste (30 points)	Texture (20 points)
Purple, clear, bright, fresh, desirable (16-20 points)	Pure, fresh fragrance, clear, mild and soft (26-30 points)	Well-balanced flavor, slightly sour, smooth, no noticeable bitterness (26-30 points)	Clear, transparent, stable, no sediment (16-20 points)
Light purple, slightly unclear, mild (11-15 points)	Strong fragrance, not too mild, slightly off (21-25 points)	Slightly light, with mild bitterness (21-25 points)	Transparent, slight cloudiness, no turbidity (11-15 points)
Dark purple, dull, unclear, mild (6-10 points)	Strong fragrance with other off flavors (16-20 points)	Sour, slightly sweet, slightly bitter (16-20 points)	Slight cloudiness, slight turbidity (6-10 points)
Dark black, unclear, dull, with red tint (0-5 points)	No fragrance, off smells (11-15 points)	Sour, bitter, unpleasant taste (11-15 points)	Poor transparency, visible turbidity, unclear (0-5 points)

Table 2. The value of factor level of orthogonal experiment

Level	Factor		
	A (Composite Schisandra chinensis extract)/%	B (Xylitol)/%	C (Citric acid)/%
1	25	6	0.008
2	30	8	0.012
3	35	10	0.016

2.4 Animal experimentation

SPF-grade mice were obtained from Nanjing Qinglongshan Animal Breeding Farm (Licence No. SCXK (SU) 2024.0001), with their body weight maintained between 20 and 25 g. All mice were housed under identical conditions.

2.5 Antioxidant activity assay

Mice were subjected to continuous oral administration for 30 days. The blank control group received 60 mL of saline; the low-dose group and middle-dose group were respectively administered 20 mL and 40 mL of Composite Schisandra chinensis beverage supplemented with 20 mL of saline; and the high-dose group received 60 mL of Composite Schisandra chinensis beverage. After the final administration, the model group and dose groups were fasted for 12 h, followed by a single oral dose of 50% ethanol at 12 mL/kg (the blank control group remained untreated). [7, 8]

2.6 Anti-fatigue assay

The treatment groups were administered the concentrate by gavage at a dose of 10 mL/kg, while the control group received an equivalent volume of saline. This regimen was maintained for 30 consecutive days. Body weight was recorded every 2 days throughout the experiment. After the final administration, the mice were fasted for 30 min and subjected to a forced swimming test. The total swimming time was recorded to evaluate anti-fatigue capacity.[9,10]

2.7 Acute toxicity experiment

Group 1: Saline control (0.9 g NaCl dissolved in distilled water to 100 mL). Group 2: Excipient control (10 g xylitol and 0.008 g citric acid dissolved in distilled water to 100 mL). Group 3: Low-dose treatment (30 g Composite Schisandra chinensis extract combined with excipients from Group 2, dissolved in distilled water to 100 mL). Group 4: Medium-dose treatment (60 g Composite Schisandra chinensis extract combined with excipients from Group 2, dissolved in distilled water to 100 mL). Group 5: High-dose treatment (120 g Composite Schisandra chinensis extract combined with excipients from Group 2, dissolved in distilled water to 100 mL). Prior to administration, the mice were fasted for 12 h and then accurately weighed. Each group was administered the respective solution via oral gavage at a dose of 10 mL/kg, repeated three times at 4-hour intervals. The mice were observed continuously for 15 days. The heart, brain, liver, spleen, kidneys, and lungs were examined for any morphological or pathological changes.[11-13]

2.8 Immunoenhancement assay

An immunocompromised mouse model was established by intraperitoneal injection of cyclophosphamide (40 mg/kg)

for 2 consecutive days. On the 5th day after the final injection, relevant immunological indices, including the splenic index, thymic index, and ear swelling difference, were measured.[14] To evaluate delayed-type hypersensitivity (DTH), mice were sensitized using dinitrofluorobenzene (DNFB). The thymus and spleen were carefully excised and weighed. The thymus and spleen indices were calculated as the ratio of organ mass (mg) to body mass (g).[15]

3. Results

3.1 Results of single factor and orthogonal experiments

The results of the single factor experiment for the Schisandra chinensis beverage formulation are presented in Table 3-5. Based on these findings, the subsequent orthogonal experiment was designed with three factors at three levels (Table 6). Analysis of the R values from the range analysis revealed that the influence of each factor on the beverage quality followed the order: C>B>A. The optimal formulation combination was determined to be A2B3C1, corresponding to 30% Composite Schisandra chinensis extract (Schisandra chinensis: Lycium barbarum: Ophiopogon japonicus=4 : 1 : 1), 8% xylitol, and 0.010% citric acid.

Table 3. Results of single factor experiment of Composite Schisandra chinensis extract additive amount

Additive amount of Composite Schisandra chinensis extract	Score of sensory evaluation
25%	79
30%	78
35%	86
40%	83
45%	82

Table 4. Results of single factor experiment of xylitol additive amount

Additive amount of xylitol	Score of sensory evaluation
2%	78
4%	79
6%	82
8%	85
10%	81

Table 5. Results of single factor experiment of xylitol additive amount

Additive amount of citric acid	Score of sensory evaluation
0.008%	84
0.010%	85
0.012%	83
0.014%	81
0.016%	79

Table 6. Results of orthogonal experiment of Composite Schisandra chinensis beverage formulation

No.	A	B	C	sensory score/scores
1	1	1	1	84
2	1	2	3	78
3	1	3	2	79
4	2	1	3	76
5	2	2	2	80
6	2	3	1	86
7	3	1	2	82
8	3	2	1	83
9	3	3	3	83
K1	299	288	309	
K2	301	287	285	
K3	296	304	283	
R	1.73	3.04	5.87	

3.2 Results of validation experiment

Under the optimal conditions, the average sensory evaluation score of the three validation experiment groups was 93.9 points, which exceeded the results from the orthogonal experiment. The resulting beverage displayed a dark red color, a robust and harmonious flavor profile, a well-balanced sweet-sour taste, and a refreshing sensation. Additionally, the beverage showed excellent stability with minimal precipitation and stratification, indicating that the flavor optimization results were stable and reliable. Therefore, the optimal formulation of the Composite Schisandra chinensis beverage was determined to be 30% Composite Schisandra chinensis extract, 8% xylitol, and 0.010% citric acid.

3.3 Results of antioxidant experiment

Table 7 presents the results of the antioxidant experiment, reflecting significant changes in oxidative stress parameters. Compared to the blank group, the model group exhibited a marked increase in serum levels of MDA and H₂O₂ (P<0.05), confirming the successful establishment of ethanol-induced oxidative damage model. Notably, in comparison with the model group, both high-dose (H Group) and medium-dose (M Group) groups showed statistically significant reductions in oxidative stress parameters (P<0.05).

Table 7. Content of MDA and H₂O₂ in different groups ($\bar{x}\pm s$, n=6)

Group	Dose of Composite Schisandra chinensis drink (mL/kg)	Content of MDA and H ₂ O ₂	
		MDA (nmol/mL)	H ₂ O ₂ (μmol/mL)
Blank group	/	0.91±0.354	0.610±0.091
Model group	/	2.24±0.548#	1.589±0.137#
Low-dose group	20	1.48±0.389	1.082±0.203
Middle-dose group	40	1.04±0.183*	0.813±0.155*
High-dose group	60	0.60±0.176*	0.704±0.118*

#P<0.05 vs blank group; *P<0.05 vs model group

3.4 Results of anti-fatigue experiment

As shown in Tables 8 and 9, the Composite Schisandra chinensis beverage concentrate at various concentrations did not induce adverse reactions in mice. All mice exhibited liveliness, good mental status, smooth fur, and consistent body weight gain across groups, indicating that gavage had no negative impact on mouse growth under the experimental conditions. Table 10 reveals that the swimming time of the control group was significantly shorter than that of the high-, medium-, and low-dose Schisandra chinensis beverage concentrate groups.

Table 8. The different dose gavage symptoms of Composite Schisandra chinensis drink extract

Group	Diet	Appearance	Behavior	Excrement	Death
Blank group	Normal	Normal	Normal	Normal	No Death
Low-dose group	Normal	Normal	Normal	Normal	No Death
Middle-dose group	Normal	Normal	Normal	Normal	No Death
High-dose group	Normal	Normal	Normal	Normal	No Death

Table 9. The average weight changes of rats (g)

Rearing days (d)	Control group	Low-dose group	Middle-dose group	High-dose group
1	28.35	28.73	29.32	27.89
3	29.01	29.49	29.57	28.02
5	29.34	29.87	29.83	28.23
7	29.72	29.75	30.07	28.11
9	29.81	28.86	29.87	28.23
11	29.99	28.77	29.89	27.94
13	30.35	28.69	29.98	27.88
15	30.67	29.10	30.01	28.15
17	30.97	29.24	30.24	28.30
19	31.24	29.39	30.51	28.57
21	31.67	29.82	31.27	29.00

Rearing days (d)	Control group	Low-dose group	Middle-dose group	High-dose group
23	32.57	29.97	31.36	29.38
25	33.04	30.54	31.81	29.86
27	33.54	30.94	32.04	30.42
29	33.06	31.52	32.33	31.03

Table 10. The swimming time of rats (min)

Number	Control group	Low-dose group	Middle-dose group	High-dose group
1	12.02	13.11	15.11	17.41
2	13.54	14.30	16.25	17.82
3	14.31	16.24	17.35	19.28
4	16.25	16.72	25.62	31.14
5	17.95	22.64	29.17	34.58
6	24.27	25.05	32.23	42.72
7	25.41	34.39	39.94	45.27
8	36.29	38.43	46.86	47.64
9	37.36	43.73	51.15	54.55
10	46.40	47.88	54.08	60.72

3.5 Results of acute toxicity experiment

As shown in Table 11, the average body weight of mice in the acute toxicity experiment increased steadily over time. Although transient weight loss was observed in a few individuals, all mice ultimately demonstrated healthy growth, indicating that gavage had no adverse effects. These results confirm that the Composite Schisandra chinensis beverage, formulated with Schisandra chinensis, Lycium barbarum and Ophiopogon japonicus, and excipients such as xylitol and citric acid, has no significant toxic or side effects.

Table 11. The average weight changes of rats in acute toxicity test (g)

	Gender	Saline	Accessory	Low-dose group	Middle-dose group	High-dose group
Pre-administration	Female	25.42	25.68	26.11	26.25	26.58
	Male	25.71	26.02	26.26	26.02	26.75
Post-administration 1 d	Female	27.00	28.16	27.63	27.82	27.40
	Male	27.45	27.79	28.27	27.79	27.42
Post-administration 3 d	Female	28.12	28.55	28.73	28.80	28.51
	Male	29.67	29.46	29.61	29.75	28.94
Post-administration 5 d	Female	30.52	30.93	30.44	30.17	31.06
	Male	30.84	31.33	30.37	30.74	31.63
Post-administration 7 d	Female	31.01	32.09	32.89	31.68	32.14
	Male	31.94	32.16	31.64	32.04	33.22
Post-administration 9 d	Female	33.27	33.71	33.86	33.06	33.67
	Male	34.53	34.73	33.97	34.21	34.49
Post-administration 11 d	Female	35.66	35.89	35.14	34.51	34.86
	Male	36.51	36.60	35.94	35.88	37.07
Post-administration 13 d	Female	36.92	36.96	36.13	35.35	35.84
	Male	37.60	38.04	37.87	37.94	37.62
Post-administration 15 d	Female	38.34	38.33	37.54	36.87	38.17
	Male	39.16	39.54	39.26	39.74	40.04

3.6 Results of immune enhancement index measurement

As shown in Table 12, the organ index measurement results revealed significant changes in the immunocompromised

model group compared to the blank control group. Specifically, the spleen index increased significantly ($P<0.05$), while the thymus index decreased significantly ($P<0.05$). Following administration of the Composite Schisandra chinensis beverage at different doses, the high-dose and medium-dose groups exhibited a significant reduction in spleen index and a significant increase in thymus index ($P<0.05$). The ear swelling difference measurement results showed that the model group had a significantly reduced ear swelling difference compared to the blank control group ($P<0.05$). However, after treatment with the Composite Schisandra chinensis beverage, the ear swelling differences in the H, M, and L groups were significantly increased compared to the model group ($P<0.05$).

Table 12. Content of organ index and the difference of ear swelling in different groups ($\bar{x}\pm s$, $n=6$)

Group	Content of organ index and the difference of ear swelling		
	Splenic index (mg/g)	Thymic index (mg/g)	Difference of ear swelling (mg)
Blank group	1.74±0.203	2.86±0.152	19.6±0.95
Model group	3.18±0.175#	1.95±0.327#	12.9±0.88#
Low-dose group	2.87±0.240	2.29±0.376	15.1±1.28*
Middle-dose group	2.70±0.175*	2.58±0.194*	16.3±1.39*
High-dose group	2.52±0.265*	2.74±0.315*	17.4±1.46*

$P<0.05$ vs blank group; * $P<0.05$ vs model group

4. Discussion

Schisandra chinensis, characterized by its complex flavor profile of sweet, sour, spicy, and bitter, exhibits an overall sour and astringent taste[16]. The concentrated extract of its Composite formulation is unsuitable for direct consumption. However, through orthogonal experiment optimization, the Composite Schisandra chinensis beverage achieved a balanced sweet-sour taste, bright and translucent color, and a pure yet robust aroma, making it suitable for public consumption. The optimal formulation, determined to be 30% Composite Schisandra chinensis extract, 8% xylitol, and 0.010% citric acid, demonstrated stable and reliable results across three validation experiments.

MDA content serves as a key indicator of antioxidant capacity, while H_2O_2 , a by-product of cellular metabolism, reflects reduced antioxidant activity in vivo. The spleen and thymus, as critical immune organs[17], provide essential insights into immune function through their indices and structural integrity. Animal experiments revealed that 30 days of continuous gavage with the Composite Schisandra chinensis beverage significantly reduced the levels of oxidative products (MDA and H_2O_2) induced by modeling, thereby mitigating oxidative damage caused by free radicals. Furthermore, the beverage extended the swimming time of mice, indicating notable anti-fatigue effects that were more pronounced at higher concentrations. Additionally, the beverage significantly increased ear swelling differences, inhibited immune organ damage, and enhanced overall immune function in mice. While high concentrations of the extract induced sluggish behavior and dull fur in some mice, likely due to gastric irritation affecting normal physiological activities, these effects were transient and non-lethal.

According to the Chinese Pharmacopoeia (ChP2020; Volume I), the recommended daily adult dosage of Schisandra chinensis ranges from 1.5 g to 6.0 g. In this study, the concentrations of Schisandra chinensis extract administered to mice were 100, 200 and 400 times higher than those in normal beverages, with no observed toxic side effects. This confirms the safety of the Composite Schisandra chinensis beverage for consumption. In conclusion, this study provides a viable approach for the development and utilization of functional beverages derived from “medicine and food homology” Schisandra chinensis, significantly enhancing its commercial value and practical applications.

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References

- [1] Sun P, Wei HQ, Zhuang CX, et al. Minor dibenzocyclooctadiene lignans with antifibrotic potential from the fruits of Schisandra chinensis. *Phytochemistry* 2025;235:114458.
- [2] Wang YS, Li MM, Sun Y, et al. Volatile oils of Schisandra chinensis (Turcz.) Baill alleviates Parkinson's disease by activating the Nrf2 pathway to positively regulate autophagy and oxidative stress. *Bioorg Chem* 2025;157:108277.
- [3] Rachalewski M, Piwko P M, Dębowska R, et al. Biocompatibility and Post-Marketing Surveillance Study of Emollient

- Plus Medical Device Cream Containing Oligofructans from *Ophiopogon japonicus* and Acetyl Heptapeptide-4 in Atopic Dermatitis Skin Care. *Cosmetics* 2024;11(4): 136-136.
- [4] Orsi CB, Gorski D, Krul EN, et al. The effects of nutritional supplementation for children and adolescents with sickle cell disease: A systematic review and meta-analyses. *Clin Nutr* 2025;47:157-168.
- [5] Kamali A, Hosseini H, Mahmoudi R, et al. The sensory evaluation and antimicrobial efficacy of *Lactobacillus acidophilus* supernatant on *Salmonella enteritidis* in milk. *Food Sci Nutr* 2023;12(3):1902-1910.
- [6] Senatore G, Virgili F, Blandini L. Global optimal actuator placement for adaptive structures: New formulation and benchmarking. *J Intel Mat Syst Str* 2025;36(3):151-173.
- [7] Bijesh R, Arun P, Muraleedharan C. Modified stoichiometric equilibrium model for sewage sludge gasification and its validation based on experiments in a downdraft gasifier. *Biomass Convers Bior* 2021;13(10):9023-9043.
- [8] Rafał W, Beata D, Dorota D, et al. Verification of the conditions for determination of antioxidant activity by ABTS and DPPH Assays-A Practical Approach. *Molecules* 2021;27(1):50-50.
- [9] Nebia B, Mokhtaria K. Antioxidant and Antimicrobial Activities of Essential Oil of *Satureja calamintha* ssp. *nepeta* (L.) Briq. from the Northwest of Algeria. *Agric Conspect Sci* 2021;86(4):349-356.
- [10] Li C, Li L, Cheng J, et al. Anti-fatigue effect of *Lateolabrax japonicus* peptides in mice and the underlying action mechanism via in vitro and in vivo assays. *Food Biosci* 2024; 58. DOI:10.1016/j.fbio.2024.103763.
- [11] Hao H, Sha A. Study on Anti-fatigue Effects and Mechanisms of Polysaccharide from *Paris polyphylla*. *Dokl Biochem Biophys* 2024;516(1):58-65.
- [12] Liu Y, Liu F, Wang C. Acute Toxicity, Neurotoxic, Immunotoxic, and Behavioral Effects of Deltamethrin and Sulfamethoxazole in Adult Zebrafish: Insights into Chemical Interactions and Environmental Implications. *Toxics* 2025;13(2):128-128.
- [13] Wu Y, Yu W, Song Z, et al. The Acute Toxicity and Cardiotoxic Effects of Levofloxacin on Zebrafish (*Danio rerio*). *Toxics*, 2025;13(2):122-122.
- [14] Santos DLAC, Moreira TMA, Silva LDRJ, et al. Evaluation of the effect of 2-borneol on the *Nauphoeta cinerea* model through oxidative stress and acute toxicity tests. *Nat Prod Res* 2025;1-9.
- [15] Tianbing W, Bang C, Tao M, et al. Identification and immunoprofiling of key prognostic genes in the tumor microenvironment of hepatocellular carcinoma. *Bioengineered* 2021;12(1):1555-1575.
- [16] Gesine BLSE, Timsse R, et al. Disrupting Roquin-1 interaction with Regnase-1 induces autoimmunity and enhances antitumor responses. *Nat Immunol* 2021;22(12):1563-1576.
- [17] Pharmacopoeia of People's Republic of China Part 1. Committee National Pharmacopoeia 2020.

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