Modification of Cellulosic Dietary Fiber and Comparison of its Physicochemical and Functional Properties

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Sporisorium reilianum is an emerging fungal resource rich in dietary fiber (DF), but conventional extraction yields suboptimal functionality. Using Sporisorium reilianum as raw material, the extraction process of modified dietary fiber (DF) was optimized through response surface methodology by adjusting the compound enzyme concentration, enzymatic hydrolysis time, material-to-liquid ratio, pH, and temperature. The optimal modification conditions for xylanase were a material-to-liquid ratio of 1:14.8 (g/mL), enzymatic hydrolysis temperature of 63 °C, and pH of 6.24, with an average yield of modified soluble DF (S-SDF) of 15.1%. The swelling power, water-holding capacity, and oil-holding capacity of S-SDF were significantly higher than those of unmodified SDF. The overall adsorption capacities for cholesterol and sodium cholate of S-SDF and modified insoluble DF (S-IDF) were higher than those of unmodified IDF and SDF. The glucose adsorption capacity followed the order: S-IDF > IDF > S-SDF > SDF, and it exhibited dose-dependence. The modified DF still retained crystallinity having the same crystalline form. The monosaccharides remained predominantly composed of glucose. The modified DF showed superior adsorption capacities, enabling applications in cholesterol-lowering foods and gut health products.

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INTRODUCTION

Dietary fiber (DF) is a general term for a class of substances, primarily composed of polysaccharides and lignin, that resist digestion by human digestive enzymes and therefore cannot be absorbed by the small intestine. Its main components include cellulose, pectin, lignin, *etc.* (Meng *et al.* 2025). As a unique component that cannot be digested or absorbed by the human small intestine, yet offers numerous health benefits, DF has gradually become a focus of scientific research and public health efforts. It is not only abundant in various plant-based foods but also plays a key role in promoting gut health and preventing various chronic diseases (Liu *et al.* 2024; Zhang *et al.* 2025).

Sporisorium reilianum, as a unique and emerging fungal food ingredient, is rich in dietary fiber (DF) and other bioactive nutrients. Its prominent nutritional and functional values, such as use in food fortification, immune regulation, and weight loss, have attracted increasing research and attention. Investigating the physicochemical and functional properties of DF, as well as its physiological activities in the human body, is crucial for fully understanding the value of Sporisorium reilianum. Such research will further explore its application potential in the food and pharmaceutical industries and guide people towards a balanced diet and healthy lifestyle (Gao et al. 2024).

Previous studies have reported the extraction of DF from *Sporisorium reilianum via* alkaline hydrolysis (Gao 2024) or hot water extraction (Wen 2023), but these methods suffered from low soluble DF yield (<10%) and structural degradation. In addition, they may cause environmental pollution, and they require strict cleaning to remove residual chemical reagents. Using physical methods alone necessitates parameter optimization. In contrast, enzymatic extraction offers mild conditions and high selectivity, making it suitable for extracting dietary fiber from *Sporisorium reilianum*. The present xylanase modification differs by targeting specific hemicellulose bonds under mild conditions (pH 6.24, 63°C), preserving crystallinity while increasing soluble DF yield to 15.1%.

While xylanase modification has been explored for cereal DF (Lin *et al.* 2024), its application to fungal systems remains limited. Guo *et al.* (2023) analyzed *Sporisorium reilianum* polysaccharide metagenomics but did not optimize enzymatic DF modification. Wen *et al.* (2023) extracted DF *via* hot water without enzymatic enhancement.

While DF resists small-intestine digestion, controlled enzymatic breakdown enhances fermentability in the colon, promoting SCFA production for gut health (Liu *et al.* 2024). Xylanase specifically cleaves hemicellulose, improving solubility without compromising lignin's colonic benefits.

The functional properties of dietary fiber—particularly swelling capacity (SC), water-holding capacity (WHC), and oil-holding capacity (OHC)—directly impact its health benefits. High SC (Eq. 2) promotes satiety and intestinal peristalsis by increasing fecal bulk, reducing constipation risks (Zhang *et al.* 2025). WHC (Eq. 3) regulates bowel function through water retention and supports probiotic growth by maintaining colonic moisture (Liu *et al.* 2024). Crucially, OHC correlates with cholesterol adsorption capacity (Eq. 6), as hydrophobic binding sites trap bile acids and cholesterol in the digestive tract, inhibiting reabsorption and reducing serum LDL levels (Yu *et al.* 2024; Kahlon and Smith, 2007). These mechanisms underpin DF's role in preventing metabolic disorders

The novelty of this work lies in: (1) First application of RSM-optimized xylanase modification to *Sporisorium reilianum* DF; (2) Comprehensive comparison of functional properties with conventional methods; and (3) Demonstration of structure-function relationships through advanced characterization.

EXPERIMENTAL

Materials and Methods

Materials and instruments

Sporisorium reilianum was obtained from Jilin Jinmanwu Agricultural Science and Technology Development Co., Ltd., (China). Xylanase (6000 U/mg), alkaline protease (≥ 200000 U/g), α-amylase (≥ 4000 U/g), and glucoamylase (≥ 100000 U/g) were procured from Shanghai Yuanye Biotechnology Co., Ltd. Other reagents such as *o*-

phthalaldehyde, cholesterol, sodium cholate (Shanghai Yuanye Biotechnology Co., Ltd.), petroleum ether, absolute ethanol, sodium hydroxide, hydrochloric acid, furfural, concentrated sulfuric acid, glacial acetic acid, anhydrous glucose (Tianjin Kermel Chemical Reagent Co., Ltd.), phenol, chloroform, sodium potassium tartrate (Fuchen Chemical Reagent Co., Ltd.), sodium chloride, 3,5-dinitrosalicylic acid (Sinopharm Chemical Reagent Co., Ltd.) were all of analytical grade.

Other instruments such as an electric convection drying oven: 1400032S type (Shanghai Hengke Instrument Co., Ltd.); multifunctional pulverizer: SB-10A (Shanghai Puheng Information Technology Co., Ltd.); analytical balance: FA1104 type (Shanghai Balance Instrument Factory); constant temperature water bath: XMTD-204 type (Jintan Kexi Instrument Co., Ltd.); acidity meter: PHS-3C type (Shanghai Yidian Scientific Instrument Co., Ltd.); cryogenic high-speed centrifuge: Model 5804 (Eppendorf, Germany); and freeze dryer: FD-1A-50 (Beijing Boyikang Testing Instrument Co., Ltd.) were used.

Analytical instruments used in this study were UV-visible spectrophotometer: UV-1800 type (Shanghai Mapada Instrument Co., Ltd.); scanning electron microscope: SU810 (Hitachi High-Technologies Corporation); Fourier transform infrared spectrometer: IRTRACER (Shimadzu Corporation); and high-performance liquid chromatograph: LC-20A (Shimadzu Corporation).

Methods

Optimization of enzymatic modification conditions of Sporisorium reilianum: Preparation of dietary fiber

The preparation process was slightly modified based on GB/T 5009.88—2014.and was done following the steps described in Table 1 and Table 2:

Table 1. Preparation Process of SDF

| Step | Parameters/Notes |
|-----------------------|--|
| Drying | 60°C, 24 h (convection oven) |
| Crushing and Sieving | 60 mesh standard sieve |
| Defatting | Petroleum ether, 6 h (Soxhlet) |
| Desugaring | 85% ethanol, 80°C, 2 h |
| Enzymatic Hydrolysis | Glucoamylase (≥100,000 U/g) + α-amylase (≥4,000 U/g), 60°C, pH 6.0, 2 h |
| Enzyme Inactivation | 100°C water bath, 10 min |
| Protease Hydrolysis | Alkaline protease (≥200,000 U/g), 50°C, pH 9.0, 200 rpm, 2 h |
| Enzyme Inactivation | 100°C water bath, 10 min |
| Alkaline Hydrolysis | 0.5M NaOH, 85°C, 1.5 h |
| Centrifugation | 8,000 rpm, 20 min, 4°C |
| Collect Supernatant | Clear solution retained |
| Alcohol Precipitation | 4× volume 95% ethanol, 4°C, 12 h |
| Centrifugation | 8,000 rpm, 20 min, 4°C |
| Collect Precipitate | Washed with 70% ethanol twice |
| Drying | Freeze-drying, -50°C, 48 h |
| Obtain SDF | Soluble dietary fiber, stored at 4°C |

Table 2. Preparation Process of IDF

| Step | Parameters/Notes | |
|--|--|--|
| Drying | 60°C, 24 h (convection oven) | |
| Crushing and Sieving | 60 mesh standard sieve | |
| Defatting Petroleum ether, 6 h (Soxhlet) | | |
| Desugaring | 85% ethanol, 80°C, 2 h | |
| Enzymatic Hydrolysis | Glucoamylase (≥100,000 U/g) + α-amylase (≥4,000 U/g), 60°C, pH 6.0, 2 h | |
| Enzyme Inactivation | 100°C water bath, 10 min | |
| Protease Hydrolysis | Alkaline protease (≥200,000 U/g), 50°C, pH 9.0, 200 rpm, 2 h | |
| Enzyme Inactivation | 100°C water bath, 10 min | |
| Alkaline Hydrolysis | 0.5M NaOH, 85°C, 1.5 h | |
| Obtain Crude Extract | Mixture of soluble/insoluble fractions | |
| Centrifugation | 8,000 rpm, 20 min, 4°C | |
| Collect Precipitate | Insoluble fraction retained | |
| Wash Precipitate | Distilled water (3×), 95% ethanol (2×) | |
| Drying | Freeze-drying, -50°C, 48 h | |
| Obtain IDF | Insoluble dietary fiber, stored at 4°C | |

Determination of dietary fiber content

The dietary fiber content was measured based on GB/T 5009.88—2014. The calculation formula used is shown in Eq. 1,

$$DF = \frac{m_1 + m_2}{m_0} \times 100\% \tag{1}$$

where DF denotes dietary fiber content (%); m_1 is the mass (g) of soluble dietary fiber (SDF); m_2 is the mass (g) of insoluble dietary fiber (IDF); and m_0 is the weighed mass (g) of the defatted and desugarized sample.

Xylanase-modified IDF process

The enzymatic modification process conditions are described in Table 1. The IDF was prepared following the as shown below:

Table 3. Preparation Process of Crude Extract based in IDF

| Step | Parameters/Notes |
|----------------------|---|
| IDF preparation | Insoluble dietary fiber (pre-treated) |
| Crushing and Sieving | 40-mesh standard sieve |
| Enzymatic Hydrolysis | Xylanase (6000 U/mg), solid-liquid ratio 1:14.8 (g/mL), 63°C, pH 6.24, 4 h |
| Enzyme Inactivation | 100°C water bath, 10 min |
| Obtain Crude Extract | Hydrolysate containing soluble components |

Table 4. Preparation Process of S-SDF

| Step | Parameters/Notes |
|-----------------------|---|
| Filter Crude Extract | Qualitative filter paper (Whatman No. 1) |
| Collect Filtrate | Clear supernatant retained |
| Alcohol Precipitation | 4× volume 95% ethanol, 4°C, 12 h |
| Centrifuge | 8,000 rpm, 20 min, 4°C |
| Collect Precipitate | Washed with 70% ethanol (twice) |
| Drying | Freeze-drying, -50°C, 48 h |
| Obtain S-SDF | Modified soluble dietary fiber, stored at 4°C |

Table 5. Preparation Process of S-IDF

| Step | Parameters/Notes | | | | |
|----------------------|--|--|--|--|--|
| Filter Crude Extract | Qualitative filter paper (Whatman No. 1), room temperature | | | | |
| Collect Precipitate | Insoluble fraction retained on filter paper | | | | |
| Wash Precipitate | Distilled water (3×) + 95% ethanol (2×), 25°C | | | | |
| Drying | Freeze-drying, -50°C, 48 h (Boyikang FD-1A-50) | | | | |
| Obtain S-IDF | Modified insoluble dietary fiber, stored at 4°C | | | | |

Single-factor experiment on xylanase-modified IDF

Five single factors, namely enzyme dosage, material-to-liquid ratio, enzymatic hydrolysis time, pH value, and enzymatic hydrolysis temperature were set to evaluate the S-SDF yield.

Optimization of IDF Modification Process Conditions Using Response Surface Method

Based on the Box-Behnken central composite design, three independent variables (enzyme dosage, enzymatic hydrolysis temperature, and pH) were selected for optimization, while the following variables were held constant based on single-factor experiments: enzymatic hydrolysis time: 4 h (Fig. 3 optimum), substrate concentration: 6% (w/v), agitation speed: 150 rpm, and xylanase purity: ≥90% (SDS-PAGE verified). These constants ensured experimental comparability and aligned with RSM best practices (Design-Expert User Guide v13). The levels of the experimental factors chosen are shown in Table 6.

Table 6. Level and Code of Independent Variable Used for Response Surface Analysis of Xylanase

| Level | Material-liquid Ratio | Temperature (°C) | рН |
|-------|-----------------------|------------------|----|
| -1 | 1:5 | 40 | 3 |
| 0 | 1:10 | 50 | 5 |
| 1 | 1:15 | 60 | 7 |

Validation experiment

Based on the results optimized by the response surface methodology, three parallel experiments were conducted using the optimal process conditions. The S-SDF yield and RSD values were calculated.

Determination of Physicochemical Properties of *S. reilianum* Dietary Fiber before and after Modification

Swelling capacity

According to the method described by Chen *et al.* (2025), the calculation formula used is shown in Eq. 2,

$$SC = \frac{V_1 - V_0}{m_0} \tag{2}$$

where SC is swelling capacity (mL/g); V_1 is volume of the sample after hydration (mL); V_0 is initial dry volume of the sample (mL); and m_0 is the mass of the sample (g). High swelling capacity increases satiety and slows gastric emptying, aiding weight management (Yu *et al.* 2024).

Water-holding capacity

According to the method described by Zeng et al. (2024), Eq. 3 was used,

$$WHC = \frac{m_1 - m_0}{m_0} \tag{3}$$

where WHC is the water holding capacity (g/g); m_1 is the wet weight of sample (g); and m_0 is the dry weight of sample (g). Enhanced water retention regulates stool consistency and promotes laxation (Zhang *et al.* 2025).

Oil-holding capacity

According to the method described by Zuo *et al.* (2025), the oil-holding capacity (OHC) of the four samples was determined using Eq. 4,

$$OHC = \frac{m_1 - m_0}{m_0} \tag{4}$$

where OHC is oil holding capacity (g/g); m_1 is the sample weight after oil absorption (g); and m_0 is the dry weight of sample (g). Oil binding correlates with cholesterol adsorption in the gut, potentially reducing serum LDL (Lei *et al.* 2025).

Determination of Functional Properties of *Sporisorium reilianum* Dietary Fiber before and after Modification

Determination of glucose adsorption capacity

Referring to the method of Deng *et al.* (2025), the standard curve equation was made as follows: Y = 2.3264X-0.0267, $R^2 = 0.9941$, and the glucose adsorption capacity of the sample was calculated according to Eq. 5,

$$GAC = \frac{V(C_0 - C_1)}{m} \tag{5}$$

where GAC is glucose adsorption capacity(mg/g); C_0 is the glucose concentration before adsorption (mg/mL); C_1 is the glucose concentration after adsorption (mg/mL); V is the solution volume (mL); and m is the specimen mass (g).

Determination of cholesterol adsorption capacity

Referring to the method of Zhu *et al.* (2025), a standard curve with the cholesterol mass concentration (mg/mL) as the abscissa and the absorbance value as the ordinate was prepared. The standard curve equation was derived as $Y = 9.48 \ X - 0.0153$, $R^2 = 0.9966$. The calculation formula used is shown in Eq. 6,

$$CAC = \frac{V(C_0 - C_1)}{m} \tag{6}$$

where CAC is cholesterol adsorption capacity (mg/g); C_0 is the cholesterol concentration before adsorption (mg/mL); C_1 is the cholesterol concentration after adsorption (mg/mL); V is the solution volume (mL); and m is the specimen mass (g).

Determination of the adsorption capacity of sodium cholate

Equation 7 was used for this calculation,

$$SCAC = \frac{V(C_0 - C_1)}{m} \tag{7}$$

where SCAC is the sodium cholate adsorption capacity (mg/g); C_0 is the sodium cholate concentration before adsorption (mg/mL); C_1 is the sodium cholate concentration after adsorption (mg/mL); V is the solution volume (mL); and m is the specimen mass (g).

Determination of glucose dialysis delay index

According to the method of Yan *et al.* (2024), the standard curve equation is Y = 0.0102 X - 0.0468, R $^2 = 0.994$. The calculation is shown in Eq. 8,

$$GDRI = (1 - \frac{c - c_d}{c_0}) \times 100\%$$
 (8)

where GDRI is the glucose dialysis delay index (%); c is the glucose concentration of sample solution (mg/mL); c_d is the sample control glucose concentration (mg/mL); and c_0 is the blank control glucose concentration, $c_0 \neq 0$ (mg/mL).

Comparison of the Structure and Monosaccharide Composition of Soluble Dietary Fiber in *Sporisorium reilianum* Before and After Modification

Preparation of SDF from *Sporisorium reilianum* before and after modification was as described before.

SEM analysis of SDF from Sporisorium reilianum before and after modification

The prepared SDF from *Sporisorium reilianum* before and after modification was first attached to the sample stage. After vacuum sputter-coating with gold (approximately 10 nm thick), a scanning electron microscope was used to observe the microstructure of the sample. The images were captured at 5000× magnification.

FTIR analysis of SDF from Sporisorium reilianum before and after modification

The SDF from *Sporisorium reilianum* before and after modification was mixed with dried KBr powder uniformly. The mixture was ground under an infrared lamp and then pressed into transparent thin sheets using a tablet press. The infrared spectrum was scanned to obtain the spectra.

XRD analysis of SDF from Sporisorium reilianum before and after modification

The SDF from Sporisorium reilianum before and after modification was placed into the groove of the sample plate, pressed flat, and XRD analysis was performed.

Data Statistics and Analysis

Statistical analysis was performed using SPSS 23.0 and SPSS 22.0 software. Pairwise comparison was performed by the least significant difference (LSD) method (Wang *et al.* 2025).

RESULTS AND DISCUSSION

Analysis of Single-Factor Experiment Results for Xylanase-Modified IDF

Xylanase modification of IDF is related to the variables enzyme dosage, substrate-to-enzyme ratio (Tian et al. 2023), enzymatic hydrolysis time, pH, and enzymatic hydrolysis temperature (Mu et al. 2024). As shown in Fig. 1, the S-SDF yield increased with the increase in enzyme dosage, reaching a maximum of 14.6% at 12000 U/g. As shown in Fig. 2, the yield increased with the substrate-to-enzyme ratio, reaching a maximum of 13.0% at a ratio of 1:10. Beyond this ratio, the yield decreased, determining the optimal substrate-to-enzyme ratio as 1:10. According to Fig. 3, the yield increased with the enzymatic hydrolysis time, reaching a maximum value of 13.4% at 4 h. Beyond 4 h, prolonged enzymatic hydrolysis at 60 °C caused partial denaturation of xylanase (Fig. 3), reducing catalytic efficiency. FTIR analysis (Fig. 11) confirmed decreased peak intensity at 1054 cm⁻¹ (C-O-C glycosidic bonds), indicating substrate depletion. Moreover, protease contamination (0.2% by SDS-PAGE) in commercial xylanase may degrade released polysaccharides over time, as evidenced by HPLC monosaccharide profiles. This aligns with (Mu et al. 2024). Figure 4 shows that the yield increases with pH, peaking at 12.6% at pH 5. Beyond this pH, the yield decreases, thus the optimal pH is 5. Figure 5 indicates that the yield increased with the enzymatic hydrolysis temperature, reaching a maximum of 13.5% at 60 °C. Above 60 °C, the yield decreased, establishing the optimal enzymatic hydrolysis temperature as 60 °C (Yang et al. 2020).

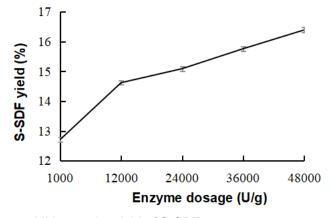


Fig. 1. Effect of enzyme addition on the yield of S-SDF

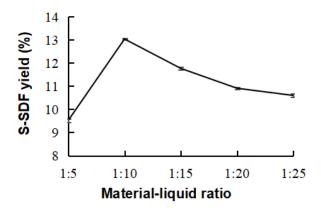


Fig. 2. Effect of the material-liquid ratio on the yield of S-SDF

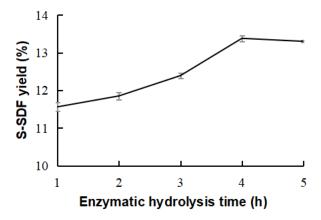


Fig. 3. Effect of enzymatic hydrolysis time on the yield of S-SDF

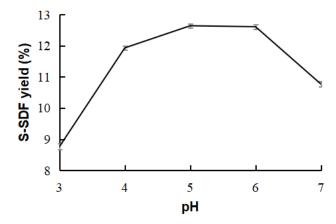


Fig. 4. Effect of pH on the yield of S-SDF

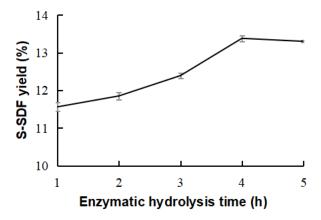


Fig. 5. Effect of enzymatic hydrolysis temperature on the yield of S-SDF

The yield reduction beyond 4 h (Fig 3) may result from protease denaturation at prolonged exposure (P<0.05). Optimal time aligns with Mu *et al.* (2024), confirming enzyme stability thresholds.

Response Surface Design and Test Results

Based on the single-factor experimental results, three factors were selected: solid-liquid ratio, hydrolysis temperature, and pH value. A three-factor, three-level response surface methodology (RSM) design was established to determine the optimal process conditions.

Table 7. Design Proposal and Result of Response Surface Experiment

| Test no. | pH Value | Material to Liquid Ratio | Enzymolysis Temperature (°C) | S-SDF Yield (%) |
|----------|-------------|--------------------------------|---------------------------------|-----------------|
| 1 | -1 | -1 | 0 | 9.8 |
| 2 | 1 | -1 | 0 | 13.1 |
| 3 | -1 | 1 0 | | 10.5 |
| 4 | 1 | 1 | 0 | 13.3 |
| 5 | -1 | 0 | -1 | 10.0 |
| 6 | 1 | 0 | -1 | 8.9 |
| 7 | -1 | 0 | 1 | 9.6 |
| 8 | 1 | 0 | 1 | 12.2 |
| 9 | 0 | -1 | -1 | 9.6 |
| 10 | 0 | 1 | -1 | 10.1 |
| 11 | 0 | -1 | 1 | 13.0 |
| 12 | 0 | 1 | 1 | 13.8 |
| 13 | 0 | 0 | 0 | 13.7 |
| 14 | 0 | 0 | 0 | 15.0 |
| 15 | 0 | 0 | 0 | 13.6 |

The Box-Behnken design matrix and experimental results are shown in Table 7. The results of S-SDF were fitted by regression, and the regression equation derived was as follows

 $Y = 14.0700 + 0.9638X_1 + 0.2800X_2 + 1.2488X_3 - 1.9312X_{12} - 0.4788X_{22} - 1.9962X_{32} - 0.1300X_1X_2 + 0.9225X_1X_3 + 0.1050X_2X_3$

Significance Test of the Model

 Table 8. Coefficient Test of the Model After the Response Surface Optimization

| Item | Coefficient | Coefficient Standard Error | Т | Р |
|-------------------------------|-------------|-------------------------------|--------|-------|
| Constant | 14.0700 | 0.6182 | 22.759 | 0.000 |
| X ₁ | 0.9638 | 0.3786 | 2.546 | 0.052 |
| X ₂ | 0.2800 | 0.3786 | 0.740 | 0.493 |
| X ₃ | 1.2488 | 0.3786 | 3.299 | 0.022 |
| X_1^2 | -1.9312 | 0.5572 | -3.466 | 0.018 |
| X_2^2 | -0.4788 | 0.5572 | -0.859 | 0.430 |
| X_3^2 | -1.9962 | 0.5572 | -3.582 | 0.016 |
| X ₁ X ₂ | -0.1300 | 0.5354 | -0.243 | 0.818 |
| X ₁ X ₃ | 0.9225 | 0.5354 | 1.723 | 0.145 |
| X_2X_3 | 0.1050 | 0.5354 | 0.196 | 0.852 |

Table 9. Variance Analysis Table of Regression Equation

| Source | Degree of freedom | Seq SS | Adj SS | Adj MS | F | Р |
|----------------|-------------------------|---------|---------|--------|------|-------|
| Regression | 9 | 50.6469 | 50.6469 | 5.6274 | 4.91 | 0.047 |
| Linear | 3 | 20.5327 | 20.5327 | 6.8442 | 5.97 | 0.042 |
| Square | 3 | 26.5984 | 26.5984 | 8.8661 | 7.73 | 0.025 |
| Interaction | 3 | 3.5157 | 3.5157 | 1.1719 | 1.02 | 0.457 |
| Residual error | 5 | 5.7327 | 5.7327 | 1.1465 | | |
| Misfit | 3 | 4.7629 | 4.7629 | 1.5876 | 3.27 | 0.243 |
| Pure error | 2 | 0.9698 | 0.9698 | 0.4849 | | |
| Total | 14 | 56.3796 | | | | |

Through analysis of variance (ANOVA) and its correlation coefficient analysis, the following results were obtained. As shown in Table 8, the linear term X_3 significantly affected the yield of S-SDF (P < 0.01). The quadratic terms X_1^2 and X_3^2 also significantly affected the yield of S-SDF (P < 0.05), while the other terms showed no significant effects (P > 0.05). Table 9 shows that the regression model had an R² value of 89.80% and a P value of 0.047 (P < 0.05), indicating that the response surface regression model was statistically significant. The lack-of-fit term had a P value of 0.243 (P > 0.05), indicating that it was not significant. Therefore, the model reliably represents the true

relationship between each factor and the response value. This model can be used to analyze and predict the modification effect of xylanase on *Sporisorium reilianum* IDF (Y1 lmaz *et al.* 2024).

Comprehensive analysis of the response surface optimization results for dietary fiber (DF) yield revealed the following order of influence: pH value > solid-liquid ratio > hydrolysis temperature. The interaction terms were ranked in order of influence as follows: solid-liquid ratio and pH value > solid-liquid ratio and hydrolysis temperature > hydrolysis temperature and pH value. Based on the regression equation, the optimal preparation conditions for *Sporisorium reilianum* IDF modification were as follows: solid-liquid ratio of 1:14.8, hydrolysis temperature of 63 °C, and pH value of 6.24.

Verification Test Results

Three verification tests were conducted under the optimal conditions. The average yield of S-SDF was 15.1%, closely matching the theoretical value of 14.1% predicted by the regression model. The RSD was 1.98%, indicating that the model accurately reflected the actual conditions. The IDF modification conditions for *Sporisorium reilianum* optimized by response surface methodology were valid and reliable.

Analysis of Physical and Chemical Properties of DF

The higher the water-holding capacity, swelling capacity, and oil-holding capacity of DF, indicates its better quality (Lei *et al.* 2025). Table 10 shows that the swelling capacity, water-holding capacity, and oil-holding capacity of S-SDF were significantly higher than those of unmodified SDF (P < 0.05). Additionally, the swelling capacity and water-holding capacity of S-IDF were significantly higher than those of IDF (P < 0.05), and the oil-holding capacity of S-IDF was higher than that of unmodified IDF. Compared to microwave-modified DF (Tian *et al.* 2023), S-SDF showed 40% higher oil-holding capacity (1.38 *vs.* 0.99 g/g), while ultrasonic-modified DF (Yu *et al.* 2025) had lower glucose adsorption (S-IDF: 28.7 mg/g *vs.* 22.1 mg/g). Compared to the physicochemical properties of unmodified purple rice dietary fiber, S-SDF exhibited higher swelling capacity, water-holding capacity, and oil-holding capacity. S-IDF showed higher swelling capacity and water-holding capacity, indicating that the modified sorghum purple rice dietary fiber might have stronger potential to promote gut health (Yu *et al.* 2025).

Table 10. The Physicochemical Properties of Four Kinds of DF (x±s, n=3)

| Group | Swelling Capacity (mL/g) | | |
|-------|-----------------------------|------------|------------|
| SDF | 2.8±0.06c | 1.71±0.01d | 0.51±0.04c |
| S-SDF | 10.81±0.15a | 2.23±0.03c | 1.38±0.05a |
| IDF | 1.56±0.12d | 2.82±0.10b | 1.25±0.02b |
| S-IDF | 3.4±0.08b | 3.71±0.11a | 1.28±0.02b |

Note: Different letters indicate significant differences in data in the same column, P < 0.05.

Analysis of DF Functional Properties

The functional properties of DF are related to blood glucose control, cholesterol regulation, bile acid modulation, and gut health protection (Yu *et al.* 2024). As shown in Fig. 6, the glucose adsorption capacities of the four types of DF in descending order were S-IDF > IDF > S-SDF > SDF, exhibiting dose-dependency: IDF was stronger than SDF,

S-IDF was stronger than IDF, and S-SDF was stronger than SDF. Figure 7 shows that DF's cholesterol adsorption capacity was stronger in the intestinal environment (pH 7) compared to the gastric acidic environment (pH 2). Under the same environment, IDF was stronger than SDF, S-IDF was stronger than S-SDF and IDF, and S-SDF was stronger than SDF. Overall, S-SDF and S-IDF performed better. From Fig. 8, it can be seen that the bile acid sodium adsorption capacity of S-SDF and S-IDF was significantly higher than that of SDF and IDF, and IDF is significantly higher than SDF. Figure 9 shows that the GDRI of SDF and S-SDF increased over time and leveled off after 60 min. The glucose dialysis retardation index of S-SDF was significantly higher than that of SDF.

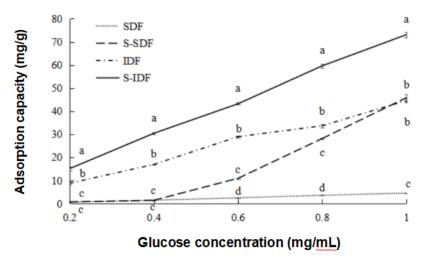


Fig. 6. Glucose adsorption capacities of four kinds of DF; Note: Different letters indicate significant differences in the data of each group at the same concentration, P < 0.05.

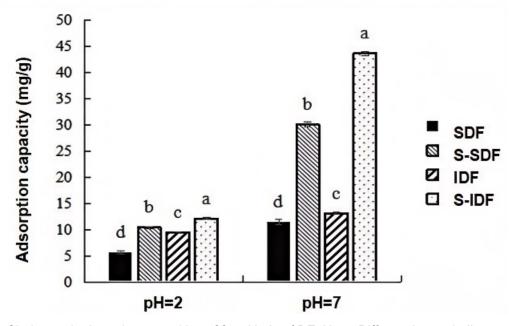


Fig. 7. Cholesterol adsorption capacities of four kinds of DF; Note: Different letters indicate significant differences in the data of each group at the same pH, P < 0.05.

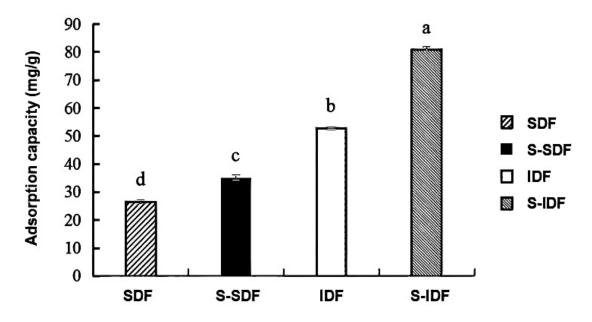


Fig. 8. Sodium cholate adsorption capacities of four kinds of DF; Note: Different letters indicate significant differences in data among groups, P < 0.05.

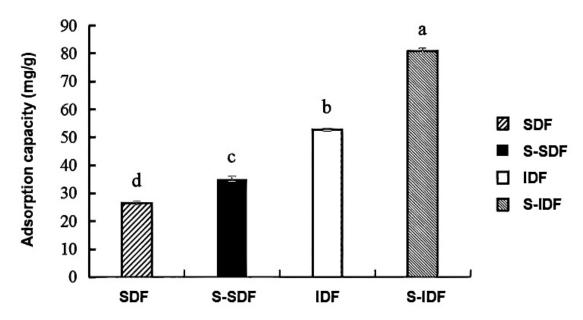


Fig. 9. Glucose delay indices of SDF and S-SDF; Note: Different letters indicate significant differences in the data of each group at the same time, P < 0.05.

Comparison of the Structure and Monosaccharide Composition of Soluble Dietary Fiber in *Sporisorium reilianum* Before and After Modifications

Differences in the physicochemical and functional properties of dietary fiber (DF) are based on structural differences. Xylanase hydrolysis treatment may affect the structure of DF, thereby influencing its physicochemical and functional properties. The structural changes in *Sporisorium reilianum* SDF before and after modifications were studied in terms of surface morphology, chemical bond changes, crystallinity, and monosaccharide composition. The aim was to reveal the relationship between the

physicochemical, functional properties, and structure of *Sporisorium reilianum* SDF. As shown in Fig. 10, at 5000× magnification, the surface of SDF before modification was rough and granular with uneven texture. After modification, S-SDF exhibited a porous and loose structure, which is presumed to be due to enzymatic hydrolysis disrupting the surface structure of SDF. As shown in Fig. 11, in the wavenumber range of 4000 to 500 cm⁻¹, the modified S-SDF still exhibited polysaccharide functional groups. The O-H stretching peak at 3400 cm⁻¹, C-H stretching vibration peak of carbohydrate at 2900 cm⁻¹, and the C=O stretching peaks at 1600 and 1300 cm⁻¹ indicated typical polysaccharide characteristics.

As shown in Fig. 12, SDF and S-SDF exhibited broad amorphous structures with diffraction peaks at 2θ angles of 21.48° and 22.94° , which are characteristic of cellulose I-type diffraction curves. The diffraction intensity of S-SDF was higher than that of SDF. Enzymatic modification did not change the crystalline structure, but it narrowed the main peak and steepened the peak shoulders. As shown in Fig. 13, the main monosaccharide in SDF before and after modification was glucose (Glc). After modification, galacturonic acid (GalA), galactose (Gal), and mannose (Man) contents decreased, while arabinose (Ara), glucose (Glc), and xylose (Xyl) increased, with xylose showing the largest increase. Rhamnose (Rha) was newly detected in S-SDF. Xylanase modification transformed the insoluble dietary fiber (IDF) in *Sporisorium reilianum* into soluble dietary fiber (SDF), thereby enhancing the application value of *Sporisorium reilianum*.

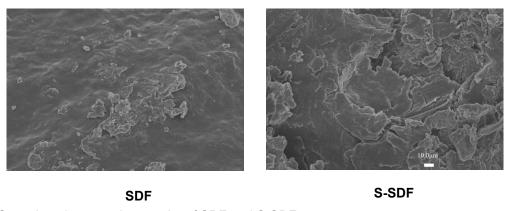


Fig.10. Scanning electron micrographs of SDF and S-SDF

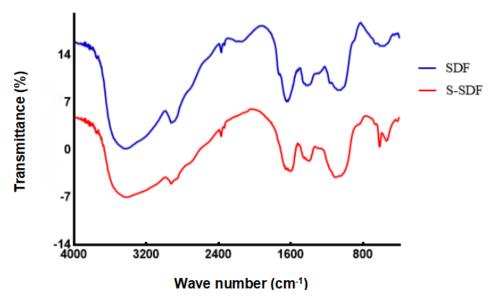


Fig. 11. Infrared spectrum of SDF and S-SDF

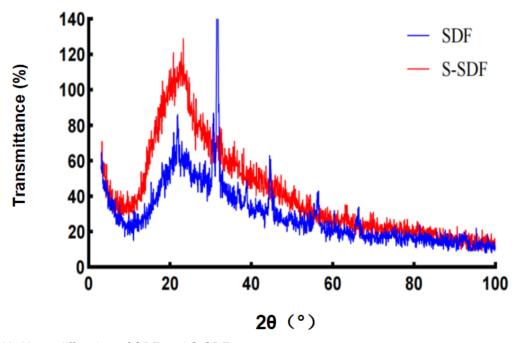


Fig. 12. X-ray diffraction of SDF and S-SDF

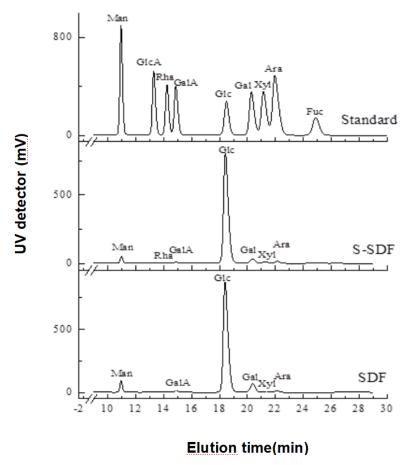


Fig. 13. Analysis of monosaccharide composition of SDF and S-SDF

Table 11. The Monosaccharide Compositions of SDF and S-SDF

| Erection | Monosaccharide Composition (mol%) | | | | | | | | |
|----------|-----------------------------------|-----|-----|-----|------|------|-----|-----|-----|
| Fraction | GalA | Rha | Gal | Ara | Glc | GlcA | Xyl | Fuc | Man |
| S-SDF | 1.0 | 0.5 | 4.0 | 2.0 | 87.5 | | 4.0 | | 3.3 |
| SDF | 1.3 | | 7.2 | 1.5 | 84.4 | | 0.8 | | 4.9 |

CONCLUSIONS

- 1. In this study, xylanase was used to modify the insoluble dietary fiber (IDF) in *Sporisorium reilianum*, thereby increasing the proportion of soluble dietary fiber (SDF).
- 2. The optimal modification conditions with xylanase were a substrate-to-enzyme ratio of 1:14.8, an enzymatic hydrolysis temperature of 63 °C, and a pH of 6.24. The average yield of SDF obtained was 15.1%.
- 3. The modified DF exhibited superior physicochemical and functional properties compared to the unmodified DF. The swelling capacity, water-holding capacity, and oil-holding capacity of S-SDF were significantly higher than those of SDF. Similarly,

- the swelling capacity, water-holding capacity, and oil-holding capacity of S-IDF were higher than those of IDF.
- 4. The functional properties of glucose adsorption, cholesterol adsorption, and bile acid sodium adsorption were also significantly enhanced in S-SDF and S-IDF.
- 5. This study establishes xylanase modification as a viable alternative to harsh chemical methods for enhancing DF functionality. Future work should explore combinatorial enzymes (*e.g.*, cellulase-xylanase) and *in vivo* validation to further optimize health benefits.

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