Investigating the Mechanism of Xiayuxue Decoction in Improving Slow Transit Constipation Based on Network Pharmacology and Animal Experiments

Fei CAI[1△], Yuxia LI[2△], Yi HUANG[1], Xinglong HUANG[1], Lingrui LOU[1], Xiaoyan LI[1], Wenbo YI[1,3*], Xinrong FAN[3*], Yudan CHEN[2], Dong GAO[2]

1. Chongqing Qijiang District Hospital of Traditional Chinese Medicine, Chongqing 401420, China; 2. College of Integrative Medicine, Fujian University of Traditional Chinese Medicine, Fuzhou 350108, China; 3. Experimental Research Center, China Academy of Chinese Medical Sciences, Beijing 100007, China

Abstract [**Objectives**] To investigating the mechanism of Xiayuxue Decoction in improving slow transit constipation (STC) based on network pharmacology and animal experiments. [**Methods**] Active components and targets of Xiayuxue Decoction (XYXD) were identified using the TCMSP, HERB, and PubChem databases, supplemented by literature mining. STC-related genes were retrieved from the GEO database. A drug—component—target—disease network was constructed, and core targets and pathways were analyzed. An STC rat model was established. After grouping and drug administration, defecation parameters, inflammatory and oxidative stress markers, and related protein expression were assessed. [**Results**] Network pharmacology identified 43 intersecting targets, primarily enriched in inflammation- and metabolism-related pathways, especially the NLRP3 inflammasome pathway. Animal experiments revealed impaired intestinal function alongside abnormal oxidative stress and inflammatory indices in the model group. Both XYXD and mosapride significantly ameliorated these abnormalities, with XYXD demonstrating a more pronounced effect. [**Conclusions**] XYXD may treat STC by inhibiting NLRP3 inflammasome-mediated chronic inflammation to alleviate colonic injury and by upregulating the 5-HT/5-HTR pathway to enhance intestinal motility.

Key words Xiayuxue Decoction (XYXD), NLRP3 inflammasome, 5-HT/5-HTR, Slow transit constipation (STC), Network pharmacology

1 Introduction

Slow transit constipation (STC) is a highly representative secondary pathological condition following spinal cord injury (SCI)[1]. As a refractory functional bowel disorder, it is clinically characterized by diminished intestinal peristalsis, significantly prolonged defecation intervals, and hardened stools, which severely compromise patients' physical and mental health as well as their quality of life. Although its pathogenesis has not yet been fully elucidated, recent studies have indicated that neurogenic inflammation plays a central role in the onset and progression of intestinal motility disorders[2-5]. SCI may trigger a local inflammatory cascade, including in the intestine, which not only directly damages the enteric nervous system (ENS), but also disrupts the synthesis and secretion of key neurotransmitters, thereby establishing a vicious cycle of "motility deficiency—inflammatory accumulation—structural remodeling.

In this pathological process, hyperactivation of the NLRP3 inflammasome is considered a critical link in intestinal immune injury[5]. Studies have shown that pro-inflammatory signals in the intestine can induce NLRP3 assembly, leading to Caspase-1 activation and subsequent cleavage of pro-IL-1β, resulting in the release of substantial amounts of mature IL-1β, which in turn exacerbates mucosal damage and disrupts the serotonin (5-HT) signaling pathway[6-9]. As the most important excitatory neurotransmitter in the gut, 5-HT coordinates intestinal motility through activation of its specific receptors, such as 5-HT3R and 5-HT4R. Therefore, inhibiting NLRP3-mediated inflammatory injury and restoring 5-HT signaling has emerged as a key therapeutic strategy for the prevention and treatment of STC.

At present, clinical treatments for STC include prokinetics, enemas, and biofeedback. Although these approaches can provide short-term symptom relief, they carry risks of drug resistance, high recurrence rates, and potential complications such as intestinal obstruction[10-11]. Traditional Chinese medicine (TCM) has accumulated extensive clinical experience in improving intestinal motility, offering unique advantages characterized by "multi-component, multi-target, and holistic regulation"[12]. Xiayuxue Decoction (XYXD), originating from Zhang Zhongjing's *Synopsis of the Golden Chamber*, is composed of Dahuang (Rhei Radix et Rhizoma), Taoren (Persicae Semen), and Tujiechong (Eupolyphaga Steleophaga). It is a classic formula known for breaking stasis, dissipating masses, and unblocking meridians. In this formula, Dahuang clears heat, expels stasis, and unblocks the bowels to relieve accumulation; Taoren promotes qi circulation and moistens the intestines; Tujiechong breaks hard masses, removes stasis, and unblocks the meridians. The combined use of these three herbs aligns with the therapeutic principles of "prolonged illness invading the collateral vessels with intestinal stasis" and "the six fu-organs should be kept unobstructed"[13-14]. Clinical studies have demonstrated that XYXD exerts significant therapeutic effects in alleviating chronic constipation[15].

Although clinical practice has revealed that XYXD can effec-

Received: August 4, 2026 Accepted: September 26, 2026
Supported by the Chongqing Natural Science Foundation (CSTB2022NSCQ-MSX1611).

△These authors contributed equally to this work.
Fei CAI, master's degree candidate, attending traditional Chinese medicine physician. Yuxia LI, master's degree candidate, assistant researcher. * Corresponding author. Wenbo YI, master's degree candidate, chief physician of traditional Chinese medicine; Xirong FAN, doctoral degree, professor.

tively alleviate STC, its specific molecular mechanisms, particularly its modulatory effects on the NLRP3 inflammatory pathway and the 5-HT/5-HTR signaling axis, remain to be elucidated. Therefore, in this study, we established a rat model of compound STC using T8 spinal cord transection combined with loperamide hydrochloride administration, and integrated network pharmacology predictions to investigate the molecular mechanisms by which XYXD improves intestinal motility. This research not only provides experimental evidence to support the clinical application of this classic formula but also deepens the theoretical understanding of the multi-target regulation of the intestine by traditional Chinese medicine.

2 Materials and methods

2.1 Animals A total of 32 adult male SPF-grade Sprague-Dawley (SD) rats, aged 8 weeks and weighing (200 ± 20) g, were purchased from Hangzhou Medical College (License No.: SCXK (Zhejiang) 2019-02). All rats were housed in the SPF barrier facility of the Laboratory Animal Center, Fujian University of Traditional Chinese Medicine (License No.: SYXK (Fujian) 2019-0007), under controlled conditions with a temperature of (25 ± 2) ℃, relative humidity of 50% – 60%, and a 12 h light/dark cycle. The rats had free access to food and water throughout the experiment. This study was approved by the Animal Ethics Committee of Fujian University of Traditional Chinese Medicine (Ethics No.: 3W2025170), and all procedures strictly adhered to the guidelines for the welfare and ethical use of laboratory animals.

2.2 Drugs XYXD was prepared using herbal granules: Dahuang granules (Batch No.: 24000791) and Taoren granules (Batch No.: 22045521) were purchased from Beijing Kangrentang Pharmaceutical Co., Ltd.; Tujiechong granules (Batch No.: 0624000151) were purchased from Gansu Foci Hongri Pharmaceutical Co., Ltd. The granules were dissolved and concentrated in ultrapure water to a final concentration of 1 g crude drug/mL and stored at 4 ℃. Mosapride citrate tablets (Yabao Pharmaceutical Group Co., Ltd., National Drug Approval No.: H20090123) were dissolved in ultrapure water to a concentration of 0.5 mg/mL. Loperamide hydrochloride (Xi'an Janssen Pharmaceutical Co., Ltd., National Drug Approval No.: H19983095) was dissolved in ultrapure water to a concentration of 0.8 mg/mL.

2.3 Reagents RIPA lysis buffer (P0013B), BCA protein assay kit (P0010S), and HRP-labeled goat anti-rabbit IgG (A0208) were purchased from Beyotime Biotechnology (Shanghai, China). Rabbit anti-β-actin (4970S) was obtained from Cell Signaling Technology. Rabbit anti-NLRP3, anti-5-HT3R, and anti-5-HT4R polyclonal antibodies were purchased from Beijing Bioss Biotechnology Co., Ltd. ELISA kits for 5-HT and IL-1β were purchased from Wuhan Elabscience Biotechnology Co., Ltd. SOD assay kit (WST-1 method) and MDA assay kit (TBA method) were purchased from Wuhan Elabscience Biotechnology Co., Ltd.

2.4 Instruments The following instruments were used: ELx800 microplate reader (BioTek, USA), Nikon 551I biological microscope (Nikon, Japan), S21-4 constant temperature water bath (Shanghai Boxun), Power Pac™ Basic electrophoresis system, Mini Trans-Blot transfer cell, and Universal Hood III gel imaging system (Bio-Rad, USA).

2.5 Identification of drug targets The constituent herbs of XYXD (Dahuang, Taoren, and Tujiechong) were searched in the PubChem database (https://pubchem.ncbi.nlm.nih.gov/). The Canonical SMILES of each component were extracted from the "Chemical Structure" section. These SMILES structures were then input into the TargetNet database (http://targetnet.scbdd.com/) for target prediction. Potential targets with a prediction probability (Prob) ≥0.9 were selected to ensure reliability. The predicted targets were integrated to obtain the potential targets of the formula.

2.6 Identification of disease targets and overlapping targets Disease targets related to "slow transit constipation" were retrieved from the GeneCards (https://www.genecards.org/) and OMIM (https://omim.org/) databases. A Venn diagram was generated to identify overlapping targets between the drug and the disease, which were considered potential nodes for XYXD's intervention in STC.

2.7 Construction of Protein-Protein Interaction (PPI) network and screening of core targets The standardized gene names of the overlapping targets were uploaded to the STRING database (https://string-db.org/) with the species set to Homo sapiens and a minimum interaction confidence score of ≥0.7. The protein interaction data, including nodes, edges, and interaction scores, were downloaded and imported into Cytoscape 3.9.1 for network visualization and analysis. Node importance was assessed based on topological parameters such as degree centrality and betweenness centrality. Nodes with higher degree values were considered more central in the network, visualized by node size and color gradient.

2.8 GO enrichment and KEGG pathway analysis Gene Ontology (GO) enrichment analysis, including biological process (BP), cellular component (CC), and molecular function (MF), was performed using R language with a significance threshold of $P<0.05$. The top 10 enriched terms in each category were selected and visualized using bubble plots. For Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis, the top 15 pathways ranked by adjusted P-value (P_{adjust}) were selected for visualization.

2.9 Establishment and evaluation of STC rat model Rats were fasted with free access to water for 12 h prior to surgery. Anesthesia was induced with 5% isoflurane and maintained with 2% isoflurane. The rats were placed in a prone position on a heated pad. A midline incision was made in the back, and the T7-T9 vertebral laminae were sequentially exposed and removed to expose the thoracic spinal cord. SCI (SCI) was induced by complete transection at the T8 level using microscissors. Hemostasis was achieved using gelatin sponges. In the control group, only laminectomy was performed without SCI. The muscles and skin were sutured, and antibiotics were applied locally to prevent infection.

Postoperatively, hindlimb twitching and tail spastic swinging were observed. After recovery from anesthesia, bilateral hindlimb flaccid paralysis was observed, and a Basso-Beattie-Bresnahan (BBB) locomotor score of 0 was considered indicative of successful model establishment. All rats received intramuscular penicillin (200 000 U) daily for seven days, along with manual bladder

evacuation (twice daily) and passive limb exercises to prevent urinary tract infections and pressure ulcers.

From day 1 after surgery, the model, XYXD, and mosapride groups received loperamide hydrochloride suspension (0.3 mg/kg, 10 mL/kg) by gavage for seven consecutive days to establish a compound STC model. The control group received an equivalent volume of normal saline.

2.10 Grouping and drug administration Thirty-two rats were randomly divided into four groups ($n=8$ per group) using a random number table: control group, model group, XYXD group (0.467 8 g/kg), and mosapride group (2.5 mg/kg). Doses were determined based on body surface area conversion between humans and rats. Drug administration began immediately after model establishment. The control and model groups received an equivalent volume of distilled water by gavage. All groups were administered a volume of 10 mL/kg once daily for 28 consecutive days.

2.11 Tissue collection, gastrointestinal motility, and defecation function assessment

2.11.1 Time to first black stool, fecal pellet count, fecal wet weight, fecal water content, and fecal consistency score. Before treatment and 30 min after the final administration, three rats from each group were randomly selected and administered activated charcoal suspension (2 mL/rat) by gavage. The time to first black stool defecation was recorded. Feces were collected before treatment and at the end of the experiment to assess fecal pellet count, wet weight, and water content. Fecal samples collected over 12 h were dried at 80 ℃ for six hours. Fecal water content was calculated using the formula: Water content (%) = (Wet weight − Dry weight)/wet weight × 100%. The consistency of the final defecation was assessed using the Bristol Stool Form Scale.

2.11.2 Tissue collection. Before sacrifice, each rat was administered 2 mL of 10% activated charcoal suspension by gavage. After 10 min, rats were anesthetized by intraperitoneal injection of 20% urethane. Blood was collected from the abdominal aorta, allowed to stand for two hours, and centrifuged at 3 500 rpm for eight minutes. The serum supernatant was collected and stored at −80 ℃. The entire small intestine from the stomach to the cecum was dissected for small intestinal propulsion rate measurement. Colon tissues were collected; portions were fixed in 4% paraformaldehyde for histopathology and immunohistochemistry (IHC), and the remaining tissues were immediately snap-frozen in liquid nitrogen and stored at −80 ℃.

2.11.3 Small intestinal propulsion rate. The small intestine was carefully separated from the mesentery and straightened. The total length of the small intestine from the pylorus to the ileocecal junction was measured, and the distance from the pylorus to the leading edge of the charcoal was measured as the propulsion distance. The small intestinal propulsion rate was calculated as: Propulsion rate (%) = (Propulsion distance/Total intestinal length) × 100%.

2.12 Histopathological analysis Colon tissues fixed in 4% paraformaldehyde were dehydrated in graded ethanol, cleared in xylene, and embedded in paraffin. The paraffin blocks were sectioned into 4-μm-thick slices, deparaffinized, stained with hematoxylin and eosin (H&E), dehydrated, mounted, and observed under a light microscope.

2.13 ELISA and oxidative stress marker detection Serum and colon tissue levels of 5-HT and IL-1β were measured using ELISA kits according to the manufacturer's instructions. Serum superoxide dismutase (SOD) activity was measured using the WST-1 method, and malondialdehyde (MDA) levels were measured using the TBA method.

2.14 Western blot analysis Colon tissues were homogenized in RIPA lysis buffer and centrifuged at 12 000 rpm for 10 min at 4 ℃. The supernatant was collected, and protein concentration was determined using the BCA method. Protein samples were denatured, separated by SDS-PAGE, and transferred onto PVDF membranes. Membranes were blocked with 5% non-fat milk for two hours, then incubated overnight at 4 ℃ with primary antibodies against 5-HT3R (1 ∶ 1 000), 5-HT4R (1 ∶ 2 000), NLRP3 (1 ∶ 2 000), and β-actin (1 ∶ 50 000). After washing with TBST, membranes were incubated with HRP-conjugated secondary antibodies for one hour. Protein bands were visualized using chemiluminescence, and densitometric analysis was performed using ImageJ software. The relative expression of target proteins was normalized to β-actin.

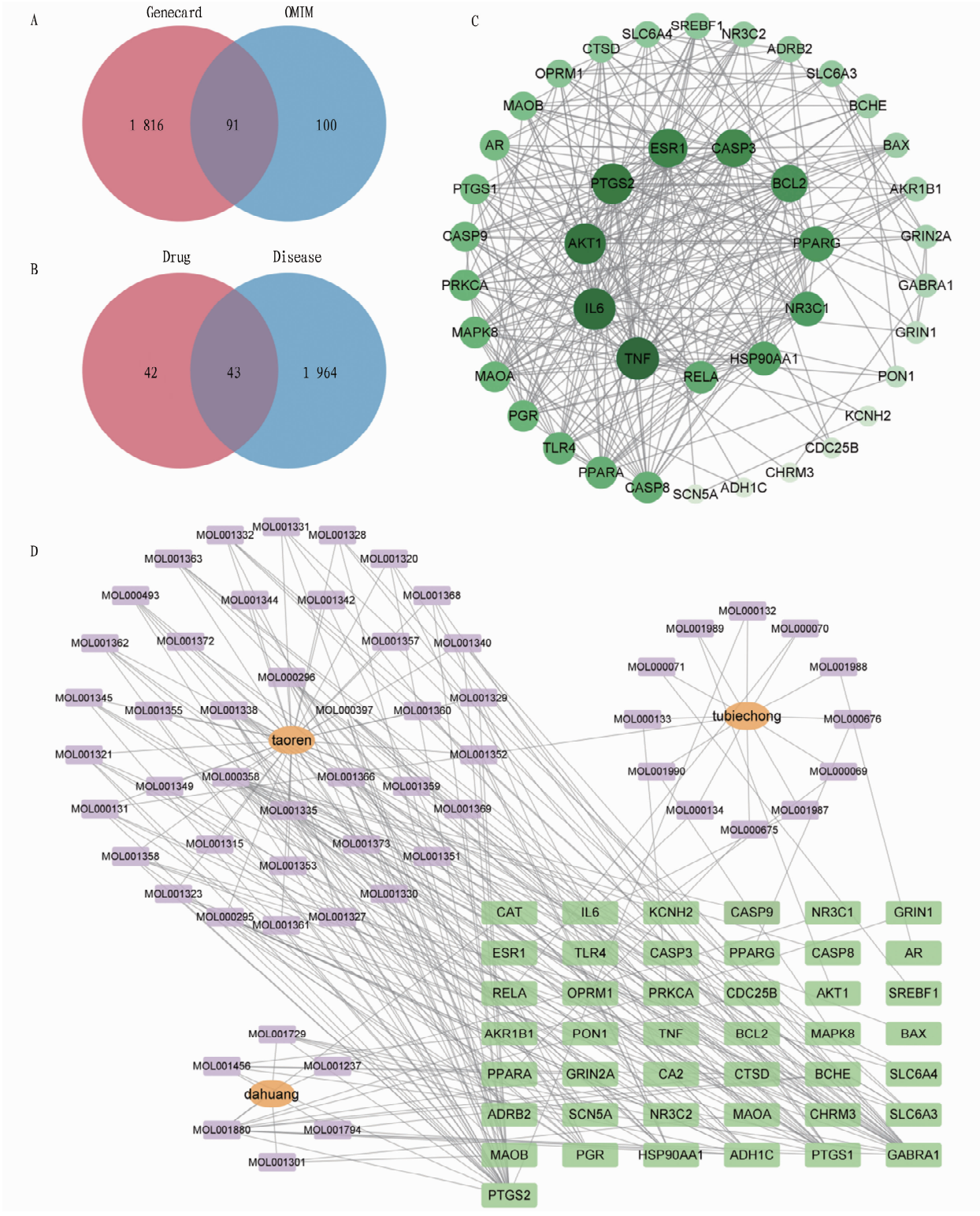
2.15 Statistical analysis Statistical analysis was performed using SPSS 24.0 software. Data were expressed as mean ± standard deviation ($\bar{x} \pm s$). Normality and homogeneity of variance were tested. For data meeting normal distribution and homogeneity of variance, comparisons between two groups were performed using unpaired t test, comparisons among multiple groups were performed using one-way analysis of variance (ANOVA), and post hoc pairwise comparisons were performed using Student-Newman-Keuls (SNK-Q) test. A P-value <0.05 was considered statistically significant.

3 Results and analysis

3.1 Screening of potential targets of XYXD for the treatment of STC Following retrieval from the GeneCards and OMIM databases and removal of duplicates, a total of 2 007 STC-related pathogenic targets were identified. Concurrently, active components and their corresponding predicted targets for the three herbs in XYXD were obtained from the TCMSP and TargetNet databases, yielding 85 targets. By mapping drug targets against disease targets, 43 overlapping targets were ultimately identified (Fig. 1A − B). These intersecting targets were considered the core biological nodes mediating the therapeutic effects of XYXD in STC. A "drug—component—core target—disease" interaction network was constructed using Cytoscape 3.10.2 (Fig. 1D), preliminarily revealing the multi-component, multi-target synergistic characteristics of XYXD.

3.2 PPI network construction and identification of key nodes To elucidate the protein-protein interactions involved in XYXD intervention in STC, the 43 overlapping targets were imported into the STRING database (confidence score ≥ 0.4). After excluding two isolated nodes, a PPI network comprising 41 protein nodes was constructed (Fig. 1E). Topological analysis was performed using Cytoscape, and nodes were ranked according to de-

gree centrality. The results showed that hub targets with high connectivity were enriched in pathways related to inflammation regulation and cellular stress, including KCNH2, GRIN1, SCN5A, GABRA1, CHRM3, and PRKCA, suggesting that XYXD may intervene in the pathological progression of STC by modulating these key proteins.

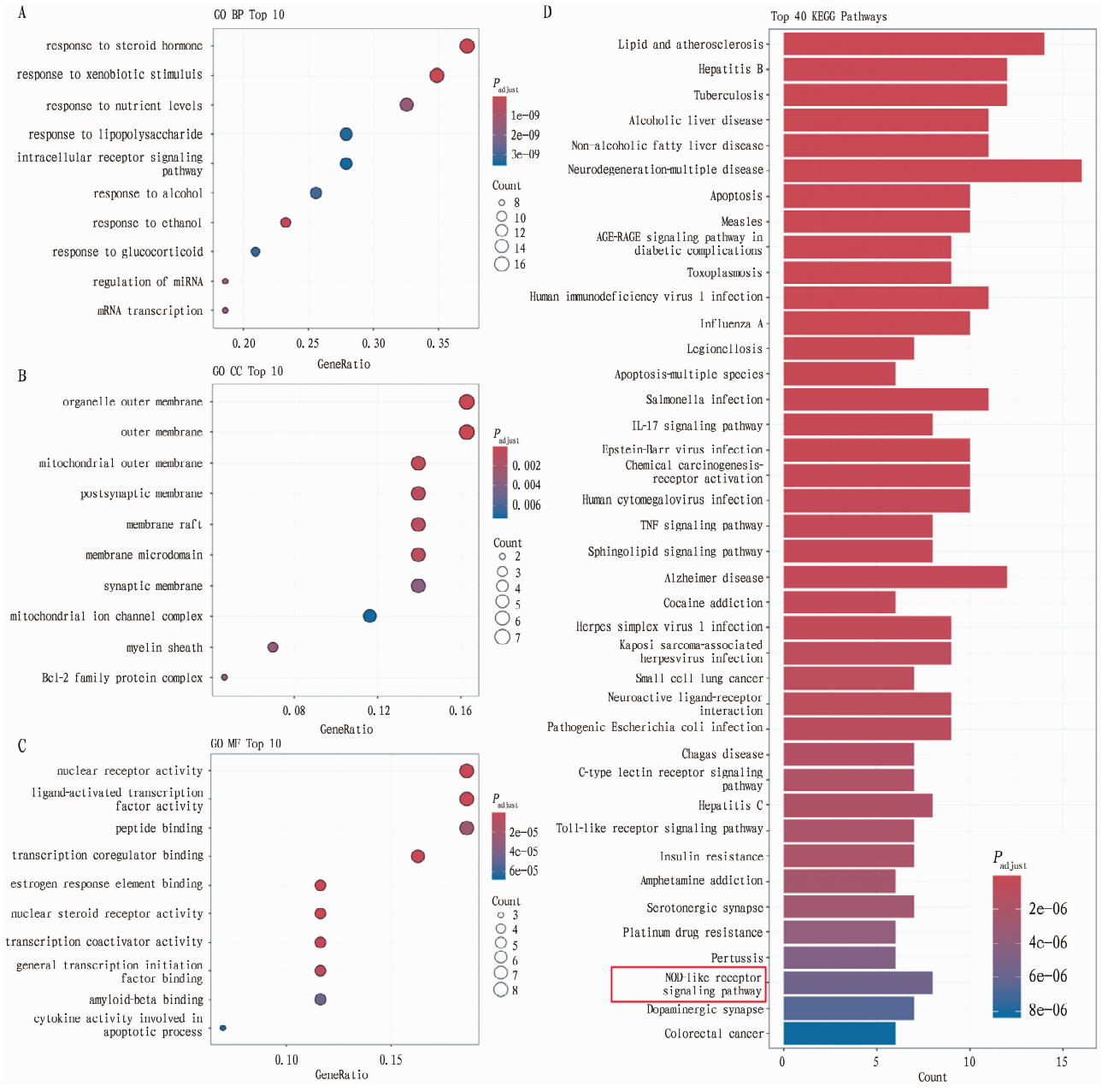


NOTE A – B. Venn diagrams of XYXD targets and STC disease-related genes; C. Protein-protein interaction (PPI) network analysis of XYXD-STC; D. "Herb—active component—target" network analysis.

Fig. 1 Bioinformatics analysis of key targets and molecular networks of XYXD in the treatment of STC

3.3 GO and KEGG enrichment analysis GO functional enrichment analysis was performed on the 41 overlapping targets. Biological process (BP) terms were mainly enriched in steroid response, lipid metabolism regulation, oxidative stress, and miRNA transcription regulation. Cellular component (CC) terms were significantly enriched in the mitochondrial outer membrane, synaptic membrane, and membrane microdomains (Fig. 2A – B). Molecular function (MF) terms were primarily associated with nuclear receptor activity, protein kinase regulation, and transcription factor binding (Fig. 2C).

KEGG pathway analysis (Fig. 2D) revealed that the core targets were significantly enriched in pathways such as lipid metabolism and atherosclerosis, the IL-17 signaling pathway, the TNF signaling pathway, and the NOD-like receptor signaling pathway. Notably, the enrichment of the NOD-like receptor pathway directly implicates the NLRP3 inflammasome, a key initiator of inflammatory responses. Combined with the GO analysis results, which highlighted oxidative stress, mitochondrial stress, and membrane structural damage, these findings preliminarily suggest that XYXD may alleviate colonic tissue immune injury by inhibiting NLRP3-mediated inflammatory responses. This bioinformatics prediction provides a clear direction for subsequent *in vivo* experimental validation.



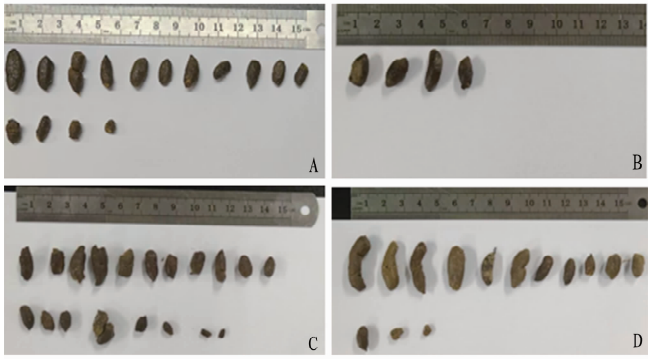
NOTE A – C. GO enrichment analysis of key targets, including biological processes (BP), cellular components (CC), and molecular functions (MF); D. KEGG pathway enrichment analysis, in which the NOD-like receptor pathway was identified as a key pathway.

Fig. 2 Functional enrichment and pathway analyses of key targets of XYXD in the treatment of STC

3.4 General condition and body weight comparison among different groups During the experimental period, rats in the control group were energetic, with lustrous fur and free movement. From days 3 to 4 after model establishment, rats in the model group gradually exhibited signs of lethargy, dull fur, and a tendency to huddle together with reduced activity. Following drug intervention, rats in both the XYXD and mosapride groups showed varying degrees of improvement in mental status and activity level, with the XYXD group demonstrating superior effects compared to the mosapride group.

3.5 Effects of XYXD on defecation function and gastrointestinal motility in STC rats Evaluation after the completion of drug intervention (Fig. 3 – 4) showed that, compared with the control group, rats in the model group exhibited a significantly prolonged time to first black stool defecation, as well as marked decreases in fecal pellet count, wet weight, and water content ($P<0.05$), confirming severe intestinal transit impairment in the model. Compared with the model group, both the XYXD and mosapride groups significantly shortened the time to first black stool defecation and improved all fecal parameters ($P<0.05$).

In the small intestinal propulsion experiment (Fig. 5), the small intestinal propulsion rate in the model group was significantly lower than that in the control group ($P<0.05$). Following drug

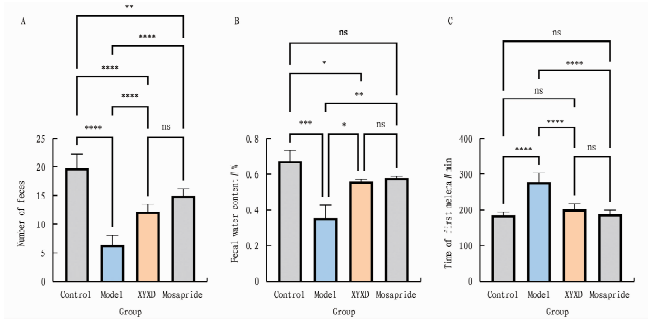


NOTE A. Control group; B. Model group; C. XYXD group; D. Mosapride group. $n=5$.

Fig. 3 Comparison of fecal traits and particle number of rats in each group

administration, both the XYXD and mosapride groups exhibited a marked increase in small intestinal propulsion rate ($P<0.05$). In particular, the XYXD group was slightly superior to the mosapride group in shortening the time to first black stool defecation ($P<0.05$), whereas no statistically significant difference was observed between the two groups in terms of small intestinal propulsion rate ($P>0.05$).

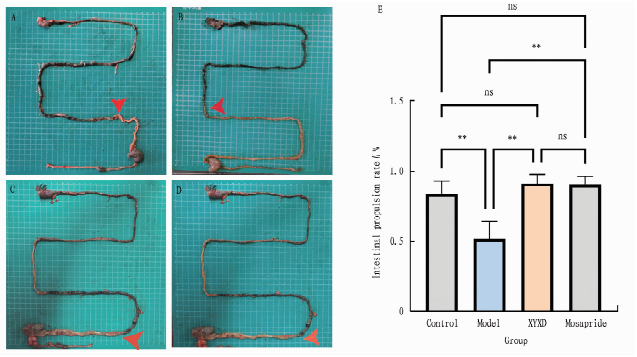
3.6 Effects of XYXD on histopathological injury and oxidative stress in colon tissue H&E staining results (Fig. 6) showed that compared with the control group, the model group exhibited disordered colonic tissue structure, significant thickening of the mucosal layer, irregular glandular architecture, and extensive inflammatory cell infiltration in the lamina propria and submucosa, indicating marked inflammation in the colon. Compared with the



NOTE A. Comparison of number of feces among groups; B. Comparison of fecal water content among groups; C. Comparison of time to first melena among groups. $^{*}P<0.05$, $^{**}P<0.01$, $^{***}P<0.001$, $^{****}P<0.0001$; $n=5$.

Fig. 4 Comparison of gastrointestinal motility and intestinal function among the rats in each group

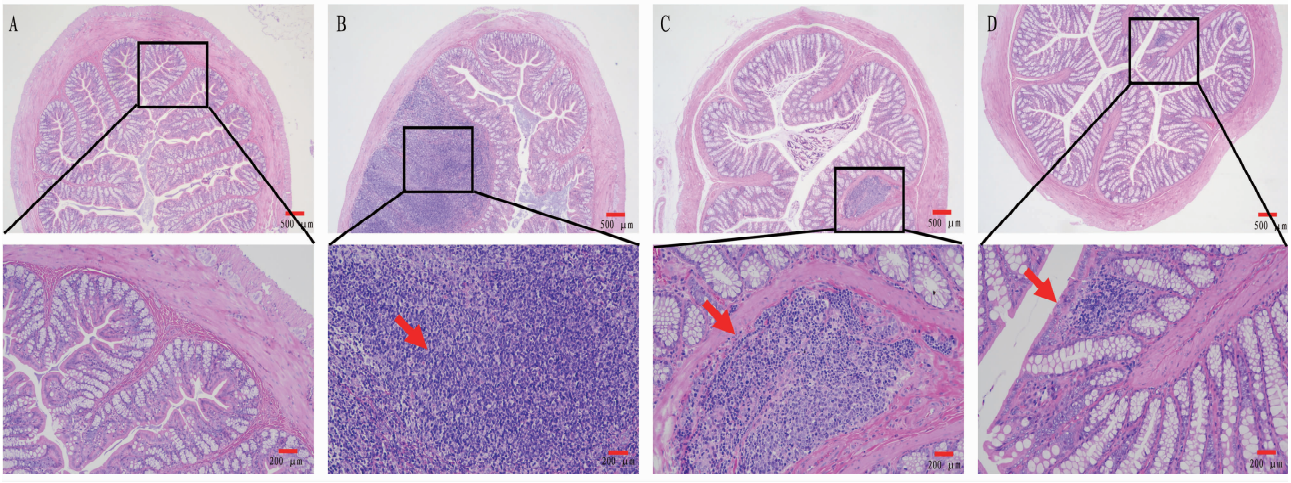
model group, the XYXD group still displayed focal inflammatory cell infiltration, mainly localized in the submucosa; however, the degree of inflammation was substantially alleviated, with glandular structures largely preserved and overall tissue integrity improved. The mosapride group showed a more intact colonic tissue structure with regularly arranged glands in the mucosal layer and only mild inflammatory cell infiltration in localized areas.



NOTE A. Small intestinal propulsion rate in the control group; B. Small intestinal propulsion rate in the model group; C. Small intestinal propulsion rate in the XYXD group; D. Small intestinal propulsion rate in the Mosapride group; E. Statistical graph of small intestinal propulsion rates in each group. $^{*}P<0.05$, $^{**}P<0.01$; $n=3$.

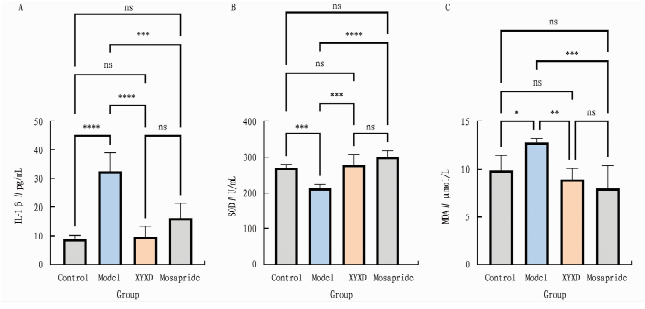
Fig. 5 Comparison of small intestinal propulsion rate in rats in each group

Serological analysis revealed (Fig. 7) that compared with the control group, the model group exhibited significantly elevated levels of the pro-inflammatory cytokine IL-1β and the oxidative stress marker MDA, along with a marked decrease in the activity of the antioxidant enzyme SOD ($P<0.001$). Following XYXD intervention, IL-1β and MDA levels were significantly downregulated, while SOD activity was substantially restored ($P<0.001$). These findings indicate that XYXD exerts potent anti-inflammatory and anti-oxidative stress effects, with improvements in inflammatory markers comparable to those observed in the mosapride group.



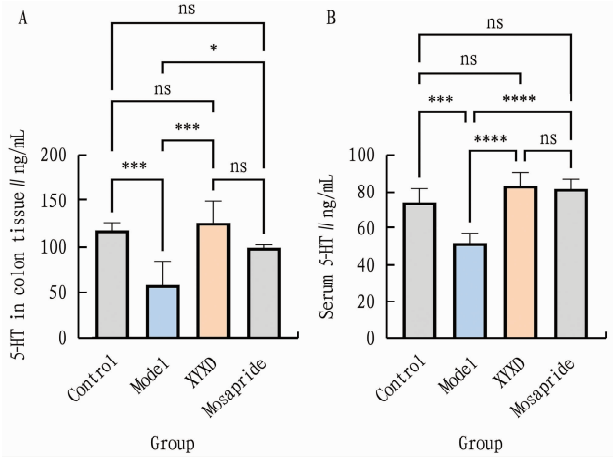
NOTE A. Control group showing intact mucosal architecture with regularly arranged glands; B. Model group, arrow indicates extensive inflammatory cell infiltration; C. XYXD group, arrow indicates substantially alleviated inflammation, with glandular structures largely preserved and overall tissue integrity improved; D. Mosapride group, arrow indicates mild inflammatory cell infiltration in localized areas, ×40 (scale bars = 500 μm); lower panels, ×200 (scale bar = 200 μm). $n = 3$.

Fig. 6 Effects of XYXD on the morphology of colon tissue in rats (HE)



NOTE A. Comparison of serum IL-1β levels among groups; B. Comparison of serum SOD activity among groups; C. Comparison of serum MDA levels among groups. $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$, $^{****}P < 0.000\,1$; $n = 5$.

Fig. 7 Effects of XYXD group on serum IL-1β, SOD and MDA contents in STC rats



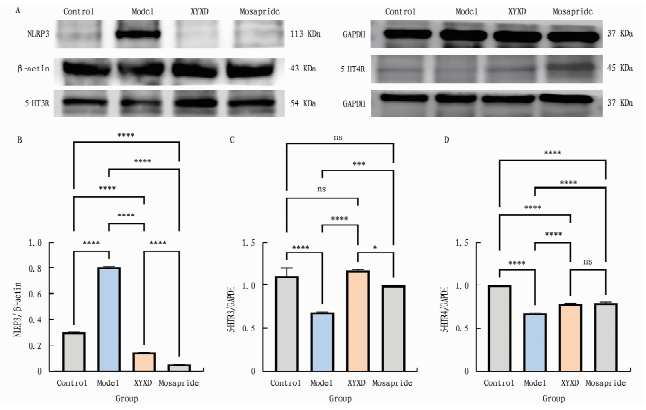
NOTE A. Comparison of colonic 5-HT levels among groups; B. Comparison of serum 5-HT levels among groups. $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$, $^{****}P < 0.000\,1$; $n = 5$.

Fig. 8 Comparison of 5-HT content in serum and colon of rats in each group

3.7 Effects of XYXD on 5-HT expression in STC rats

ELISA analysis revealed that compared with the control group, the 5-HT content in colon tissue was significantly decreased in the model group ($P < 0.001$). Following XYXD intervention, colonic 5-HT levels were effectively elevated ($P < 0.05$), suggesting that XYXD modulates the secretion of intestinal neurotransmitters. The comparison among groups is presented in Fig. 8.

3.8 Effects of XYXD on NLRP3, 5-HT3R, and 5-HT4R protein expression in STC rats Western blot analysis was performed to assess the protein expression levels of NLRP3 and 5-HT-related receptors. Compared with the control group, the model group exhibited significantly increased NLRP3 protein expression and significantly decreased expression levels of 5-HT3R and 5-HT4R in colon tissue ($P < 0.05$). Compared with the model group, both the XYXD and mosapride groups showed significantly



NOTE A – D. Comparison of NLRP3/5-HTR3/5-HTR4 protein expression levels in colon tissue among groups. $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$, $^{****}P < 0.000\,1$; $n = 3$.

Fig. 9 Electrophoresis of colon tissue protein expression in each group

reduced NLRP3 expression and significantly increased expression levels of 5-HT3R and 5-HT4R ($P<0.05$). The comparison among groups is shown in Fig. 9.

4 Discussion

The refractory nature of STC often stems from a vicious cycle of intestinal neuronal degeneration and microenvironmental dysregulation. In this study, through network pharmacology predictions followed by *in vivo* experimental validation, we demonstrated that XYXD improves colonic transit function via a dual mechanism involving anti-inflammatory effects and pro-motility actions. This finding not only provides evidence for the clinical application of the classic formula from the *Synopsis of the Golden Chamber*, known for its principle of "breaking blood stasis and expelling blood stasis", but also elucidates the molecular mechanism by which it alleviates intestinal motility disorders from the perspective of inflammatory regulation.

XYXD is a classic representative formula in TCM. Originally, it was used to treat postpartum abdominal pain due to retained blood stasis and various epigastric and abdominal pain syndromes[13-14]. In recent years, it has been widely applied in clinical practice for a variety of conditions, including anti-fibrotic therapy, renal protection, and anti-tumor treatment, among others[15-20]. With the expanding application and growing attention to TCM, the clinical utility of XYXD has been extended; notably, when used in combination therapy for constipation, it not only significantly alleviates symptoms and improves patients' quality of life but also demonstrates a favorable safety profile[15]. Functional constipation is influenced by multiple factors, including dietary habits, lack of physical activity, and psychological factors. Prolonged disease progression not only leads to defecation difficulties and perianal complications but also potentially increases the risk of colorectal cancer. Currently, the pathogenesis of functional constipation remains incompletely understood. In Western medicine, laxatives are commonly used in clinical practice; although they provide rapid relief, long-term use can lead to drug dependence and may induce melanosis coli[10-11]. In contrast, traditional Chinese medicine offers unique advantages in the treatment of chronic constipation, characterized by stable therapeutic efficacy and low toxicity[12].

In this study, network pharmacology was first employed to predict the potential molecular mechanisms underlying the intervention of XYXD in STC. The analysis revealed a total of 41 intersecting targets between XYXD and STC, which were significantly enriched in inflammation- and metabolism-related pathways, including the NOD-like receptor pathway, the IL-17 signaling pathway, and lipid metabolism. These findings suggest that XYXD may synergistically regulate intestinal inflammatory responses and neurotransmitter systems through multi-target and multi-pathway mechanisms, thereby restoring intestinal motility. This provides a robust theoretical foundation and mechanistic rationale for the subsequent *in vivo* experiments.

The core features of STC include diminished intestinal motility, prolonged defecation intervals, and hard stools, with local intestinal inflammation and neurotransmitter dysregulation following SCI serving as key pathological underpinnings. In this study, the model established by T8 spinal cord transection combined with loperamide administration falls within the pathological scope of NBD. NBD is a critical factor contributing to disability and depression in patients with SCI, characterized by prolonged colonic transit time and loss of intestinal reflex function[21]. Although this study aimed to investigate the general mechanisms underlying STC, NBD represents the most intractable subtype of STC with the most severe neurological impairment; therefore, findings from this model hold significant indicative value for elucidating the deep-seated pharmacological effects of XYXD.

From a pathophysiological perspective, SCI not only results in physical disruption of supraspinal central control over the intestine but also triggers local intestinal microenvironment remodeling[22-23]. The autonomic nervous system dysregulation following SCI leads to reduced colonic blood flow, tissue hypoxia, and subsequent accumulation of metabolites in the colonic mucosa, a pathological state that closely aligns with the TCM description of STC as "intestinal collateral stasis and obstruction of qi movement"[24]. As a classic formula for "activating blood circulation and resolving stasis", the rationale behind XYXD's therapeutic effect on NBD does not rely solely on mechanical prokinetic action; rather, it exerts secondary injury intervention on the ENS by improving local circulation and the inflammatory microenvironment[25-26].

In this study, rats in the model group exhibited a significant prolongation in the time to first black stool defecation, marked reductions in fecal pellet count, water content, and small intestinal propulsion rate, as well as pathological changes in colonic tissue including mucosal thickening, glandular disorganization, and inflammatory cell infiltration. Following XYXD intervention, all the above parameters were significantly improved. Moreover, the recovery of general conditions such as mental status and activity level in the XYXD group was superior to that in the mosapride group, suggesting that XYXD effectively promotes intestinal peristalsis, increases fecal water content, and improves defecation function. Mosapride, a commonly used prokinetic agent in clinical practice, exerts its effects by stimulating 5-HT4 receptors on cholinergic interneurons and myenteric plexus neurons in the gastrointestinal tract, thereby promoting acetylcholine release and enhancing gastrointestinal motility. Although the mosapride group ameliorated STC symptoms in this study, XYXD demonstrated superior efficacy in certain parameters, which may be attributed to its multi-target, multi-pathway mechanisms, reflecting the holistic regulatory advantage of TCM.

Recent evidence has indicated that inflammatory responses play a pivotal role in the progression of STC. As a core component of the innate immune system, hyperactivation of the NLRP3 inflammasome drives the maturation and release of IL-1β, thereby exacerbating colonic mucosal injury[8]. In this study, we observed

significant upregulation of both NLRP3 protein and IL-1β levels in the colon of model rats, suggesting that SCI triggers local NLRP3 inflammasome activation in the intestine, which may lead to impaired intestinal motility through sustained inflammatory cascades. Following XYXD intervention, colonic NLRP3 expression and inflammatory infiltration were markedly reduced, and tissue structural integrity was restored. These findings not only validate the predictions from network pharmacology but also align with the TCM theory of "stasis-induced inflammation".

Numerous studies have confirmed a close association between aberrant NLRP3 activation and constipation. The study by Zeng Chunlan et al[27]. demonstrated that in the model group, colonic expression of NLRP3 and caspase-1 was elevated, accompanied by decreased intestinal motility and reduced fecal water content; intervention with mirabilite downregulated NLRP3 and improved intestinal status, suggesting that dysregulation of this pathway represents an important mechanism. In a constipation study by Kasti *et al.*, a loperamide-induced mouse model of constipation showed activation of the NLRP3-GSDMD pyroptosis pathway in the colon, with upregulation of NLRP3, caspase-1, and IL-1β, along with intestinal epithelial pyroptosis and disruption of the intestinal barrier; intervention with hawthorn-probiotic postbiotics inhibited NLRP3 activation, alleviated damage, and relieved constipation, confirming that NLRP3 mediates pyroptosis in the progression of constipation[28]. Huang Hao *et al.* found that in a high-fat diet-induced obesity-associated constipation mouse model, colonic NLRP3 was hyperactivated and IL-1β levels were elevated, accompanied by abnormalities in the myenteric plexus pathway; NLRP3 gene knockout alleviated inflammation and restored intestinal motility[29]. Furthermore, patients with constipation-predominant irritable bowel syndrome exhibited higher expression levels of NLRP3 and IL-1β compared with healthy individuals, which were positively correlated with intestinal motility dysfunction[30], further supporting the association between the two. The progression of SCI-associated STC is accompanied by inflammatory responses. SCI-induced activation of the NLRP3 inflammasome in the intestine directly compromises the integrity of the myenteric plexus (enteric nervous system, ENS) by mediating the release of the pro-inflammatory cytokine IL-1β.

The present study confirmed that XYXD significantly inhibits the NLRP3 pathway and reduces neuronal toxicity resulting from excessive immune activation, which can be interpreted as a contemporary explication of the principle "removing stasis to protect the collaterals". In this formula, Dahuang expels stasis and activates blood circulation, Taoren moistens dryness, and Tujiechong unblocks meridians and breaks up accumulations. The combined use of these three herbs achieves the therapeutic effect of "removing stasis and unblocking collaterals" by suppressing NLRP3 activity and thereby blocking inflammatory responses, thus alleviating intestinal injury caused by excessive immune activation.

As a crucial neurotransmitter in the intestine, 5-HT regulates intestinal motility, secretion, and sensory function through binding to different receptors, among which 5-HT3R and 5-HT4R play key roles in promoting smooth muscle contraction and enhancing intestinal propulsion[31]. Under conditions of partial neural pathway disruption, the compensation of intestinal motility relies primarily on efficient signal transmission within the ENS neurotransmitter network.

In this study, we observed significantly reduced 5-HT levels in both the colon and serum of model rats, along with downregulated protein expression of 5-HT3R and 5-HT4R in colonic tissue, suggesting that dysregulation of the 5-HT system may be a key contributor to diminished intestinal motility in STC rats. This finding is consistent with the results of multiple previous studies. It has been reported that both mRNA and protein expression levels of 5-HT3R and 5-HT4R in the colon are significantly decreased in STC model rats, accompanied by reduced intestinal propulsion rate and fecal water content. Similarly, in a loperamide-induced mouse model of constipation, colonic expression of 5-HT3R and 5-HT4R was significantly downregulated, while intervention with *Torreya grandis* seed oil effectively reversed these changes and promoted recovery of intestinal peristalsis[32]. In addition, in a STC rat model established by intragastric administration of rhein, decreased mRNA and protein expression of 5-HT3R and 5-HT4R in the colon, along with impaired intestinal transit function, was also observed. Collectively, these studies confirm that impairment of the 5-HT signaling pathway, particularly downregulation of receptor expression, represents a critical link in the pathogenesis of STC.

Following XYXD intervention, both serum and colonic 5-HT levels were significantly elevated in rats, and the protein expression of 5-HT3R and 5-HT4R was markedly upregulated. In particular, the improvement in colonic 5-HT levels was superior to that observed in the mosapride group. These findings suggest that XYXD promotes the synthesis and release of 5-HT, upregulates the expression of its functional receptors, and enhances gastrointestinal parasympathetic excitability, thereby accelerating intestinal peristalsis. This strategy of "receptor sensitization" effectively compensates for the deficiency of central motility drive, enabling the impaired intestine to re-establish an effective peristaltic pattern. Similar mechanisms have been reported in other intervention studies. For instance, abdominal massage combined with a 5-HT4 receptor agonist significantly upregulated colonic 5-HT3R and 5-HT4R mRNA expression in STC rats and improved intestinal propulsion rate[33]. Acupuncture at points such as Houhai and Housanli also upregulated colonic 5-HT3R and 5-HT4R mRNA expression, alleviating constipation symptoms[34-35]. In addition, high-dose raw *Atractylodes macrocephala* combined with *Aurantii Fructus* was shown to promote intestinal motility by upregulating colonic 5-HT3R and 5-HT4R expression in STC rats[36], while *Bifidobacterium* and *Lactobacillus* triple viable tablets were similarly reported to increase colonic protein expression of 5-HT, 5-HT3R, and 5-HT4R, thereby improving intestinal transit function[37]. Collectively, these studies provide strong supporting evi-

dence for the modulation of the 5-HT system by XYXD in ameliorating STC, suggesting that intervention strategies targeting 5-HT3R/5-HT4R hold promising potential for the treatment of STC.

In the present study, XYXD employed was a clinically commonly used granule, whose quality control was performed in accordance with the manufacturer's established standards. For future research, we will establish the fingerprint of its chemical components using techniques such as HPLC/MS and further identify the specific active ingredients responsible for its therapeutic effects.

In summary, by integrating network pharmacology predictions with *in vivo* experimental validation, this study demonstrates that XYXD effectively improves defecation function and intestinal motility in STC rats. The underlying mechanism is closely associated with inhibition of NLRP3 inflammasome activation, modulation of the 5-HT signaling pathway, and alleviation of oxidative stress. This study provides a scientific experimental basis for the clinical application of XYXD in the treatment of STC and establishes a theoretical foundation for its broader application.

5 Conclusions

XYXD may exert its therapeutic effects on STC by inhibiting NLRP3 inflammasome-mediated chronic inflammation, thereby alleviating colonic tissue injury, and by upregulating the 5-HT/5-HTR signaling pathway to enhance intestinal motility.

Institutional Review Board Statement:

The study was conducted in accordance with the Declaration of Helsinki and approved by the Institutional Review Board of Fujian University of Chinese Medicine, China (ID No: 3w2025170) for studies.

References

[1] WILLITS AB, KADER L, ELLER O. Spinal cord injury-induced neurogenic bowel: A role for host-microbiome interactions in bowel pain and dysfunction[J]. Neurobiology of Pain, 2024, 15: 100156.

[2] LIU D, MAO M, LIU W. The role of the TRPV4 channel in intestinal physiology and pathology[J]. Journal of Inflammation Research, 2024, 17: 9307 – 9317.

[3] PANTHER EJ, DODD W, CLARK A, *et al.* Gastrointestinal microbiome and neurologic injury[J]. Biomedicines, 2022, 10(2): 500.

[4] WILLITS AB, KADER L, CHOUDHURY S. Intrarectal antagonism of calcitonin gene-related peptide prevents spinal cord injury-associated neurogenic bowel phenotypes[J]. Journal of Neurotrauma, 2025. DOI:10.1177/08977151251406659

[5] XIE Z, ROSE L, FENG J, *et al.* Enteric neuronal Piezo1 maintains mechanical and immunological homeostasis by sensing force[J]. Cell, 2025, 188(9): 2417 – 2432. e19.

[6] HEREDIA DJ, DICKSON EJ, BAYGUINOV PO, *et al.* Localized release of serotonin (5-hydroxytryptamine) by a fecal pellet regulates migrating motor complexes in murine colon[J]. Gastroenterology, 2009, 136(4): 1328 – 1338.

[7] LI T, FU B, ZHANG X, *et al.* Overproduction of gastrointestinal 5-HT promotes colitis-associated colorectal cancer progression via enhancing NLRP3 inflammasome activation[J]. Cancer Immunology Research, 2021, 9(9): 1008 – 1023.

[8] YIN F, JIANG Z, TONG Y, *et al.* Batatasin III alleviates slow transit constipation by regulating gut microbiota and inhibiting the NLRP3-IL-1β pathway[J]. Journal of Nutritional Biochemistry, 2026, 147: 110110.

[9] ZHANG S, TIAN D, XIA Z, *et al.* Chang-Kang-Fang alleviates diarrhea predominant irritable bowel syndrome (IBS-D) through inhibiting TLR4/NF-κB/NLRP3 pathway[J]. Journal of Ethnopharmacology, 2024, 330: 118236.

[10] BHARUCHA AE, LACY BE. Mechanisms, evaluation, and management of chronic constipation[J]. Gastroenterology, 2020, 158(5): 1232 – 1249.

[11] VLISMAS LJ, WU W, HO V. Idiopathic slow transit constipation: pathophysiology, diagnosis, and management[J]. Medicina, 2024, 60(1): 108.

[12] WANG L, WU F, HONG Y, *et al.* Research progress in the treatment of slow transit constipation by traditional Chinese medicine[J]. Journal of Ethnopharmacology, 2022, 290: 115075.

[13] CHEN XH, FU SQ, ZHU XY, *et al.* Exploration and analysis of syndrome and formula of classic famous formula Xiayuxue decoction and its application expansion[J]. Shanghai Journal of Traditional Chinese Medicine, 2023, 57(3): 56 – 58. (in Chinese).

[14] SANG HL, ZHAO M. Analysis on Xiayuxue decoction syndrome in Synopsis of Golden Chamber[J]. Chinese Journal of Basic Medicine in Traditional Chinese Medicine, 2022, 28(5): 686 – 688. (in Chinese).

[15] SHI RW, ZHANG T, LU WM, *et al.* Clinical observation on Xiayuxue decoction enema combined with acupoint application in the treatment of opioid-related constipation[J]. Western Journal of Traditional Chinese Medicine, 2022, 35(4): 124 – 126. (in Chinese).

[16] WEI DD, LI SS, WANG YJ, *et al.* Mechanism of Xiayuxue decoction intervening in renal fibrosis in rats through crosstalk of Wnt/β-catenin and TGF-β1/Smad signaling pathways[J]. Chinese Journal of Experimental Traditional Medical Formulae, 2021, 27(10): 8 – 14. (in Chinese).

[17] WEI DD, ZHANG T, GUO S, *et al.* Intervention effect of Xiayuxue decoction on myocardial fibrosis in rats based on regulation of TGFβ1/Smad2/ERK1 and Nrf2/HO-1 pathways[J]. Journal of Liaoning Traditional Chinese Medicine, 2025, 52(8): 175 – 182, 223 – 224. (in Chinese).

[18] ZHANG H, HU PP, SHAO JG, *et al.* Experimental study on Xiayuxue decoction inducing apoptosis of human hepatocellular carcinoma xenograft cells in nude mice[J]. Hebei Journal of Traditional Chinese Medicine, 2020, 42(6): 896 – 901, 800. (in Chinese).

[19] ZHANG JZ, XU QX, HU ZB, *et al.* Mechanism of Xiayuxue decoction combined with collateral-pricking therapy for hepatic fibrosis based on blood stasis and liver collateral disease theory[J]. Chinese Journal of Folk Medicine and Therapy, 2024, 32(15): 15 – 18, 128. (in Chinese).

[20] ZHAO X, WANG ZY, TAO L, *et al.* Intervention effect and mechanism of Xiayuxue decoction on renal injury in a mouse model of non-alcoholic fatty liver disease[J]. Journal of Clinical Hepatology, 2024, 40(11): 2213 – 2220. (in Chinese).

[21] TURE SD, OZKAYA G, SIVRIOGLU K. Relationship between neurogenic bowel dysfunction severity and functional status, depression, and quality of life in individuals with spinal cord injury[J]. Journal of Spinal Cord Medicine, 2023, 46(3): 424 – 432.

[22] JING Y, BAI F, YU Y. Spinal cord injury and gut microbiota: A review[J]. Life Sciences, 2021, 266: 118865.

(To page 24)

conazole nitrate cream in treating tinea pedis[D]. Nanchang: Jiangxi University of Traditional Chinese Medicine, 2024. (in Chinese).
[14] FANG Y, QI T, HE X, *et al.* Research progress on application and mechanism of antifungal traditional Chinese medicines[J]. Chinese Journal of Mycology, 2025, 20(4): 439-444. (in Chinese).
[15] ZHAO C, MIAO YY, XU J, *et al.* Evaluation of the anti-inflammatory, analgesic and skin wound repair activities of different extracts of *Senecio scandens* Buch-Ham[J]. Chinese Journal of Hospital Pharmacy, 2025, 45(8): 851-858. (in Chinese).
[16] ZHANG SN, YE C, LAN QH, *et al.* Research progress on quercetin resource, its pharmacological effect and major dosage form[J]. Journal of Wenzhou Medical University, 2025, 55(1): 66-74. (in Chinese).
[17] HU ZX, TONG L, GENG YM, *et al.* A review on pharmacological activities and preparations of luteolin[J]. Clinical Journal of Chinese Medicine, 2022, 14(10): 141-145. (in Chinese).
[18] YIN L, LIU QH, LIAO XW, *et al.* Research progress on the antibacterial effects of berberine and its derivatives[J]. Chemical Reagents, 2026, 48(5): 13-21. (in Chinese).
[19] XIE S, WANG WQ, LI WJ, *et al.* Research progress and prospect of pharmacological effects, pharmacokinetics and adverse reactions of matrine[J]. Chinese Traditional and Herbal Drugs, 2025, 56(10): 3712-3728. (in Chinese).
[20] FU JY, SHANG QH, JIANG L, *et al.* Research progress on the efficacy, indications, clinical applications, and modern pharmacological effects of Baibu[J]. Chinese Journal of Medicinal Guide, 2025, 27(9): 896-901. (in Chinese).
[21] WU B. Research progress on pseudolaric acid B[J]. Evaluation and Analysis of Drug-Use in Hospitals of China, 2015, 15(10): 1422-1423. (in Chinese).
[22] TANG LL. Study on the mechanism of Jiawei ShengjiYuhong ointment regulating the expression of TNF-α and AQP3 to promote wound healing of damp-heat anal fistula after operation[D]. Xianyang: Shaanxi University of Chinese Medicine, 2022. (in Chinese).
[23] XIAO WD. Identification and functional verification of protein coding genes and IncRNAs in Trichophyton mentagrophytes-induced rabbit dermatophytosis[D]. Ya'an: Sichuan Agricultural University, 2018. (in Chinese).
[24] LI BT, LI HX, ZHAO ZX, *et al.* Exploring the mechanism of Guizhi Fuling Wan in treating psoriasis based on network pharmacology[J]. Guizhou Medical Journal, 2026, 50(3): 451-455, 462. (in Chinese).
[25] LIN JL, JIAO WX, ZHOU ZY, *et al.* Research progress in anti-apoptotic effect of traditional Chinese medicine on neural cells[J]. Chinese Journal of Basic Medicine in Traditional Chinese Medicine, 2023, 29(9): 1569-1573. (in Chinese).
[26] KALONI D, DIEPSTRATEN ST, STRASSER A, *et al.* BCL-2 protein family: Attractive targets for cancer therapy[J]. Apoptosis, 2023, 28(1-2): 20-38.
[27] VARA JúF, CASADO E, DE CASTRO J, *et al.* PI3K/Akt signalling pathway and cancer[J]. Cancer Treatment Reviews, 2004, 30(2): 193-204.
[28] TIAN LL, YIN ZR, CAO GZ, *et al.* Anti-aging effect and molecular mechanism of Xiyangshen Sanqi Danshen Granules based on metabolomics and bioinformatics[J]. China Journal of Chinese Materia Medica, 2024,49(7): 1932-1946. (in Chinese).

(From page 10)

[23] JING Y, YU Y, BAI F, *et al.* Effect of fecal microbiota transplantation on neurological restoration in a spinal cord injury mouse model: Involvement of brain-gut axis[J]. Microbiome, 2021, 9(1): 59.
[24] JIANG Y, LIU G, DENG B, *et al.* The efficacy and pharmacological mechanisms of traditional Chinese medicine in the treatment of spinal cord injury[J]. Journal of Orthopaedic Surgery and Research, 2025, 20(1): 748.
[25] MA WT, LIU XL, TAO L, *et al.* Improvement effect of Xia Yu Xue decoction on hepatic inflammation and steatosis in mice with alcoholic liver disease[J]. Journal of Clinical Hepatology (Electronic Edition), 2021, 13(2): 40-51. (in Chinese).
[26] SHA HX. Therapeutic effect and related mechanism of classic formula "Xia Yu Xue decoction" on acute radiation enteritis in rats[D]. Nanjing: Nanjing University of Chinese Medicine, 2019. (in Chinese).
[27] ZENG CL, SHUAI LJ, DUAN QQ. Effect of Glauber's Salt on intestinal flora in rats with functional constipation by regulating NLRP3/caspase-1 signaling pathway[J]. Journal of Special Products Research, 2025, 47(6): 39-46. (in Chinese).
[28] KASTI A, KATSAS K, NIKOLAKI MD, *et al.* The role and the regulation of NLRP3 inflammasome in irritable bowel syndrome: A narrative review[J]. Microorganisms, 2025, 13(1): 171.
[29] HUANG H, PENG Y, XIAO L, *et al.* Electroacupuncture improves NLRP3-mediated inflammation and gastric motility in diabetic gastroparesis rats by activating AMPK and regulating mitochondrial homeostasis [J]. Acupuncture Research, 2025, 50(10): 1133-1142. (in Chinese).
[30] ELMENEZA S. Role of NLRP3 inflammasomes in disorders of children's digestive systems: A narrative review[J]. Pediatric Reports, 2025, 17(5): 103.
[31] ZHU Y, CHENG J, YIN J, *et al.* Effects of sacral nerve electrical stimulation on 5-HT and 5-HT3AR/5-HT4R levels in the colon and sacral cord of acute spinal cord injury rat models[J]. Molecular Medicine Reports, 2020, 22(2): 763-773.
[32] WANG X, GUO R, YU Z, *et al.* Torreya grandis kernel oil alleviates loperamide-induced slow transit constipation via up-regulating the colonic expressions of Occludin/Claudin-1/ZO-1 and 5-HT3R/5-HT4R in BALB/c mice[J]. Molecular Nutrition & Food Research, 2024, 68(4): e2300615.
[33] WANG DL, MA XW. Effect of abdominal massage on neurotransmitters and 5-HT receptor expression in rats with slow transit constipation[J]. West China Journal of Traditional Chinese Medicine, 2021, 34(7): 29-33. (in Chinese).
[34] YI HR. Regulatory effect of Buzhong Yiqi decoction modified on qi deficiency type constipation based on the theory of "all diseases arise from qi"[D]. Nanchang: Jiangxi University of Traditional Chinese Medicine, 2023. (in Chinese).
[35] YI LM, WEI MY, ZHOU J, *et al.* Effect of dexmedetomidine injection at Zusanli acupoint on postoperative PONV and gastrointestinal hormone levels[J]. Chinese Journal of Traditional Chinese Medicine Science and Technology, 2024, 31(4): 580-585. (in Chinese).
[36] GONG YX, WANG H, HOU Y, *et al.* Effect of large-dose raw Atractylodes rhizome combined with Aurantii Fructus on the expression of 5-HT3R and 5-HT4R in the colon of rats with slow transit constipation [J]. Chinese Journal of Integrated Traditional and Western Medicine, 2019, 39(8): 988-992. (in Chinese).
[37] SU HX, FU ZY, GAO YZ, *et al.* Clinical efficacy of Jiangshi granules combined with probiotics in the treatment of IBS-D of spleen-stomach weakness type and its effect on serum 5-HT, SP and inflammatory factors[J]. Lishizhen Medicine and Materia Medica Research, 2023, 34(9): 2172-2174. (in Chinese).