

# Identifying medication regularity of traditional Chinese medicine and potential pharmacological mechanism of Jiedu Sangen decoction in colorectal cancer treatment by data mining and network pharmacology

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## Author contributions

PPW, LTS designed the study. XJR wrote the manuscript, which was revised by LTS and PPW. XJR, XCY, and LHQ conducted the data acquisition, extraction, and analysis. XCY and YWX visualized data. All authors read and approved the final manuscript.

## Competing interests

The authors declare no conflicts of interest.

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## Abbreviations

CRC, colorectal cancer; TCM, traditional Chinese medicine; JSD, Jiedu Sangen decoction; FA, formic acid; CUR, Curtain gas; ISVF, IonSapary Voltage Floating; IDA, information-dependent acquisition; DP, Declustering potential; PPI, protein-protein interaction; DC, degree BC, betweenness; CC, closeness; BP, biological processes; CC, cellular components; MF, molecular functions; KEGG, Kyoto Encyclopedia of Genes and Genomes; MNCs, mononuclear cells; TME, tumor microenvironment; TCR, T cell receptor.

## Citation

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## Abstract

**Objective:** Colorectal cancer (CRC) is one of the most common malignancies in the world, and traditional Chinese medicine (TCM) is widely used in its treatment in China. However, the medication rules of TCM for CRC treatment remain unclear. Therefore, data mining combined with network pharmacology was utilized to establish treatment principles and rules. **Methods:** The CRC cases treated at Zhejiang Provincial Hospital of Chinese Medicine from January 1, 2016 to October 31, 2021 were analyzed using data mining methods. UPLC-Q/TOF-MS analysis was performed to identify the chemical composition of Jiedu Sangen decoction (JSD). Network pharmacology was used to reveal the therapeutic mechanism of JSD. **Results:** A total of 312 cases 2,998 prescriptions that met the inclusion criteria used 343 kinds of traditional Chinese medicines. The nature of the herbs used in treatment was mainly warm and mild. The taste was mainly sweet, bitter, and pungent. The meridian tropisms were mainly the spleen meridian, followed by lung and stomach meridians. Tonifying and replenishing herbs were the most frequently used in treatment. High-frequency herbs were classified into 11 categories by cluster analysis, and 42 association rules were obtained by association rule analysis. Combined with complex network analysis, 3 core prescriptions for clinical CRC treatment were obtained. Jiedu Sangen decoction contains 64 chemical ingredients, out of which 31 active ingredients were identified, including polydatin, caffeic acid, and glutamic acid, along with 130 potential targets such as AKT1, SRC, and MAPK1. Jiedu Sangen decoction may play a role in regulating inflammation, immunity, metabolism, and hormones in the development of CRC via pathways such as the relaxin signaling pathway, IL-17 signaling pathway, prolactin signaling pathway, and T cell receptor signaling pathway. **Conclusions:** This study summarizes the treatment and medication principles for clinical CRC treatment, promoting the inheritance and development of the traditional Chinese medical experience.

**Keywords:** data mining; network pharmacology; traditional Chinese medicine; colorectal cancer; Jiedu Sangen decoction

## Introduction

Colorectal cancer (CRC) is the third most common malignancy and the second leading cause of cancer-related death worldwide, accounting for about 10% of all cancer diagnoses and deaths each year [1]. By 2030, there are estimated to be 2.2 million new cases globally [2]. With an increasing incidence and mortality in China, CRC threatens people's health seriously, especially those under 50 years old [3, 4]. Despite significant advances in endoscopic, surgical, and adjuvant treatment, the overall survival and life quality of survivors remain poor [5], with symptoms such as bowel symptoms, pain, and fatigue [6, 7]. Moreover, for some patients, adjuvant chemotherapy may compromise functional recovery and be associated with inferior survival [8]. Integrative medicine can alleviate the side effects of surgery, radiotherapy, and chemotherapy, improve patients' quality of life, and prevent tumor metastasis and recurrence [9, 10].

Traditional Chinese medicine (TCM) is the primary form of complementary and alternative medicine for patients with CRC in China, which is also popular worldwide [11]. Clinical trials have found that Xiao-Chai-Hu-Tang, a traditional Chinese medicine formula, could improve depressive symptoms and intestinal dysbiosis in cancer patients, and its anti-tumoral effect was also confirmed in mouse models by regulating intestinal flora, suppressing the activation of the TLR4/MyD88/NF- $\kappa$ B pathway, and regulating inflammatory cytokine levels [12]. Reports have also demonstrated that Sino decoction with acupuncture together could lower the incidence of postoperative ileus, as well as complications such as pain, abdominal distension, and fever in CRC patients after resection [13]. Sijunzi decoction may suppress tumor development and be associated with better prognosis of CRC patients by upregulating the expression of KLF4 [14]. Moreover, combination therapy with Gegen Qinlian decoction and anti-PD-1 drugs showed a significant antitumor effect in vivo by remodeling gut microbiota and the tumor microenvironment [15].

Traditional Chinese medicine has a long history of treating colorectal cancer. Although the term "colorectal cancer" cannot be found in ancient Chinese classics. It tends to be the category of "Changfeng" (A disease characterized by hematochezia caused by the entry of pathogenic wind into the intestines), "Zangdu" (A disease characterized by pain, hot and a weight-bearing sensation within the anus), and "Jijv" (A diseases caused by the accumulation of pathogenic qi in the intestine) in traditional Chinese concepts when combined with the clinical manifestations of CRC [16]. Through TCM syndrome differentiation, CRC can be classified into several syndromes, including dampness-heat accumulation, stasis and toxin stagnation, spleen and kidney deficiencies, and so on [17]. From the aspect of the disease stage, the syndromes of CRC are qi stagnation (A pathological state in which the movement of qi is inhibited) and blood stasis, damp-heat-toxin accumulation in the early stage, vital qi deficiency (A pathological state of body weakness due to deficiency of qi) and pathogenic qi (qi is the most basic substance that makes up the human body and sustains its vital activities) excess in the middle stage, and vital qi deficiency in the late stage [18]. For the damp-heat-toxin accumulation syndrome, the Xianlian Jiedu formula helped regulate the proportion of immune cells and prevent EMT in intestinal tissue [19]. Meta-analysis indicated that Jianpi Jiedu herbs could relieve the symptom burden of CRC patients with spleen deficiency [20]. Although TCM has formed its system for the diagnosis and treatment of disease, there is currently a lack of standards in CRC, which tends to rely on the subjective judgment of TCM physicians. Therefore, analysis tools are needed to summarize rules and establish criteria.

Data mining is the process of extracting latent information hidden in a vast volume of practical application data [21]. It offers valuable guidance for disease treatment and has attracted growing attention in medical research. Cluster analysis and association analysis are commonly used to explore syndrome differentiation [22] and

medication rule [23]. Given that the prescription rules of TCM therapy in CRC are unclear, conducting data mining on the TCM treatment of CRC is crucial. To this end, this study analyzed 2,998 prescriptions to obtain medication rules in terms of nature, taste, meridian tropism, and other factors.

Jiedu Sangen decoction (JSD) is an important TCM formula for the CRC treatment, which is composed of Tengligen (*Radix Actinidiae Argutae*), Huzhanggen (*Polygoni cuspidati rhizoma*), and Shuiyangmeigen (Root of *Adina rubella*). However, there is limited research on the antitumor ability of JSD, and its underlying mechanism is not well understood, which limits its clinical application. Network pharmacology, which shares a similar holistic philosophy with TCM, provides a novel approach to exploring the active substances, targets, and mechanisms of TCM formulae by constructing "compound-proteins/genes-disease" interaction networks [24]. It has been widely used in pharmacological mechanisms [25], intelligent recommendation systems of TCM formula [26], and toxicity prediction [27]. Therefore, network pharmacology was used in this study to investigate the relationship between Jiedu Sangen decoction and CRC.

This study intends to use data mining and network pharmacology to explore the experience and medication regularity for the TCM treatment of CRC based on clinical records provided by Zhejiang Provincial Hospital of Chinese Medicine. Thereby enriching and optimizing integrated TCM-based treatment of CRC. The design is shown in Figure 1.

## Methods

### Clinical information

A total of 312 cases and 2,998 prescriptions of CRC were obtained from a professor at Zhejiang Provincial Hospital of Chinese Medicine between January 1, 2016 and October 31, 2021. Inclusion criteria were as follows: (1) Individuals who have histologically confirmed colorectal cancer by clinical or pathological/cytological examination; (2) Patients are treated with traditional Chinese medicine decoction; (3) Those with complete medical records, including prescription information; (4) There is no restriction on gender and age. Exclusion criteria were as follows: (1) Those diagnosed with colorectal cancer metastases; (2) Individuals with other primary malignancies; (3) Traditional Chinese medicine for external use; (4) Uncooperative individuals. All the protocols and procedures of this study were approved by the Zhejiang Province Hospital of TCM Ethics Committee (2023-KL-121-01).

### Data entry and processing

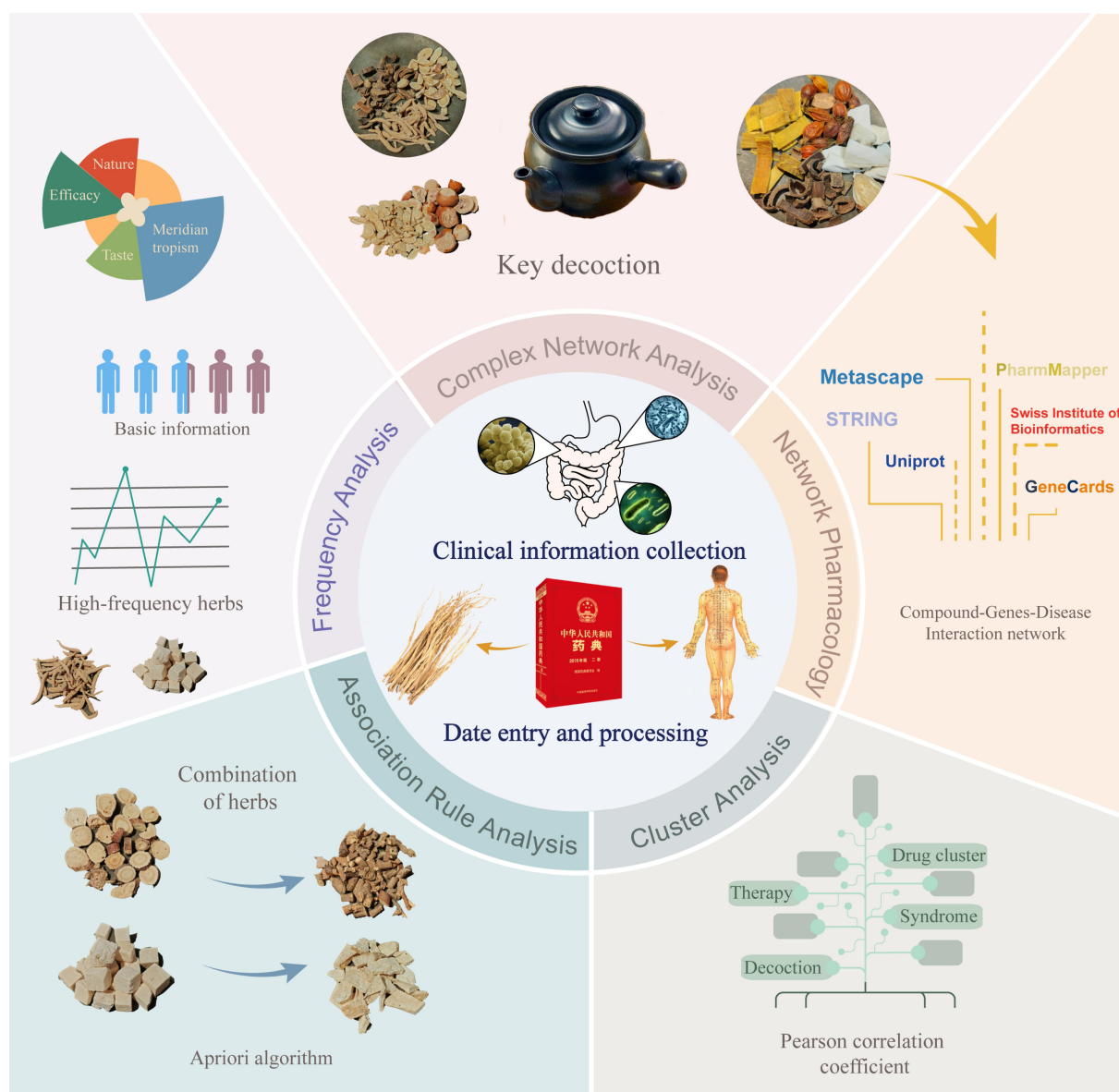
The collected 2,998 valid cases were input into Microsoft Excel, including patients' names, ages, genders, Western medicine diagnoses, TCM syndrome, and prescriptions. The names of herbs appearing in the prescription were standardized based on the *Chinese Pharmacopoeia (2020 edition)* [28] and *Traditional Chinese Pharmacology* [29], such as the "Daoudouzi (*Canavaliae semen*)" was unified as "Daodou (*Canavaliae semen*)", "Yangchunsha (*Amomi fructus*)" was unified as "Sharen (*Amomi fructus*)", "Danpi (*Moutan cortex*)" was unified as "Mudanpi (*Moutan cortex*)". The nature, flavor, meridian tropism, and efficacy of each herb were input and digitized in the database of Chinese medicinal herbs, according to the *Chinese Pharmacopoeia (2020 edition)*.

After the data entry was completed, another two persons checked the data to ensure accuracy and reliability.

### Frequency analysis

Frequency analysis was used to examine the frequency of use, nature, taste, meridian tropism, and the efficacy of each herb involved. All analyses were performed in Microsoft Excel 2019 and GraphPad Prism (version 8.4). The formula used was:

$$frequency(a) = \frac{\alpha}{\beta} \times 100\%$$



**Figure 1** Identifying medication regularity of traditional Chinese medicine and potential pharmacological mechanism of Jiedu Sangen decoction in colorectal cancer treatment by data mining and network pharmacology.

Where  $\alpha$  stands for the usage times of a specific herb, while  $\beta$  refers to the overall number of prescriptions or herbs.

#### Cluster analysis

52 high-frequency herbs with a frequency of herb use  $\geq 150$  times were carried out by hierarchical cluster analysis based on the Pearson correlation coefficient by SPSS statistics 26.0. The specific steps were as follows: First, each sample was set as one cluster and the Pearson distance between each other was calculated. Then, the nearest two clusters, which means they have the maximum correlation coefficient, were merged into one cluster. Then the Pearson distance between the newly generated cluster and each old cluster was recalculated. The calculation and merging process were repeated until all clusters were merged into one category. The results were displayed as a dendrogram. The Pearson distance between clusters was calculated using the following formula:

$$r = \frac{\sum_{i=1}^n (X_i - \bar{X})(Y_i - \bar{Y})}{\sqrt{\sum_{i=1}^n (X_i - \bar{X})^2 \sum_{i=1}^n (Y_i - \bar{Y})^2}}$$

Where  $X$  and  $Y$  represent two different clusters. The correlation coefficient ranges from  $[-1, 1]$ . Positive values represent a perfect correlation, negative values represent a negative correlation, and 0 indicates no correlation.

#### Association rule analysis

All herbs in this study performed association rule analysis using SPSS Modeler18.0 and bioinformatics (<http://www.bioinformatics.com.cn>). The candidate item sets were obtained using the Apriori algorithm. The results were presented in the form of  $X \rightarrow Y$ , where  $X$  represented antecedent and  $Y$  represented consequent. Each association rule contained four kernel indicators: support, confidence, expected confidence, and lift. The probability of  $X$  and  $Y$  presenting in one prescription was indicated by the level of support, which is positively correlated with the significance of the rule. The accuracy of the results is indicated by the confidence degree, which is the likelihood of  $Y$  appearing in a prescription based on  $X$  being in the same prescription. The expected confidence presents the likelihood of the consequences when the  $X$  is independent of the consequent. Lift is the ratio of the frequency of  $Y$  appearing in the presence of  $X$  to the frequency of  $Y$ , which is related to the mined association rule value [30]. The

formulae were expressed as follows:

$$\text{Support}(X) = \frac{|\{T|T \in D \text{ and } XCT\}|}{|D|} \quad (3)$$

$$\begin{aligned} \text{Support}(X \rightarrow Y) &= \text{Support}(X \cup Y) \\ &= \frac{|\{T|T \in D \text{ and } (X \cup Y)CT\}|}{|D|} \end{aligned} \quad (4)$$

$$\text{Confidence}(X \rightarrow Y) = \frac{\text{Support}(X \cup Y)}{\text{Support}(X)} = \frac{|\{T|T \in D \text{ and } (X \cup Y)CT\}|}{|\{T|T \in D \text{ and } (X)CT\}|} \quad (5)$$

$$\text{Expected Confidence}(X \rightarrow Y) = \frac{|\{T|T \in D \text{ and } (Y)CT\}|}{|D|} \quad (6)$$

$$\text{Lift}(X \rightarrow Y) = \frac{\text{Expected Confidence}(X \rightarrow Y)}{\text{Confidence}(X \rightarrow Y)} \quad (7)$$

Set the minimum value of support to 10%, the minimum threshold of confidence to 70%, and the maximum number of antecedent items to 1.

### Complex network analysis

SPSS Modeler 18.0 and Cytoscape 3.9.1 were used to find the potential core prescription composition of TCM in the treatment of CRC. We regarded the constituent herbs for CRC treatment as nodes and the connections between the two herbs as edges. Set the minimum number of links that could be displayed to 150, the upper limit of weak links to 50, and the lower limit of strong links to 150. The link size and transparency showed continuous changes.

### Preparations and chemical constituent analyses of JSD

JSD is composed of three herbs, namely Tengligen (*Radix Actinidiae Argutae*), Huzhanggen (*Polygoni cuspidati rhizoma*), and Shuiyangmeigen (Root of *Adina rubella*). All herbs were obtained from the Zhejiang Provincial Hospital of Chinese Medicine and extracted with boiling distilled water for 1 h. The extract was concentrated in a rotary evaporator (R-220 Pro, Buchi, Switzerland), stored at  $-80^{\circ}\text{C}$  for 24 h, and dried in a freeze dryer (VirTis AD2.0 EL, SP Scientific, America) for 48 h. 10 mg of the freeze-dried JSD powder was accurately weighed, dispersed in 1 ml methanol, and extracted with ultrasonic for 30 min. Then, it was centrifuged to obtain supernatant for UPLC-Q-TOF/MSE analysis.

Chromatographic analysis was performed using a Waters ACQUITY I-Class Plus UPLC system (Waters Company, United States) equipped with an ACQUITY UPLC BEH C18 column (150 x 2.1 mm, 1.7  $\mu\text{m}$ ). The mobile phase was composed of 0.1% formic acid (FA) in acetonitrile (A) and formic acid (B). The gradient profile was as follows: 0-3 min, 99%-85% B; 3-8 min, 85%-80% B; 8-15 min, 80%-60% B; 15-16 min, 60-40% B; 16-17 min, 40-1% B; 17-20 min, 1% B. The flow rate was 0.3 mL/min, the column temperature was  $40^{\circ}\text{C}$ , and the injection volume was 1  $\mu\text{L}$ .

Chemical analysis was conducted on SCIEX X-500R quadrupole time-of-flight mass spectrometry (AB SCIEX Company, United States), using TurboIonSpray ion source and ESI positive and negative ion scanning modes. The settings were as follows: Ion Source Gas1 (Gas1) was 55 L/min, Ion Source Gas2 (Gas2) was 45, and Curtain gas (CUR) was 35. The source temperature was set to  $600^{\circ}\text{C}$ . IonSapary Voltage Floating (ISVF) was 5500 V/-4500 V (positive and negative ion modes); TOF MS scan mas-to-charge (m/z) ranged from 50 to 1500 Da and production scan m/z ranged from 25 to 1000 Da. The accumulation time of the TOF MS scan was 0.25 s/spectra, while the product ion scan accumulation time was 0.035 s/spectra; the secondary mass spectrum was obtained using information-dependent acquisition (IDA) mode. Declustering potential (DP) was set to  $\pm 60$  V in positive and negative ion mode, collision energy was set to  $35 \pm 15$  eV, and candidate ions to monitor per cycle were set to 12, and the excluded isotopes within 4 Da. Data were collected and processed by

SCIEX OS software. TCM MS/MS Library, a secondary database of TCM configured by SCIEX OS, was searched according to the primary accurate mass number of compounds, isotope distribution ratio, and secondary mass spectrum.

### Target collection and screening

SwissADME (<http://www.swissadme.ch/index.php>) was utilized to screen potential components, with active ingredients defined as having GIabsorption "High" and Druglikeness "Yes > 2". PharmMapper (<http://www.lilab-ecust.cn/pharmmapper/>) was then used to screen and predict high-related targets, conditional on NormFit  $\geq 0.6$ . The full name of the gene was transformed into the gene symbol using Uniprot (<https://www.uniprot.org/>). To obtain CRC-related targets, "colorectal cancer", "colorectal adenomas", and "colorectal neoplasms" were used as keywords in GeneCards (<https://www.genecards.org/>). After integrating results and removing duplicates, the top 50% of relevance scores were considered differential targets. The overlapped genes between the herbal and disease targets were visualized using bioinformatics (<http://www.bioinformatics.com.cn>). Cytoscape3.7.2 was used to construct a target-compound network. The protein-protein interaction (PPI) network was established using the STRING database (<https://cn.string-db.org>). The results were imported into CytoscapeV3.7.2, and the CytoNCA plugin was used to calculate the degree (DC), betweenness (BC), and closeness (CC) of intersection genes.

### GO and KEGG enrichment analysis

The overlapped genes were analyzed using the Metascape database (<https://metascape.org/gp/index.html>) with "H. sapiens" selected as the species for biological processes (BP), cellular components (CC), molecular functions (MF), and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis. The results were visualized by bioinformatics.

### Result

#### Basic information

In our study, a total of 312 patients who met the inclusion criteria were included. There were 174 males and 138 females (Figure 2a), aged from 29 to 87, with an average age of  $61.43 \pm 11.87$  years. The highest number of cases were in the 60 to 70 years group, and the prevalence rate generally increased with age until 70 years of age (Figure 2b). The majority of patients were middle-aged or older. Figure 2c shows the location of tumors, with the whole colon and rectum being the main distribution areas.

#### Frequency analysis

**Medication frequency.** 343 herbs were used in 2,998 prescriptions, with a cumulative total frequency of herb use of 32,129. The top 10 frequently used herbs were Gancao (*Glycyrrhizae radix*), Fuling (*Poria*), Baizhu (*Atractylodis macrocephalae rhizoma*), Dangshen (*Codonopsis radix*), Banxia (*Pinelliae rhizoma*), Chaihu (*Bupleuri radix*), Tengligen (*Radix Actinidiae Argutae*), Chenpi (*Citri reticulatae pericarpium*), Huangqi (*Astragali radix*), and Huzhanggen (*Polygoni cuspidati rhizoma*) (Figure 3). Most of them belonged to tonics, while some could eliminate dampness and clear heat. The frequency of herbs was ranked from high to low, and those with a frequency  $\geq 150$  times were classified as high-frequency herbs (Table 1).

**Nature, taste, meridian tropism, and of herbs.** The nouns "Four Qi" and "Five Wei" first appeared in *Shennong Ben Cao Jing*. "Qi" (nature) and "Wei" (taste) represent not only the objective existence of herbs that can be detected by the senses, but also abstract generalizations of their functions, displaying the characteristics of each herb. "Four Qi" mainly comprises of cold, hot, warm, and cool. Additionally, herbs that have moderate effects are regarded as mild. In our study, warm herbs (7,904 times) and mild herbs (7,574 times) were the most commonly used, with frequencies of up to 31.36% and 30.05%, respectively (Figure 4a).

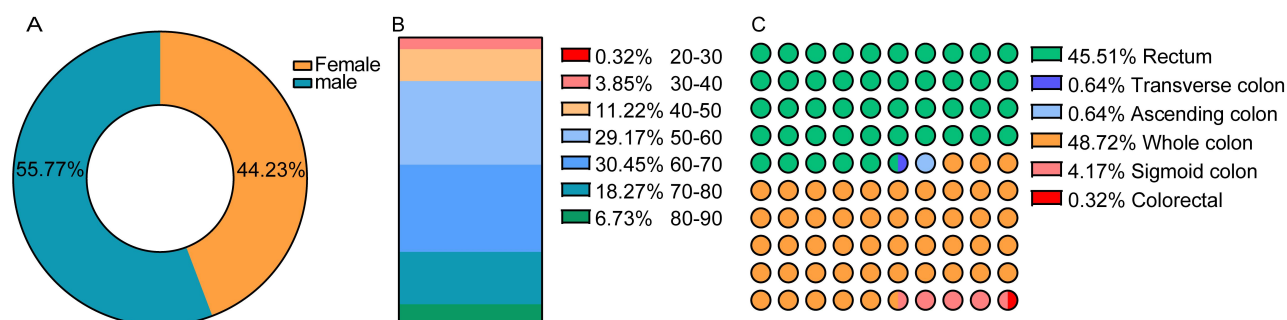


Figure 2 Basic information of 312 CRC patients

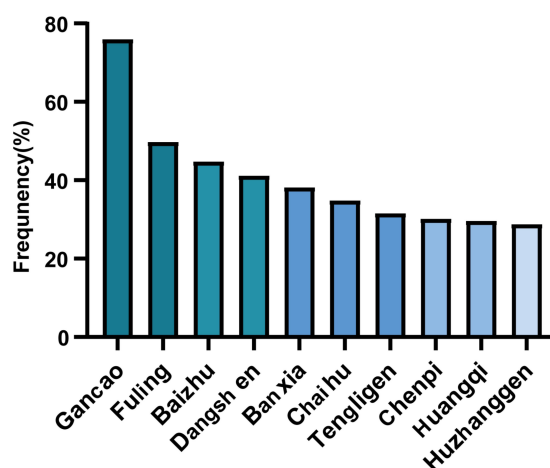


Figure 3 Frequency chart of top 10 herbs in prescriptions

Table 1 High-frequency herbs used in the TCM treatment for CRC

| No. | Herb                                      | Count | Frequency 1 (%) | Frequency 2 (%) |
|-----|-------------------------------------------|-------|-----------------|-----------------|
| 1   | <i>Glycyrrhizae radix et rhizoma</i>      | 2280  | 76.05           | 7.10            |
| 2   | <i>Poria</i>                              | 1494  | 49.83           | 4.65            |
| 3   | <i>Atractylodis macrocephalae rhizoma</i> | 1343  | 44.80           | 4.18            |
| 4   | <i>Codonopsis radix</i>                   | 1237  | 41.26           | 3.85            |
| 5   | <i>Pinelliae rhizoma</i>                  | 1146  | 38.23           | 3.57            |
| 6   | <i>Bupleuri radix</i>                     | 1047  | 34.92           | 3.26            |
| 7   | <i>Radix Actinidiae Argutae</i>           | 949   | 31.65           | 2.95            |
| 8   | <i>Citri reticulatae pericarpium</i>      | 907   | 30.25           | 2.82            |
| 9   | <i>Astragali radix</i>                    | 891   | 29.72           | 2.77            |
| 10  | <i>Polygoni cuspidati rhizoma</i>         | 865   | 28.85           | 2.69            |
| 11  | <i>Coicis semen</i>                       | 778   | 25.95           | 2.42            |
| 12  | <i>Jujubae fructus</i>                    | 735   | 24.52           | 2.29            |
| 13  | Root of <i>Adina rubella</i>              | 667   | 22.25           | 2.08            |
| 14  | <i>Paeoniae radix alba</i>                | 596   | 19.88           | 1.86            |
| 15  | <i>Astragali radix</i>                    | 510   | 17.01           | 1.59            |
| 16  | <i>Talcum</i>                             | 488   | 16.28           | 1.52            |
| 17  | <i>Coptidis rhizoma</i>                   | 446   | 14.88           | 1.39            |
| 18  | <i>Angelicae sinensis radix</i>           | 437   | 14.58           | 1.36            |
| 19  | <i>Aucklandiae radix</i>                  | 411   | 13.71           | 1.28            |
| 20  | <i>Pseudostellariae radix</i>             | 353   | 11.77           | 1.10            |
| 21  | <i>Hordei fructus germinatus</i>          | 341   | 11.37           | 1.06            |

Table 1 High-frequency herbs used in the TCM treatment for CRC (Continued)

| No. | Herb                                      | Count | Frequency 1 (%) | Frequency 2 (%) |
|-----|-------------------------------------------|-------|-----------------|-----------------|
| 22  | <i>Zingiberis rhizoma</i>                 | 339   | 11.31           | 1.06            |
| 23  | <i>Amomi fructus</i>                      | 308   | 10.27           | 0.96            |
| 24  | <i>Cimicifugae rhizoma</i>                | 308   | 10.27           | 0.96            |
| 25  | <i>Ilex cornuta</i>                       | 297   | 9.91            | 0.92            |
| 26  | <i>Dioscoreae rhizoma</i>                 | 276   | 9.21            | 0.86            |
| 27  | <i>Galli gigerii endothelium corneum</i>  | 273   | 9.11            | 0.85            |
| 28  | <i>Puerariae lobatae radix</i>            | 262   | 8.74            | 0.82            |
| 29  | <i>Ixeris denticulata</i>                 | 260   | 8.67            | 0.81            |
| 30  | <i>Crataegi fructus</i>                   | 248   | 8.27            | 0.77            |
| 31  | <i>Aconiti lateralis radix praeparata</i> | 242   | 8.07            | 0.75            |
| 32  | <i>Aurantii fructus immaturus</i>         | 242   | 8.07            | 0.75            |
| 33  | <i>Massa Medicata Fermentata</i>          | 238   | 7.94            | 0.74            |
| 34  | <i>Cinnamomi ramulus</i>                  | 235   | 7.84            | 0.73            |
| 35  | <i>Alismatis rhizoma</i>                  | 232   | 7.74            | 0.72            |
| 36  | <i>Magnoliae officinalis cortex</i>       | 231   | 7.71            | 0.72            |
| 37  | <i>Forsythiae fructus</i>                 | 215   | 7.17            | 0.67            |
| 38  | <i>Atractylodis rhizoma</i>               | 214   | 7.14            | 0.67            |
| 39  | <i>Zingiberis rhizoma recens</i>          | 210   | 7.00            | 0.65            |
| 40  | <i>Albiziae cortex</i>                    | 208   | 6.94            | 0.65            |
| 41  | <i>Rehmanniae radix praeparata</i>        | 207   | 6.90            | 0.64            |
| 42  | <i>Eucommiae cortex</i>                   | 206   | 6.87            | 0.64            |
| 43  | <i>Phellodendri chinensis cortex</i>      | 200   | 6.67            | 0.62            |
| 44  | <i>Cinnamomi cortex</i>                   | 197   | 6.57            | 0.61            |
| 45  | <i>Achyranthis bidentatae radix</i>       | 185   | 6.17            | 0.58            |
| 46  | <i>Ostreae concha</i>                     | 178   | 5.94            | 0.55            |
| 47  | <i>Gastrodiae rhizoma</i>                 | 167   | 5.57            | 0.52            |
| 48  | <i>Rhei radix et rhizoma</i>              | 166   | 5.54            | 0.52            |
| 49  | <i>Guanghuoxiang</i>                      | 160   | 5.34            | 0.50            |
| 50  | <i>Chuanxiong rhizoma</i>                 | 158   | 5.27            | 0.49            |
| 51  | <i>Rehmanniae radixhuang</i>              | 155   | 5.17            | 0.48            |
| 52  | <i>Ziziphi spinosae semen</i>             | 155   | 5.17            | 0.48            |

Frequency 1 (%) = number of herb/total number of prescriptions; Frequency 2 (%) = number of herb/total number of herb

“Five Wei” refers to pungent, sour, sweet, bitter, and salty, with astringency and weakness also included due to the development of the theory that astringency attaches to sour and weakness attaches to sweet. As shown in Figure 4b, the frequency of herbs’ taste is ranked from high to low as sweet (36.43%), bitter (24.41%), pungent (22.50%), weak (7.90%), sour (5.79%), astringent (2.51%), and salty (0.47%).

Meridian tropism demonstrates the specificity of pharmacological activity in a particular region of the body, which is the combination of the function of herbs with viscera and meridians. It can be classified into twelve categories: liver, heart, spleen, lung, kidney, gallbladder, small intestine, stomach, large intestine, bladder, pericardium, and triple energizer. According to Figure 4c, most of the herbs used in the CRC treatment belong to the spleen (23.69%), lung (20.09%), and stomach (15.19%) meridians, followed by the heart (9.49%) and liver (8.99%) meridians.

The frequency distribution of high-frequency herbs based on their efficacy is shown in Table 2. Tonifying and replenishing medicinal were the most frequently used herbs in treatment, followed by heat-clearing medicinal, exterior-releasing medicinal, and dampness-draining diuretic medicinal. In the tonifying and replenishing medicinal, based on the differences in qi (qi is the most basic substance that makes up the human body and sustains its vital

activities), blood, yin (Yin is the opposite of yang. The qualities of yin include cold/cool, dimness, descending, etc.), and yang (Yang is the opposite of yin. The qualities of yang include warm/hot, bright, ascending, etc.), the frequency of herbs from high to low was qi-tonifying herbs, blood-tonifying herbs, and yang-tonifying herbs (Figure 4d).

#### Cluster analysis

According to the results of high-frequency herbal cluster analysis (Figure 5) and clinical experience, high-frequency herbs can be divided into 11 categories.

Cluster 1: Tengligen (*Radix Actinidiae Argutae*), Huzhanggen (*Polygoni cuspidati rhizoma*), and Shuiyangmeigen (*Root of Adina rubella*) constitute Jiedu Sangen decoction, which has the function of clearing heat, detoxicating, dispersing blood stasis, and removing dampness.

Cluster 2: Dahuang (*Rhei radix et rhizoma*) and Huanglian (*Coptidis rhizoma*) belong to Dahuang Huanglian Xiexin decoction. They are bitter and cold in terms of nature and flavor, which can be used to clear away heat, purge fire, and detoxicate.

Cluster 3: The combination of Suanzaoren (*Ziziphi spinosae semen*) and Chuanxiong (*Chuanxiong rhizome*) has a nourishing effect on the blood, helps to regulate the liver, and promotes tranquility of the mind.

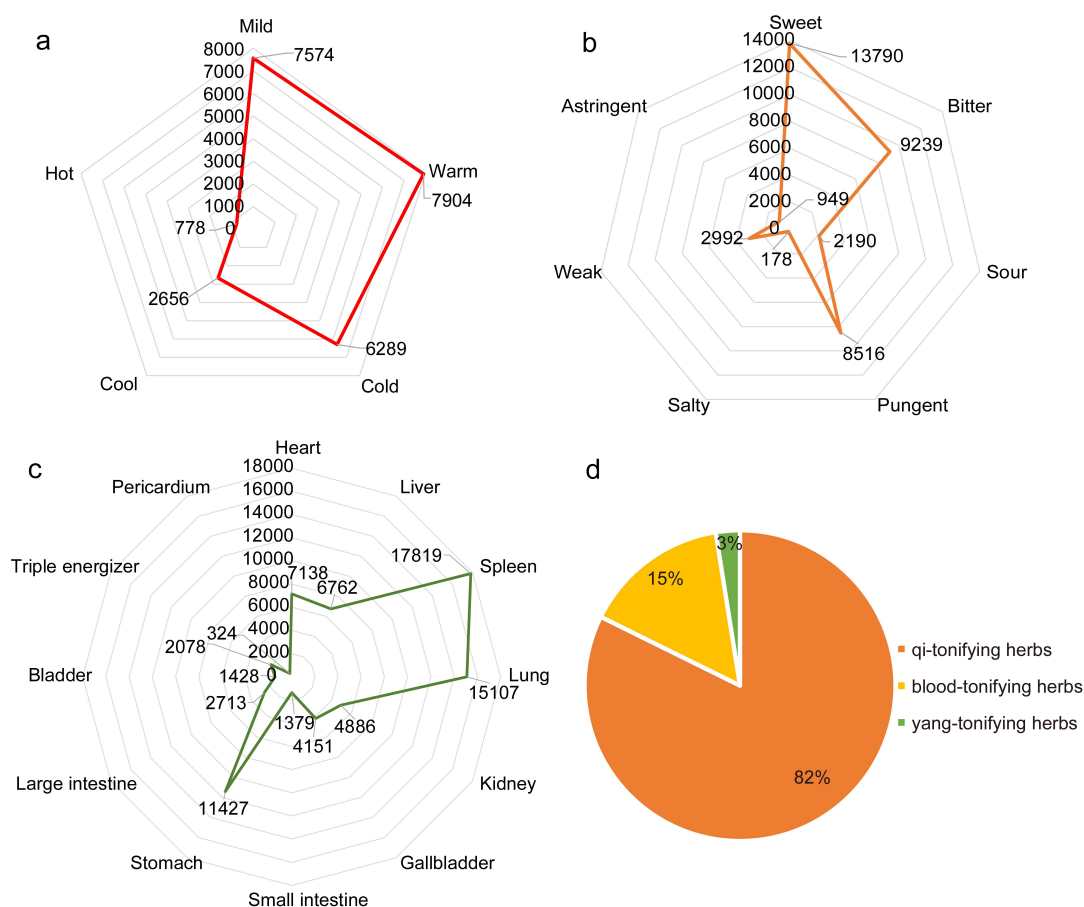


Figure 4 Frequency analysis results for nature, taste, meridian tropism, and efficacy of herbs

Table 2 The frequency distribution of high-frequency herbs based on efficacy

| No. | Efficacy                                          | Count | Frequency (%) | Herbs                                                                                                                                                                                                                                                                                                                                                  |
|-----|---------------------------------------------------|-------|---------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1   | Tonifying and replenishing medicinal              | 8180  | 25.46         | <i>Glycyrrhizae radix et rhizoma</i> , <i>Atractylodis macrocephalae rhizoma</i> , <i>Codonopsis radix</i> , <i>Jujubae fructus</i> , <i>Paeoniae radix alba</i> , <i>Astragali radix</i> , <i>Angelicae sinensis radix</i> , <i>Pseudostellariae radix</i> , <i>Dioscoreae rhizoma</i> , <i>Rehmanniae radix praeparata</i> , <i>Eucommiae cortex</i> |
| 2   | Heat-clearing medicinal                           | 3116  | 9.70          | <i>Radix Actinidiae Argutae</i> , <i>Astragali radixn</i> , <i>Coptidis rhizoma</i> , <i>Ixeris denticulata</i> , <i>Forsythiae fructus</i> , <i>Phellodendri chinensis cortex</i> , <i>Rehmanniae radixhuang</i>                                                                                                                                      |
| 3   | Exterior-releasing medicinal                      | 2729  | 8.49          | <i>Bupleuri radix</i> , <i>Root of Adina rubella</i> , <i>Cimicifugae rhizoma</i> , <i>Puerariae lobatae radix</i> , <i>Cinnamomi ramulus</i> , <i>Zingiberis rhizoma recens</i>                                                                                                                                                                       |
| 4   | Dampness-draining diuretic medicinal              | 3857  | 12.00         | <i>Poria</i> , <i>Polygoni cuspidati rhizoma</i> , <i>Coicis semen</i> , <i>Talcum</i> , <i>Alismatis rhizoma</i>                                                                                                                                                                                                                                      |
| 5   | Digestant medicinal                               | 1100  | 3.42          | <i>Hordei fructus germinatus</i> , <i>Galli gigerii endothelium corneum</i> , <i>Crataegi fructus</i> , <i>Massa Medicata Fermentata</i>                                                                                                                                                                                                               |
| 6   | Humidifying medicinal                             | 913   | 2.84          | <i>Amomi fructus</i> , <i>Magnoliae officinalis cortex</i> , <i>Atractylodis rhizoma</i> , <i>Guanghuoxiang</i>                                                                                                                                                                                                                                        |
| 7   | Qi-regulating medicinal                           | 1560  | 4.86          | <i>Citri reticulatae pericarpium</i> , <i>Aucklandiae radix</i> , <i>Aurantii fructus immaturus</i>                                                                                                                                                                                                                                                    |
| 8   | Interior-warming medicinal                        | 778   | 2.42          | <i>Zingiberis rhizoma</i> , <i>Aconiti lateralis radix praeparaia</i> , <i>Cinnamomi cortex</i>                                                                                                                                                                                                                                                        |
| 9   | Sedative medicinal                                | 363   | 1.13          | <i>Albiziae cortex</i> , <i>Ziziphi spinosae semen</i>                                                                                                                                                                                                                                                                                                 |
| 10  | Blood-activating and stasis-dissipating medicinal | 343   | 1.07          | <i>Achyranthis bidentatae radix</i> , <i>Chuanxiong rhizoma</i>                                                                                                                                                                                                                                                                                        |

Table 2 The frequency distribution of high-frequency herbs based on efficacy (Continued)

| No. | Efficacy                                                   | Count | Frequency (%) | Herbs                                             |
|-----|------------------------------------------------------------|-------|---------------|---------------------------------------------------|
| 11  | Calming liver and relieving wind medicinal                 | 345   | 1.07          | <i>Ostreae concha</i> , <i>Gastrodiae rhizoma</i> |
| 12  | Phlegm-removing, anti-tussive and anti-asthmatic medicinal | 1146  | 3.57          | <i>Pinelliae rhizoma</i>                          |
| 13  | Expelling wind and dampness medicinal                      | 297   | 0.92          | <i>Ilex cornuta</i>                               |

Frequency (%) = number of herb/total number of herbs

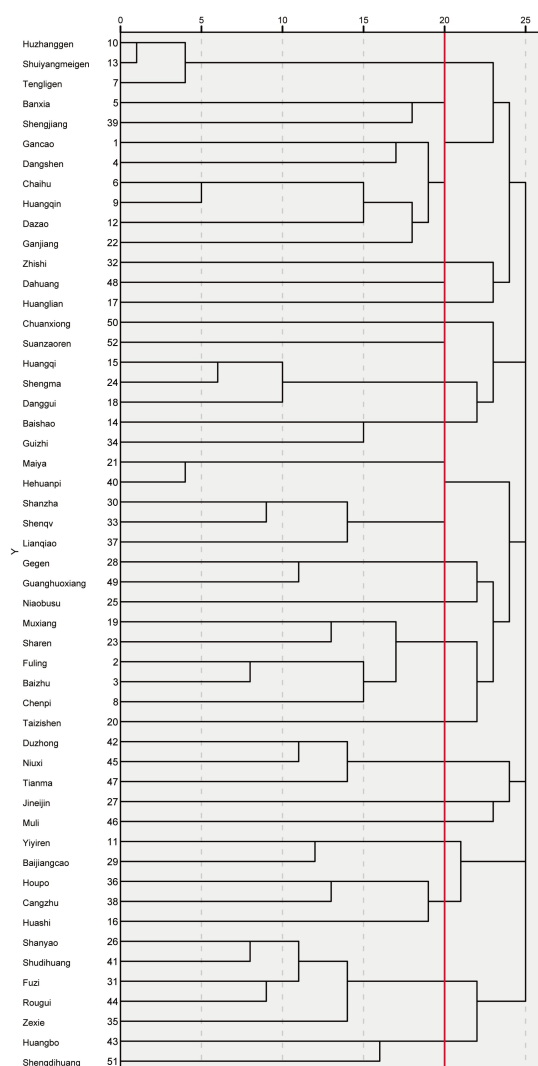


Figure 5 Cluster analysis of high-frequency herbs

Cluster 4: Huangqi (*Astragali radix*), Shengma (*Cimicifuga rhizome*), and Danggui (*Angelica sinensis radix*) are all ingredients in Buzhong Yiqi decoction. This decoction is mainly used to treat **qi deficiency** syndrome since it can invigorate **qi** and elevate **yang** without causing stagnation.

Cluster 5: Guizhi (*Cinnamomi Ramulus*), with its pungent nature and warm taste, promotes **Wei (protective function)** as well as **Yang (active and dynamic factors)**, while eliminating exterior pathogens. Baishao (*Paeoniae alba radix*), with its sour and sweet flavor, helps to converge **Ying (nutritive function)** and invigorate **Yin (repressive and stationary factors)**. The compatibility of Guizhi and Baishao can harmonize the relations between **Ying** and **Wei**, **Yin** and **Yang**, which are the basis of the normal function of the human body.

Cluster 6: Maiya (*Hordei fructus germinatus*) relieves indigestion caused by fruit, Shanzha (*Crataegi fructus*) relieves indigestion caused by meat, and Shenqu (*Massa Medicata Fermentata*) relieves indigestion

caused by pasta products. These three ingredients are combined in Jiaoshanxian (**The coking product of Maiya, Shanzha, and Shenqu**), which can also dispel the heat and tranquilize the mind with the aid of Lianqiao (*Forsythiae fructus*) and Hehuanpi (*Albiziae cortex*).

Cluster 7: The herbs of Cluster 7 have the effect of resolving exterior dampness and warming the **middle Jiao (The middle jiao is located between the diaphragm and navel and includes the spleen, stomach, liver and gallbladder.)** to alleviate diarrhea.

Cluster 8: Tianma (*Gastrodiae rhizoma*), Niuxi (*Achyranthis bidentatae radix*), and Duzhong (*Eucommiae cortex*) are all ingredients in Tianma Gouteng Yin decoction. This decoction has a calming effect on the liver, extinguishes wind, clears away heat, activates blood circulation, and tonifies the liver and kidney.

Cluster 9: The goal of Cluster 9 is to eliminate toxins and dissipate masses.

Cluster 10: Cluster 10 contributes to dehumidification.

Table 3 Association rules of two herbs

| No. | Consequent                                | Antecedent                                | Support (%) | Confidence (%) | Lift (%) |
|-----|-------------------------------------------|-------------------------------------------|-------------|----------------|----------|
| 1   | <i>Poria</i>                              | <i>Glycyrrhizae radix et rhizoma</i>      | 49.80       | 76.76          | 1.02     |
| 2   | <i>Poria</i>                              | <i>Atractylodis macrocephalae rhizoma</i> | 49.80       | 72.07          | 1.61     |
| 3   | <i>Atractylodis macrocephalae rhizoma</i> | <i>Glycyrrhizae radix et rhizoma</i>      | 44.73       | 87.55          | 1.16     |
| 4   | <i>Atractylodis macrocephalae rhizoma</i> | <i>Poria</i>                              | 44.73       | 80.24          | 1.61     |
| 5   | <i>Codonopsis radix</i>                   | <i>Glycyrrhizae radix et rhizoma</i>      | 41.26       | 89.09          | 1.18     |
| 6   | <i>Pinelliae rhizoma</i>                  | <i>Glycyrrhizae radix et rhizoma</i>      | 38.19       | 85.15          | 1.13     |
| 7   | <i>Bupleuri radix</i>                     | <i>Glycyrrhizae radix et rhizoma</i>      | 34.92       | 83.09          | 1.10     |
| 8   | <i>Bupleuri radix</i>                     | <i>Astragali radix</i>                    | 34.92       | 71.54          | 2.41     |
| 9   | <i>Radix Actinidiae Argutae</i>           | <i>Glycyrrhizae radix et rhizoma</i>      | 31.62       | 75.84          | 1.01     |
| 10  | <i>Citri reticulatae pericarpium</i>      | <i>Glycyrrhizae radix et rhizoma</i>      | 30.22       | 83.66          | 1.11     |
| 11  | <i>Citri reticulatae pericarpium</i>      | <i>Poria</i>                              | 30.22       | 76.16          | 1.53     |
| 12  | <i>Astragali radix</i>                    | <i>Bupleuri radix</i>                     | 29.65       | 84.25          | 2.41     |
| 13  | <i>Astragali radix</i>                    | <i>Glycyrrhizae radix et rhizoma</i>      | 29.65       | 79.53          | 1.06     |
| 14  | <i>Polygoni cuspidati rhizoma</i>         | <i>Radix Actinidiae Argutae</i>           | 28.85       | 76.42          | 2.42     |
| 15  | <i>Polygoni cuspidati rhizoma</i>         | <i>Glycyrrhizae radix et rhizoma</i>      | 28.85       | 76.42          | 1.02     |
| 16  | <i>Polygoni cuspidati rhizoma</i>         | Root of <i>Adina rubella</i>              | 28.85       | 75.61          | 3.40     |
| 17  | <i>Coicis semen</i>                       | <i>Glycyrrhizae radix et rhizoma</i>      | 25.95       | 71.85          | 0.95     |
| 18  | <i>Jujubae fructus</i>                    | <i>Glycyrrhizae radix et rhizoma</i>      | 24.52       | 94.83          | 1.26     |
| 19  | Root of <i>Adina rubella</i>              | <i>Radix Actinidiae Argutae</i>           | 22.25       | 98.80          | 3.12     |
| 20  | Root of <i>Adina rubella</i>              | <i>Polygoni cuspidati rhizoma</i>         | 22.25       | 98.05          | 3.40     |
| 21  | Root of <i>Adina rubella</i>              | <i>Glycyrrhizae radix et rhizoma</i>      | 22.25       | 77.66          | 1.03     |
| 22  | <i>Paeoniae radix alba</i>                | <i>Glycyrrhizae radix et rhizoma</i>      | 19.81       | 89.56          | 1.19     |
| 23  | <i>Astragali radix</i>                    | <i>Glycyrrhizae radix et rhizoma</i>      | 16.88       | 83.99          | 1.12     |
| 24  | <i>Talcum</i>                             | <i>Glycyrrhizae radix et rhizoma</i>      | 16.24       | 98.56          | 1.31     |
| 25  | <i>Coptidis rhizoma</i>                   | <i>Glycyrrhizae radix et rhizoma</i>      | 14.84       | 77.30          | 1.03     |
| 26  | <i>Angelicae sinensis radix</i>           | <i>Glycyrrhizae radix et rhizoma</i>      | 14.58       | 86.96          | 1.16     |
| 27  | <i>Angelicae sinensis radix</i>           | <i>Atractylodis macrocephalae rhizoma</i> | 14.58       | 78.95          | 1.76     |
| 28  | <i>Aucklandiae radix</i>                  | <i>Glycyrrhizae radix et rhizoma</i>      | 13.68       | 89.02          | 1.18     |
| 29  | <i>Aucklandiae radix</i>                  | <i>Atractylodis macrocephalae rhizoma</i> | 13.68       | 88.29          | 1.97     |
| 30  | <i>Aucklandiae radix</i>                  | <i>Poria</i>                              | 13.68       | 77.07          | 1.55     |
| 31  | <i>Aucklandiae radix</i>                  | <i>Codonopsis radix</i>                   | 13.68       | 73.66          | 1.79     |
| 32  | <i>Pseudostellariae radix</i>             | <i>Glycyrrhizae radix et rhizoma</i>      | 11.74       | 90.34          | 1.20     |
| 33  | <i>Zingiberis rhizoma</i>                 | <i>Glycyrrhizae radix et rhizoma</i>      | 11.31       | 91.15          | 1.21     |
| 34  | <i>Cimicifugae rhizoma</i>                | <i>Glycyrrhizae radix et rhizoma</i>      | 10.27       | 88.96          | 1.18     |
| 35  | <i>Cimicifugae rhizoma</i>                | <i>Astragali radix</i>                    | 10.27       | 87.99          | 5.21     |
| 36  | <i>Amomi fructus</i>                      | <i>Glycyrrhizae radix et rhizoma</i>      | 10.27       | 87.01          | 1.16     |
| 37  | <i>Cimicifugae rhizoma</i>                | <i>Bupleuri radix</i>                     | 10.27       | 85.06          | 2.44     |
| 38  | <i>Cimicifugae rhizoma</i>                | <i>Codonopsis radix</i>                   | 10.27       | 81.17          | 1.97     |
| 39  | <i>Amomi fructus</i>                      | <i>Atractylodis macrocephalae rhizoma</i> | 10.27       | 80.84          | 1.81     |
| 40  | <i>Cimicifugae rhizoma</i>                | <i>Atractylodis macrocephalae rhizoma</i> | 10.27       | 77.60          | 1.73     |
| 41  | <i>Amomi fructus</i>                      | <i>Poria</i>                              | 10.27       | 77.27          | 1.55     |

Table 3 Association rules of two herbs (Continued)

| No. | Consequent           | Antecedent                           | Support (%) | Confidence (%) | Lift (%) |
|-----|----------------------|--------------------------------------|-------------|----------------|----------|
| 42  | <i>Amomi fructus</i> | <i>Citri reticulatae pericarpium</i> | 10.27       | 71.43          | 2.36     |

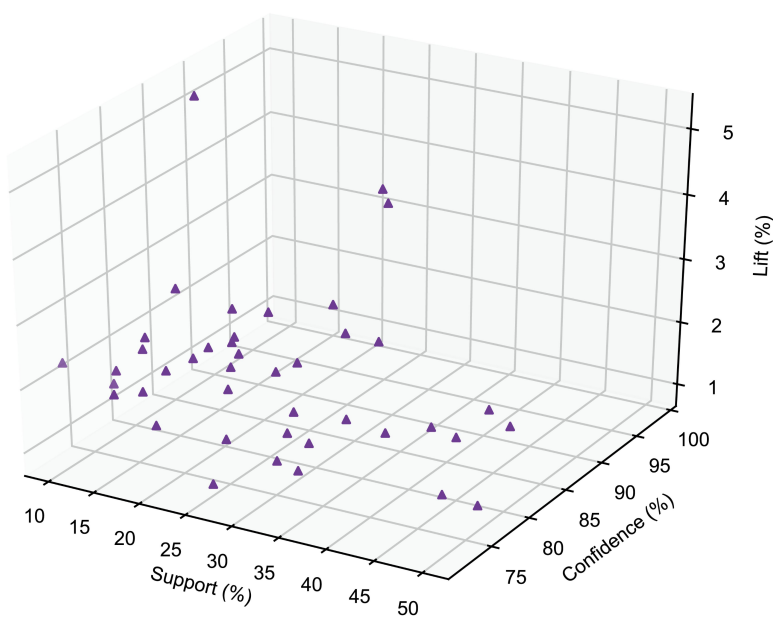


Figure 6 Scatter diagram of all rules

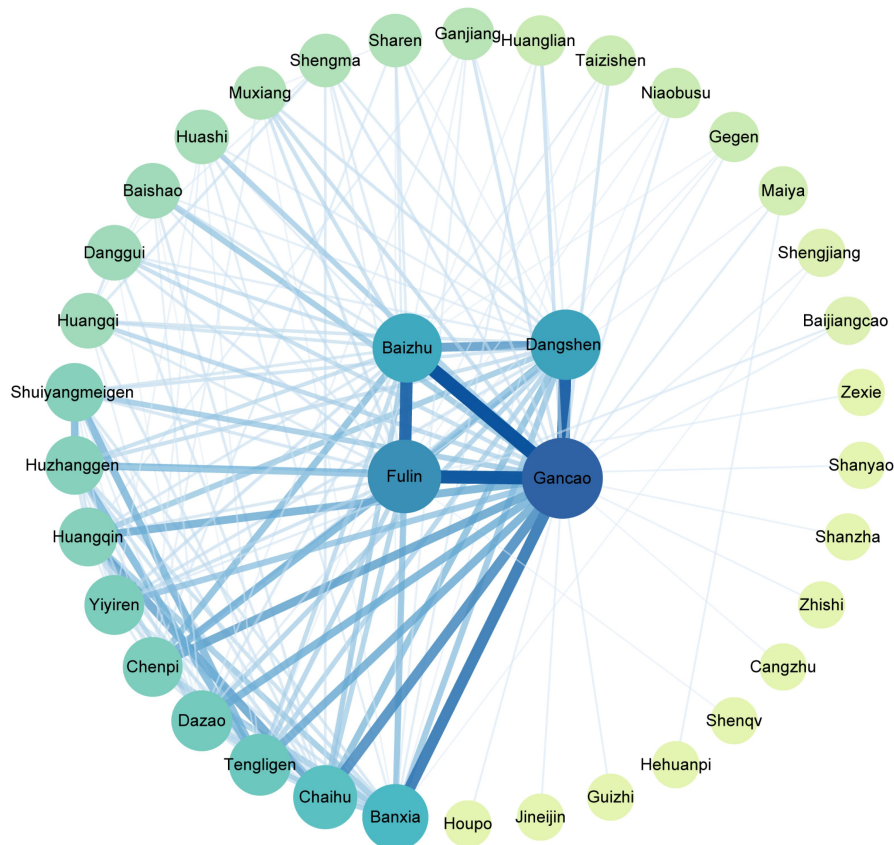


Figure 7 Complex network analysis of high-frequency herbs

Cluster 11: The combination of Huangbo (*Phellodendri chinensis cortex*) and Shengdi (*Rehmanniae radix*) can clear heat and nourish yin.

### Association rule and complex network analysis

The core herbal combination can be mined from prescriptions using the results of association rule and complex network. A total of 42 association rules were obtained. The results revealed that Fuling (*Poria*)-Gancao (*Glycyrrhizae radix*), Fuling (*Poria*)-Baizhu (*Atractylodis macrocephalae rhizome*), Baizhu (*Atractylodis macrocephalae rhizome*)-Gancao (*Glycyrrhizae radix*), and Dangshen (*Codonopsis radix*)-Gancao (*Glycyrrhizae radix*) had support over 40% (Table 3). The relationship among herbs was converted into a scatter diagram of all rules, using the X-axis for support degree, the Y-axis for confidence level, and the Z-axis for lift value (Figure 6). Figure 7 indicated that Fuling (*Poria*), Baizhu (*Atractylodis macrocephalus rhizome*), Dangshen (*Codonopsis radix*), and Gancao (*Glycyrrhizae radix*) were the core herbs for CRC treatment, consistent with the conclusion of frequency analysis. These four herbs constitute the Sijunzi decoction, which can replenish qi and strengthen the spleen.

### The interaction network between JSD and CRC targets

There were 64 chemical ingredients in JSD (Supplementary Table 1) and 11103 targets were associated with CRC (Supplementary Table 2). After screening, a total of 31 active ingredients (Table 4), 264 high-related targets of JSD, and 3250 differential targets in CRC (Supplementary Table 3) were obtained. Of these, 138 overlapped genes (Supplementary Table 3) were identified between JSD and CRC (Figure 8a). As shown in Figure 8b, the target-compound network was constructed by the overlapped genes and active ingredients of JSD. Furthermore, the overlapped targets were also imported into the STRING database to create a PPI network, which revealed the interaction between targets. After removing noninteraction proteins, there were 130 nodes and 258 edges in the network, representing 258 interactions among 130 targets. The average node degree value was 3.97 and the average local clustering coefficient was 0.461 (Figure 8c). CytoscapeV3.7.2 was used to calculate the network topological parameter, and identified the top 15 targets with BC values as AKT1, SRC, MAPK1, ESR1, HSP90AA1, ALB, RXRA, F2, TYMS, CTNNA1, CYP19A1, EGFR, CAT, PLG, and PLAU (Figure 8d).

### GO enrichment and KEGG pathway analysis

The overlapped genes between JSD and CRC were analyzed for GO enrichment and KEGG pathway using the Metascape database. The top 20 results are presented in Figure 8 (P < 0.01). The BP analysis indicated that the key targets of JSD acting on CRC involved in multiple processes, including response to hormone, cellular response to hormone stimulus, cellular response to lipid, cellular response to organic cyclic compound, response to steroid hormone, cellular response to steroid hormone stimulus, and hormone-mediated signaling pathway (Figure 9a). In CC analysis, the key targets were concentrated in vesicle lumen, secretory granule lumen, cytoplasmic the vesicle lumen, ficolin-1-rich granule lumen, ficolin-1-rich granule, membrane raft, membrane microdomain, and plasma membrane raft (Figure 9b). The significant results of MF analysis included protein kinase activity, phosphotransferase activity, alcohol group as acceptor, kinase activity, protein serine/threonine kinase activity, protein serine kinase activity, endopeptidase activity, peptidase activity, and serine-type peptidase activity (Figure 9c). The KEGG pathways were mainly involved in pathways in cancer, chemical carcinogenesis-reactive oxygen species, relaxin signaling pathway, colorectal cancer, IL-17 signaling pathway, prolactin signaling pathway, epithelial cell signaling in *Helicobacter pylori* infection, T cell receptor signaling pathway, and Th17 cell differentiation (Figure 9d).

### Discussion

Complementary and alternative medicine has made significant

advances in recent decades, both in theoretical research and clinical practice. Traditional Chinese medicine, which has been practiced for thousands of years and encompasses a wide variety of schools and theories, is widely accepted as a mainstream form of complementary and alternative therapy for cancer in Asian countries, particularly China. However, the subjectivity of TCM diagnosis and treatment, as well as the complexity of prescription compatibility, have limited the inheritance of TCM clinical experience. Furthermore, with the digital collection of clinical data and the establishment of a large number of high-quality medical public databases, TCM research faces the dilemma of rich data but poor knowledge. Data mining technology can assist in exploring the core connections between disease, diagnosis, and treatment, as well as the inner rules hidden under the fuzzy information, thereby playing an important role in the summary, transmission, and application of TCM.

In this study, we conducted a frequency analysis of TCM medication for the treatment of colorectal cancer. The top three herbs were Gancao (*Glycyrrhizae radix*), Fuling (*Poria*), and Baizhu (*Atractylodis macrocephalae rhizome*). Gancao has the effect of tonifying the spleen, benefiting qi, clearing heat, and detoxifying toxins, which is widely used to harmonize the effects of various herbs. Both Fuling and Baizhu can expel water and dampness, fortify the spleen and calm the heart. All three herbs have a spleen tonifying effect, which is in line with traditional Chinese medicine's view that the primary cause of CRC is spleen deficiency [31]. Modern medicine has also identified spleen deficiency as a pivotal pathogenesis in the development of CRC [32]. The spleen is often damaged due to deficiencies in the body at birth, unhealthy lifestyle, or long-term chronic intestinal diseases, leading to malfunctions in the production and transport of nutrients. This weakens vital qi (qi is the most basic substance that makes up the human body and sustains its vital activities), causes stagnation of blood, qi, as well as production of fire, dampness, and toxins, and has a significant impact on the occurrence and development of CRC. Pharmacological research confirmed that the *G.uralensis* polysaccharides in Gancao had an antitumor and immunomodulatory ability by up-regulating the expression of the IL-7 gene and immune cytokines [33]. *Poria* could inhibit angiogenesis via the downregulation of NF- $\kappa$ B and NF- $\kappa$ B/Rel translocation to potentiate immune response and inhibit tumor growth [34]. *Atractylenolide III* induced apoptosis of CRC cells by regulating the Bax/Bcl-2 signaling pathway [35].

The nature and taste theory and the five elements theory are vital components of traditional Chinese medicine, and have been widely studied in modern research [36, 37]. Frequency analysis showed that the application frequency of herbs with a warm nature was the highest, and the taste of herbs was mainly sweet, pungent, and bitter. The meridian tropism of herbs mainly belonged to the spleen, lungs, and stomach. In the TCM theory, the spleen has a mild nature and sweet taste, and is the hub of yin and yang operation. According to the theory of the five elements, fire generates soil, and the liver generates the spleen corresponding to the viscera. If the liver fails to nourish the spleen, it will lead to spleen deficiency and cold. And the deficiency of the spleen may produce dampness. Therefore, it is necessary to supplement the spleen with sweet-tasting herbs, which have the effect of tonifying, and promote its operation with warmth-nature herbs to dispel cold and dampness. Pungent-tasting herbs aid the upward and outward movement of qi (qi is the most basic substance that makes up the human body and sustains its vital activities), while bitter-tasting herbs facilitate downward and inward movement. The combination of pungent and bitter herbs helps the normal operation of qi, which is crucial for the normal function of the human body. The analysis of treatment strategies for CRC highlights the importance of strengthening the spleen and nourishing qi.

The etiology of CRC often appears as several symptom combinations. Cluster analysis resulted in formulas for composite syndromes. The herbs in Cluster 1, 2, 7, 9, 10, and 11 were for clearing heat, detoxifying, dispersing blood stasis, and removing dampness and were used to treat CRC patients with dampness and

Table 4 Active ingredients in JSD

| No. | Name                    | CAS        | Area    | Retention Time | Formula                                                       | Precursor Mass | Found At Mass | Mass Error (ppm) | Library Score | Isotope Ratio Difference |
|-----|-------------------------|------------|---------|----------------|---------------------------------------------------------------|----------------|---------------|------------------|---------------|--------------------------|
| 1   | Histidine               | 71-00-1    | 19280   | 1.04           | C <sub>6</sub> H <sub>9</sub> N <sub>3</sub> O <sub>2</sub>   | 156.077        | 156.0768      | 0.1              | 96.5          | 2.2                      |
| 2   | L-Carnitine             | 541-15-1   | 30240   | 1.13           | C <sub>7</sub> H <sub>15</sub> NO <sub>3</sub>                | 162.112        | 162.1125      | 0.1              | 90.8          | 0.6                      |
| 3   | Glutamic acid           | 56-86-0    | 91430   | 1.14           | C <sub>5</sub> H <sub>9</sub> NO <sub>4</sub>                 | 148.06         | 148.0605      | 0.3              | 97.9          | 0.4                      |
| 4   | Trigonelline            | 535-83-1   | 36590   | 1.2            | C <sub>7</sub> H <sub>7</sub> NO <sub>2</sub>                 | 138.055        | 138.055       | 0.4              | 86.5          | 0.8                      |
| 5   | Proline                 | 147-85-3   | 625400  | 1.23           | C <sub>5</sub> H <sub>9</sub> NO <sub>2</sub>                 | 116.071        | 116.0705      | -0.6             | 98.5          | 1.5                      |
| 6   | Nicotinic acid          | 59-67-6    | 19360   | 1.74           | C <sub>6</sub> H <sub>5</sub> NO <sub>2</sub>                 | 124.039        | 124.0393      | 0                | 95.7          | 0.6                      |
| 7   | Nicotinamide            | 98-92-0    | 19390   | 1.85           | C <sub>6</sub> H <sub>6</sub> N <sub>2</sub> O                | 123.055        | 123.0554      | 1.1              | 92.8          | 2.1                      |
| 8   | Phenylalanine           | 63-91-2    | 38590   | 3.1            | C <sub>9</sub> H <sub>11</sub> NO <sub>2</sub>                | 166.086        | 166.0863      | 0.5              | 99.8          | 1.5                      |
| 9   | Anisaldehyde            | 123-11-5   | 45450   | 3.13           | C <sub>8</sub> H <sub>8</sub> O <sub>2</sub>                  | 137.06         | 137.0598      | 0.4              | 76.6          | 0.9                      |
| 10  | Isoscapoletin           | 776-86-3   | 861800  | 4.48           | C <sub>10</sub> H <sub>8</sub> O <sub>4</sub>                 | 193.05         | 193.0494      | -1               | 97.6          | 3.2                      |
| 11  | Fraxetin                | 574-84-5   | 1431000 | 4.73           | C <sub>10</sub> H <sub>8</sub> O <sub>5</sub>                 | 209.044        | 209.0442      | -1               | 97.8          | 1.2                      |
| 12  | Epicatechin             | 490-46-0   | 495200  | 5.13           | C <sub>15</sub> H <sub>14</sub> O <sub>6</sub>                | 291.086        | 291.0862      | -0.3             | 97.6          | 4.3                      |
| 13  | Oxyresveratrol          | 29700-22-9 | 74690   | 5.52           | C <sub>14</sub> H <sub>12</sub> O <sub>4</sub>                | 245.081        | 245.0808      | -0.2             | 74.6          | 2.1                      |
| 14  | Resveratrol             | 501-36-0   | 2731000 | 7.15           | C <sub>14</sub> H <sub>12</sub> O <sub>3</sub>                | 229.086        | 229.0858      | -0.7             | 97.5          | 5.4                      |
| 15  | Polydatin               | 65914-17-2 | 480000  | 7.16           | C <sub>20</sub> H <sub>22</sub> O <sub>8</sub>                | 391.139        | 391.1387      | -0.2             | 96.7          | 8.1                      |
| 16  | Isofraxidin             | 486-21-5   | 29380   | 7.62           | C <sub>11</sub> H <sub>10</sub> O <sub>5</sub>                | 223.06         | 223.06        | -0.3             | 89.4          | 0.3                      |
| 17  | Glycetein               | 40957-83-3 | 711700  | 14.91          | C <sub>16</sub> H <sub>12</sub> O <sub>5</sub>                | 285.076        | 285.0755      | -0.7             | 78.1          | 7                        |
| 18  | Aspartic acid           | 56-84-8    | 52340   | 1.11           | C <sub>4</sub> H <sub>7</sub> NO <sub>4</sub>                 | 132.03         | 132.0304      | 1                | 89.8          | 1.4                      |
| 19  | L-Malic acid            | 97-67-6    | 662600  | 1.32           | C <sub>4</sub> H <sub>6</sub> O <sub>5</sub>                  | 133.014        | 133.0144      | 1.1              | 89.8          | 0.7                      |
| 20  | Gallic acid             | 149-91-7   | 450700  | 2.62           | C <sub>7</sub> H <sub>6</sub> O <sub>5</sub>                  | 169.014        | 169.0143      | 0.1              | 92.4          | 3.5                      |
| 21  | Androsin                | 531-28-2   | 22650   | 2.92           | C <sub>15</sub> H <sub>20</sub> O <sub>8</sub>                | 327.109        | 327.1086      | 0.2              | 87.7          | 7.2                      |
| 22  | Protocatechuic acid     | 99-50-3    | 42200   | 3.62           | C <sub>7</sub> H <sub>6</sub> O <sub>4</sub>                  | 153.019        | 153.0195      | 1                | 87.9          | 2.1                      |
| 23  | L-Tryptophan            | 73-22-3    | 11390   | 3.99           | C <sub>11</sub> H <sub>12</sub> N <sub>2</sub> O <sub>2</sub> | 203.083        | 203.0827      | 0.5              | 97.4          | 3.5                      |
| 24  | Protocatechuic Aldehyde | 139-85-5   | 68900   | 4.43           | C <sub>7</sub> H <sub>6</sub> O <sub>3</sub>                  | 137.024        | 137.0246      | 1.3              | 95            | 0.4                      |
| 25  | Esculetin               | 305-01-1   | 65240   | 4.88           | C <sub>9</sub> H <sub>6</sub> O <sub>4</sub>                  | 177.019        | 177.0195      | 0.8              | 96.5          | 4.1                      |
| 26  | Caffeic acid            | 331-39-5   | 1351000 | 4.96           | C <sub>9</sub> H <sub>8</sub> O <sub>4</sub>                  | 179.035        | 179.035       | 0.1              | 92.9          | 4.2                      |
| 27  | Catechin                | 154-23-4   | 1459000 | 5.13           | C <sub>15</sub> H <sub>14</sub> O <sub>6</sub>                | 289.072        | 289.0719      | 0.6              | 95.2          | 5.4                      |
| 28  | Anisic acid             | 100-09-4   | 22270   | 6.96           | C <sub>8</sub> H <sub>8</sub> O <sub>3</sub>                  | 151.04         | 151.0401      | 0.3              | 99.3          | 2                        |
| 29  | 3-Hydroxymorindone      | 80368-74-7 | 267800  | 14.88          | C <sub>15</sub> H <sub>10</sub> O <sub>6</sub>                | 285.04         | 285.0404      | -0.2             | 72            | 7.8                      |
| 30  | Physcion                | 521-61-9   | 705800  | 14.92          | C <sub>16</sub> H <sub>12</sub> O <sub>5</sub>                | 283.061        | 283.0611      | -0.5             | 99.6          | 8.6                      |
| 31  | Emodin                  | 518-82-1   | 2043000 | 17.82          | C <sub>15</sub> H <sub>10</sub> O <sub>5</sub>                | 269.046        | 269.0454      | -0.7             | 89.3          | 5.5                      |

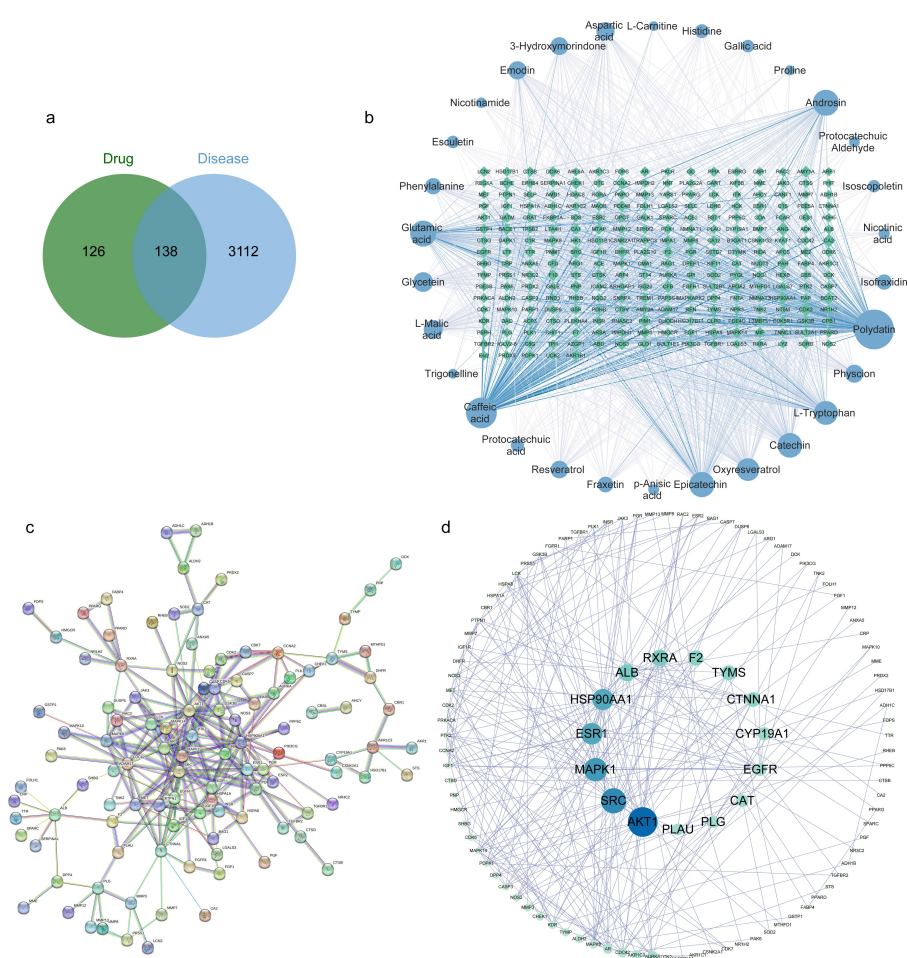


Figure 8 The interaction network between JSD and CRC targets

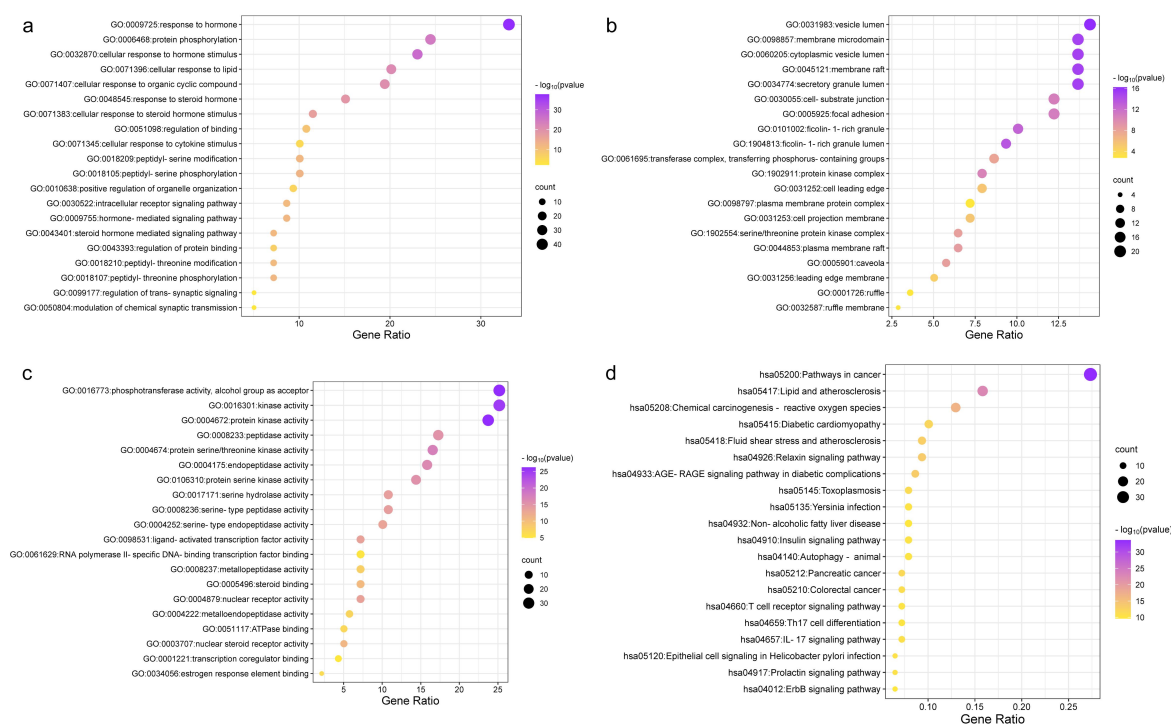


Figure 9 GO enrichment and KEGG pathway analysis

heat, caused by **qi deficiency** and blood stasis. Herbs in Cluster 3 and 5 are effective in regulating **qi** and blood, nourishing blood, regulating the liver, tranquilizing the mind, and harmonizing **Ying** and **Wei**. They were prescribed to patients with **qi** and blood disorders, **Ying** and **Wei** disharmony syndromes. Those in cluster 4 invigorate **qi**, as well as elevate **yang**, and were the core component of Buzhong Yiqi decoction, which is usually used to treat **qi deficiency** syndrome. The main indication of Cluster 6 is dyspepsia. The function of herbs in Cluster 8 was to calm the liver, activate blood circulation, and tonify the liver and kidney. The treatment methods of these clusters are consistent with the etiology and pathogenesis theory, diagnosis and therapy theory of CRC in TCM [38, 39], and provide useful ideas for clinical treatment.

Association rule analysis was proposed by Agrawal et al. [40] to explore meaningful associations among a large number of data items. Apriori algorithm is one of the most influential algorithms in it. Our results indicated that the most commonly used herbal combinations for CRC treatment were Fuling (*Poria*)-Gancao (*Glycyrrhizae radix*), Fuling (*Poria*)-Baizhu (*Atractylodis macrocephalae rhizome*), Baizhu (*Atractylodis macrocephalae rhizome*)-Gancao (*Glycyrrhizae radix*), and Dangshen (*Codonopsis radix*)-Gancao (*Glycyrrhizae radix*). The complex network is a network with a complex correlation relationship that is constructed by abstracting the internal elements of a complex system as nodes and the relationship between elements as edges, through attributes such as node degree, degree distribution, average distance, and cluster coefficient [41]. By filtering out other complex information and preserving the internal organizational structure as well as the association of the system, the complex network highlights the internal characteristics and properties of complex systems. A high correlation was found between Fuling (*Poria*), Baizhu (*Atractylodis macrocephalae rhizoma*), Dangshen (*Codonopsis radix*), and Gancao (*Glycyrrhizae radix*). These four herbs make up the Sijunzi decoction, a classic prescription for spleen's **qi deficiency** syndrome in TCM. It is derived from the *Preions of the Bureau of Taiping People's Welfare Pharmacy*, and has the function of plenishing **qi** of the spleen and removing dampness. Network pharmacology and experimental validation revealed that Sijunzi decoction promoted PI3K/Akt/mTOR pathway-mediated apoptosis and autophagy in CRC cells [42]. Furthermore, modified Sijunzi therapy was found to prevent CRC liver metastasis by activating the innate immune system [43].

In our study, Buzhong Yiqi decoction was also identified as a core prescription for CRC treatment. This decoction is from the *spleen and stomach theory*, and consists of eight herbs, namely Huangqi (*Astragali radix*), Renshen (*Ginseng radix*), Danggui (*Angelicae sinensis radix*), Gancao (*Glycyrrhizae radix*), Baizhu (*Atractylodis macrocephalae rhizoma*), Shengma (*Cimicifugae rhizoma*), Chenpi (*Citri reticulatae pericarpium*), and Chaihu (*Bupleuri radix*). Huangqi, Gancao, and Renshen replenish **qi** (**qi is the most basic substance that makes up the human body and sustains its vital activities**); Baizhu invigorates the spleen, and nourishes **qi** as well as blood; Danggui invigorates the blood and promotes its circulation; The function of Chenpi is invigorating **qi** and preventing stagnation; Chaihu and Shengma can elevate **qi**; Gancao harmonizes the effects of the other herbs. The combined use of these eight drugs has the effects of tonifying **qi**, lifting depression, and warming. A protocol of randomized controlled trial has shown that the use of this decoction could improve cancer-related fatigue and immunity in cervical carcinoma patients [44]. The combination of Buzhong Yiqi decoction with chemotherapy could enhance the body's immunity, reduce tumor indicators, improve postoperative **qi** and blood weakness, and other symptoms in patients with CRC, effectively reduce some adverse reactions of chemotherapy, and improve the life quality of patients [45, 46].

Association rule and complex network analysis also demonstrated a high correlation among Tengligen (*Radix Actinidiae Argutae*), Huzhanggen (*Polygoni cuspidati rhizoma*), and Shuiyangmeigen (Root of *Adina rubella*), which constituting Jiedu Sangen decoction. In traditional Chinese medicine theory, the combination of blood stasis and toxin leads to the occurrence and development of gastrointestinal

tumors [47]. Therefore, in addition to nourishing vital **qi**, dispelling pathogenic factors is also an important principle for treating tumors. Jiedu Sangen decoction is a representative formula for treating CRC from the perspective of dispelling pathogenic factors, which has various effects, such as clearing heat and toxic substances, eliminating dampness, reducing phlegm, activating blood circulation, and dispersing blood stasis. Modern research has found that Jiedu Sangen decoction inhibited the invasion and metastasis of colorectal cancer cell SW480 via the Hippo Signaling Pathway [48]. Moreover, the combination of Jiedu Sangen decoction and 5-fluorouracil could regulate the expression of caspase family and glycolysis-related proteins through the PI3K/AKT/HIF-1 $\alpha$  signaling pathway, inhibit glycolysis, induce apoptosis in tumor cells, and reduce the chemoresistance of colorectal cancer cells to 5-fluorouracil, thereby improving the efficacy of chemotherapy [49].

The effect of JSD in treating CRC has been clinically validated, and experimental studies have confirmed its anticancer effect, but there is limited research on the relevant mechanisms, which limits its clinical application. Therefore, we conducted network pharmacology analysis on CRC to reveal its possible mechanism. Network pharmacology is a powerful tool that can show the key compounds and mechanisms of TCM in disease treatment, and its strategy is in line with the holistic philosophy of TCM [24]. 31 active ingredients and 264 targets of JSD were found to be closely related to the pharmacodynamic activities of CRC. Among them, polydatin, caffeic acid, glutamic acid, androcin, and epicatechin were identified as the core compounds. Polydatin, a polyphenol extracted from the *Polygoni cuspidati rhizoma*, has been reported to exhibit antioxidant and anti-inflammatory effects and regulate autophagy and apoptosis [50]. It alone or in combination with human mononuclear cells (MNCs) shows antiproliferative and proapoptotic effects on CRC cells by inhibiting the inflammation and metastasis linked to the tumor microenvironment (TME) [51]. Caffeic acid can inhibit the self-renewal and radio-resistance of colorectal cancer stem cells via PI3k/AKT signaling pathway [52]. Glutamic acid promotes apoptosis in gastric cancer cells [53], which may be related to taurine metabolism [54]. ROS-induced DNA damage, activation of oncogenes or inactivation of tumor-suppressor genes are important mechanisms for the development of CRC [55]. Androcin was found to have antioxidant activity and may play an important role in the treatment of oxidative stress [56]. (-)-epicatechin, a novel anticancer drug, has a synergistic antiproliferative effect with panaxadiol on HCT-116 colorectal cancer cells [57]. AKT1, SRC, and MAPK1 were the core targets in the PPI network and the overexpression of them has been associated with various human malignancies, making them important inhibitor targets for oncology treatment. AKT1 is one of the AKT isoform and has significant effects on cell proliferation, apoptosis, tumor angiogenesis, and cancer metastasis [58]. The dysregulation of SRC is associated with malignant transformation, such as growth, invasion, and migration [59], while MAPK1 is involved in mitophagy [60], cell proliferation [61], and drug resistance [62].

Functional enrichment analyses were used to determine the potential pathways of JSD in treating CRC. KEGG enrichment analysis showed that JSD could modulate multiple signaling pathways, including relaxin signaling pathway, IL-17 signaling pathway, prolactin signaling pathway, and T cell receptor signaling pathway. The relaxin signaling pathway can induce cell invasion by reducing the expression of N- and E-cadherin through the Wnt pathway [63]. The IL-17 signaling pathway takes part in various human diseases, including inflammation, cancer, and metabolism-modulating. And there is a strong association between chronic inflammation and colorectal tumorigenesis [64]. Moreover, research has found that IL-17 and TNF $\alpha$  can cooperatively promote the survival and proliferation of CRC cells by stimulating glycolysis and growth factor production [65]. The upregulation of the prolactin signaling pathway has been reported not only in hormone-dependent cancers, but also in colon cancer development. Neradugomma N K et al. [66] found that prolactin signaling can enhance colon cancer stemness, which is associated with cellular resistance, tumor recurrence, and metastasis.

In addition to the relationship with immune diseases, study has also found that the T cell receptor (TCR) signaling pathway is closely related to tumors, and the TCR-based therapy is widely applied in cancer immunotherapy, including adoptive cellular therapy, checkpoint blockade, tumor microenvironment regulation, and vaccines [67]. Therefore, JSD may affect the development of CRC by regulating inflammation, immunity, metabolism, and hormone through the above pathways.

Based on data mining, our study found that spleen deficiency is the primary cause of CRC, and the basic principle of therapy is to strengthen the spleen and nourish qi. Cluster analysis revealed the diagnosis and therapy theory of CRC could be divided into two categories. One is to strengthen the body through nourishing and promoting the proper functioning of qi and blood, especially in the spleen and liver, while the other is to eliminate evils, including heat, dampness, poison, and dispersing blood stasis. The core prescriptions for CRC treatment are Sijunzi decoction, Buzhong Yiqi decoction, and Jiedu Sangen decoction. Sijunzi decoction and Buzhong Yiqi decoction mainly use sweet and warm herbs to nourish the body, while Jiedu Sangen decoction represents the principle of eliminating evils. Network pharmacology further revealed the active ingredients, key targets and mechanisms of JSD in CRC treatment. There are still some limitations in our study, such as not analyzing the medication rule of TCM in different stages and symptoms of CRC, and only collecting cases from one hospital, which lack the support of multicenter data. Therefore, large-scale clinical and pharmacodynamic experiments are needed for further exploration and study.

## Conclusions

This study summarized the medication regularity of TCM in the treatment of CRC, providing data support and guidance for clinical therapy and improving our understanding of CRC pathogenesis from the perspective of Chinese medicine. The data mining results showed that herbs with nature of warm, taste of sweet, and meridian tropism of the spleen were most frequently used in treating CRC. The primary principle of CRC therapy was to strengthen the spleen and nourish qi, and the core treatment method was to strengthen the body and eliminate evils. The core prescriptions were Sijunzi decoction and Buzhong Yiqi decoction, which can nourish the qi, blood, and spleen, and Jiedu Sangen decoction, which can clear heat and toxins. Network pharmacology revealed that the mechanism of Jiedu Sangen decoction in the CRC treatment mainly involved core compounds of polydatin, caffeic acid, glutamic acid, androsterone, and epicatechin, core targets of AKT1, SRC, MAPK1, through relaxin signaling pathway, IL-17 signaling pathway, prolactin signaling pathway, and T cell receptor signaling pathway.

## References

- Sung H, Ferlay J, Siegel RL, et al. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA A Cancer J Clin* 2021;71(3):209–249. Available at: <http://doi.org/10.3322/caac.21660>
- Arnold M, Sierra MS, Laversanne M, Soerjomataram I, Jemal A, Bray F. Global patterns and trends in colorectal cancer incidence and mortality. *Gut* 2016;66(4):683–691. Available at: <http://doi.org/10.1136/gutjnl-2015-310912>
- Li N, Lu B, Luo C, et al. Incidence, mortality, survival, risk factor and screening of colorectal cancer: A comparison among China, Europe, and northern America. *Cancer Lett* 2021;522:255–268. Available at: <http://doi.org/10.1016/j.canlet.2021.09.034>
- Patel SG, Karlitz JJ, Yen T, Lieu CH, Boland CR. The rising tide of early-onset colorectal cancer: a comprehensive review of epidemiology, clinical features, biology, risk factors, prevention, and early detection. *Lancet Gastroenterol Hepatol* 2022;7(3):262–274. Available at: [http://doi.org/10.1016/S2468-1253\(21\)00426-X](http://doi.org/10.1016/S2468-1253(21)00426-X)
- O’Gorman C, Stack J, O’Ceilleachair A, et al. Colorectal cancer survivors: an investigation of symptom burden and influencing factors. *BMC Cancer* 2018;18(1):1022. Available at: <http://doi.org/10.1186/s12885-018-4923-3>
- Rutherford C, Müller F, Faiz N, King MT, White K. Patient-reported outcomes and experiences from the perspective of colorectal cancer survivors: meta-synthesis of qualitative studies. *J Patient Rep Outcomes* 2020;4(1):27. Available at: <http://doi.org/10.1186/s41687-020-00195-9>
- Denlinger CS, Barsevick AM. The Challenges of Colorectal Cancer Survivorship. *J Natl Compr Canc Netw* 2009;7(8):883–894. Available at: <http://doi.org/10.6004/jnccn.2009.0058>
- Sun Z, Adam MA, Kim J, et al. Determining the Optimal Timing for Initiation of Adjuvant Chemotherapy After Resection for Stage II and III Colon Cancer. *Dis Colon Rectum* 2016;59(2):87–93. Available at: <http://doi.org/10.1097/DCR.0000000000000518>
- Liu N, Wu C, Jia R, et al. Traditional Chinese Medicine Combined With Chemotherapy and Cetuximab or Bevacizumab for Metastatic Colorectal Cancer: A Randomized, Double-Blind, Placebo-Controlled Clinical Trial. *Front Pharmacol* 2020;11:478. Available at: <http://doi.org/10.3389/fphar.2020.00478>
- Wang Q, Jiao L, Wang S, et al. Adjuvant Chemotherapy with Chinese Herbal Medicine Formulas Versus Placebo in Patients with Lung Adenocarcinoma after Radical Surgery: a Multicenter, Randomized, Double-Blind, Placebo-Controlled Trial. *Biol Proced Online* 2020;22:5. Available at: <http://doi.org/10.1186/s12575-020-00117-5>
- Sun Q, He M, Zhang M, et al. Traditional Chinese Medicine and Colorectal Cancer: Implications for Drug Discovery. *Front Pharmacol* 2021;12:685002. Available at: <http://doi.org/10.3389/fphar.2021.685002>
- Shao S, Jia R, Zhao L, et al. Xiao-Chai-Hu-Tang ameliorates tumor growth in cancer comorbid depressive symptoms via modulating gut microbiota-mediated TLR4/MyD88/NF- $\kappa$ B signaling pathway. *Phytomedicine* 2021;88:153606. Available at: <http://doi.org/10.1016/j.phymed.2021.153606>
- Yang Y, Zuo HQ, Li Z, et al. Comparison of efficacy of simo decoction and acupuncture or chewing gum alone on postoperative ileus in colorectal cancer resection: a randomized trial. *Sci Rep* 2017;7:37826. Available at: <http://doi.org/10.1038/srep37826>
- Jie Y, He W, Yang X, Chen W. Krüppel-like factor 4 acts as a potential therapeutic target of Sijunzi decoction for treatment of colorectal cancer. *Cancer Gene Ther* 2017;24(9):361–366. Available at: <http://doi.org/10.1038/cgt.2017.25>
- Lv J, Jia Y, Li J, et al. Gegen Qinlian decoction enhances the effect of PD-1 blockade in colorectal cancer with microsatellite stability by remodelling the gut microbiota and the tumour microenvironment. *Cell Death Dis* 2019;10(6):415. Available at: <http://doi.org/10.1038/s41419-019-1638-6>
- Chen Y, Liu JT, Zhu Y. The Overview in Traditional Chinese Medicine Syndrome Differentiation and Treatment of Colorectal Cancer. *J China Cancer* 2015;24(4):319–324. Chinese Available at: <http://doi.org/10.11735/j.issn.1004-0242.2015.04.A014>
- Cao Y, Liu ZH, Chen ZJ. Professor Chen Ruishen’s Experience in the Treatment of Colorectal Cancer. *J Chin Archives Traditional Chin Med* 2005;23(10):1750–1751. Chinese Available at: <http://doi.org/10.13193/j.archctm.2005.10.16.caoy.006>
- Zhang YM. Bo Liansong’s experience in treating large intestine cancer by supporting health Qi and eliminating pathogenic factors. *Shanghai J Tradit Chin Med* 2005;39(9):29. Chinese

Available at:

<http://doi.org/10.16305/j.1007-1334.2005.09.015>

19. Zhang QC, Cheng HB, Yu CT, et al. Effect of Xianlian Jiedu Formula on Intestinal Cell Types in Colorectal Cancer Model Mice with Damp-heat Stasis Toxin Pattern. *J Tradit Chin Med* 2022;63(05):461–467. Chinese Available at: <http://doi.org/10.13288/j.11-2166/r.2022.05.012>
20. Zhang S, Shi L, Mao D, et al. Use of Jianpi Jiedu Herbs in Patients with Advanced Colorectal Cancer: A Systematic Review and Meta-Analysis. *Evid Based Complement Alternat Med* 2018;2018:1–13. Available at: <http://doi.org/10.1155/2018/6180810>
21. Wu WT, Li YJ, Feng AZ, et al. Data mining in clinical big data: the frequently used databases, steps, and methodological models. *Mil Med Res* 2021;8(1):44. Available at: <http://doi.org/10.1186/s40779-021-00338-z>
22. Fang J, Li J. Research on Classification of Primary Liver Cancer Syndrome Based on Data Mining Technology. *J Healthc Eng* 2022;2022:1–14. Available at: <http://doi.org/10.1155/2022/2629509>
23. Luo W, Ding R, Guo X, et al. Clinical data mining reveals Gancao-Banxia as a potential herbal pair against moderate COVID-19 by dual binding to IL-6/STAT3. *Comput Biol Med* 2022;145:105457. Available at: <http://doi.org/10.1016/j.combiomed.2022.105457>
24. Zhang R, Zhu X, Bai H, Ning K. Network Pharmacology Databases for Traditional Chinese Medicine: Review and Assessment. *Front Pharmacol* 2019;10:123. Available at: <http://doi.org/10.3389/fphar.2019.00123>
25. Zhou W, Zhang H, Wang X, et al. Network pharmacology to unveil the mechanism of Muluodan in the treatment of chronic atrophic gastritis. *Phytomedicine* 2022;95:153837. Available at: <http://doi.org/10.1016/j.phymed.2021.153837>
26. Zhou W, Yang K, Zeng J, et al. FordNet: Recommending traditional Chinese medicine formula via deep neural network integrating phenotype and molecule. *Pharmacol Res* 2021;173:105752. Available at: <http://doi.org/10.1016/j.phrs.2021.105752>
27. Li Y, Zhang Y, Wang Y, et al. A strategy for the discovery and validation of toxicity quality marker of Chinese medicine based on network toxicology. *Phytomedicine* 2019;54:365–370. Available at: <http://doi.org/10.1016/j.phymed.2018.01.018>
28. Commission CP. *Chinese Pharmacopoeia China Medical Science Press*. Published November 28th, 2020. [https://www.gov.cn/xinwen/2020-11/28/content\\_5565608.htm](https://www.gov.cn/xinwen/2020-11/28/content_5565608.htm)
29. Zhong GS, Yang BC. *Traditional Chinese Pharmacology*. Beijing, China: China Press of Traditional Chinese Medicine; 2021.
30. Hao LQ. Analysis of data mining algorithm based on association rules. *Journal. J Taiyuan Univ (Nat Sci)* 2020;38(03):42–45. Chinese Available at: <http://doi.org/10.14152/j.cnki.2096-191X.2020.03.009>
31. Song Z, Yang YF, Sun LY, et al. Discussing theory and practice of spleen deficiency as the core pathogenesis in gastrointestinal cancer. *World Chin Med* 2021;16(09):1357–1363 + 1371. Chinese Available at: [https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7iyRpms2pqwbFRRUtoUlmHVveHXijLx\\_Kd\\_14VgKilcpIL8-HH-AVEro1FT4fsl7d&uniplatform=NZKPT](https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7iyRpms2pqwbFRRUtoUlmHVveHXijLx_Kd_14VgKilcpIL8-HH-AVEro1FT4fsl7d&uniplatform=NZKPT)
32. Sun X, Lin X, Diao J, Yu Z, Li K. Pi (Spleen)-deficiency syndrome in tumor microenvironment is the pivotal pathogenesis of colorectal cancer immune escape. *Chin J Integr Med* 2015;22(10):789–794. Available at: <http://doi.org/10.1007/s11655-015-2086-5>
33. Ayeka PA, Bian Y, Mwitari PG, et al. Immunomodulatory and anticancer potential of Gan cao (Glycyrrhiza uralensis Fisch.) polysaccharides by CT-26 colon carcinoma cell growth inhibition and cytokine IL-7 upregulation in vitro. *BMC Complement Altern Med* 2016;16:206. Available at: <http://doi.org/10.1186/s12906-016-1171-4>
34. Ríos JL. Chemical Constituents and Pharmacological Properties of *Poria cocos*. *Planta Med* 2011;77(07):681–691. Available at: <http://doi.org/10.1055/s-0030-1270823>
35. Zhang D, Li X, Song D, et al. Atractylenolide III induces apoptosis by regulating the Bax/Bcl-2 signaling pathway in human colorectal cancer HCT-116 Cells in vitro and in vivo. *Anticancer Drugs* 2022;33(1):30–47. Available at: <http://doi.org/10.1097/CAD.0000000000001136>
36. Li WL, Zhang XL, Sui F, Dai L, Huo HR, Jiang TL. Study Progress on Natures and Tastes of Chinese Herbs. *Chin J Exp Tradit Med Formulae* 2015;21(12):227–230. Chinese Available at: <http://doi.org/10.13422/j.cnki.syfx.2015120227>
37. Wen L, Liu W, Gu ZS, Tao XG, Yan ZA, Huang X. The Research Development of Yin and Yang and the Five Elements of Traditional Chinese Medicine in Recent Ten Years. *China J Tradit Chin Med* 2009;24(11):1481–1485. Chinese Available at: [https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7ir5D84hng\\_y4D11vwp0rrtWNF4Wz-M1s\\_p6ybXR2GuHk6O8clFiW2TpGh2RQfviKAM&uniplatform=NZKPT](https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7ir5D84hng_y4D11vwp0rrtWNF4Wz-M1s_p6ybXR2GuHk6O8clFiW2TpGh2RQfviKAM&uniplatform=NZKPT)
38. Feinberg J, Nielsen EE, Korang SK, et al. Nutrition support in hospitalised adults at nutritional risk. *Cochrane Database Syst Rev* 2017;5(5):CD011598. Available at: <http://doi.org/10.1002/14651858.CD011598.pub2>
39. Chen JB, Chai KQ, Chen M, Zou L, Du Y X, Zhang XY. Academic thought and clinical experience of CHAI Ke-qun in the differentiation and treatment of colonic and rectal carcinoma. *China J Tradit Chin Med Pharm* 2015;30(11):3956–3959. Chinese Available at: [https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7ir5D84hng\\_y4D11vwp0rrtWNF4Wz-M1s\\_p6ybXR2GuHk6O8clFiW2TpGh2RQfviKAM&uniplatform=NZKPT](https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7ir5D84hng_y4D11vwp0rrtWNF4Wz-M1s_p6ybXR2GuHk6O8clFiW2TpGh2RQfviKAM&uniplatform=NZKPT)
40. Agrawal R, Imieliński T, Swami A. Mining association rules between sets of items in large databases. *ACM SIGMOD Rec* 1993;22(2):207–216. Available at: <http://doi.org/10.1145/170036.170072>
41. Xia P, Gao K, Xie J, et al. Data Mining-Based Analysis of Chinese Medicinal Herb Formulae in Chronic Kidney Disease Treatment. *Evid Based Complement Alternat Med* 2020;2020:1–14. Available at: <http://doi.org/10.1155/2020/9719872>
42. Shang L, Wang Y, Li J, et al. Mechanism of Sijunzi Decoction in the treatment of colorectal cancer based on network pharmacology and experimental validation. *J Ethnopharmacol* 2023;302:115876. Available at: <http://doi.org/10.1016/j.jep.2022.115876>
43. Zhou JY, Chen M, Wu CE, Zhuang YW, Chen YG, Liu SL. The modified Si-Jun-Zi Decoction attenuates colon cancer liver metastasis by increasing macrophage cells. *BMC Complement Altern Med* 2019;19(1):86. Available at: <http://doi.org/10.1186/s12906-019-2498-4>
44. Hu J, Li X, Fang Y, Peng J. Efficacy and safety of Buzhong Yiqi Decoction in improving cancer-related fatigue and immunity of cervical carcinoma patients: A protocol of randomized controlled trial. *Medicine (Baltimore)* 2021;100(49):e27938. Available at: <http://doi.org/10.1097/MD.00000000000027938>
45. Zhang C, Yang WJ. Clinical Observation the Recurrence of Colon Cancer after Treating with BuZhong YiQiTang Combined with Chemotherapy. *Western J Tradit Chin Med* 2011;24(07):73–74. Chinese Available at: [https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7ir5D84hng\\_y4D11vwp0rrtWNF4Wz-M1s\\_p6ybXR2GuHk6O8clFiW2TpGh2RQfviKAM&uniplatform=NZKPT](https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7ir5D84hng_y4D11vwp0rrtWNF4Wz-M1s_p6ybXR2GuHk6O8clFiW2TpGh2RQfviKAM&uniplatform=NZKPT)
46. Wang ZX, Li X. Clinical Study of Buzhong Yiqi Decoction

- Combined with Chemotherapy in the Treatment of Advanced Colorectal Cancer. *Shaanxi J Tradit Chin Med* 2020;41(10):1414–1417. Chinese Available at: [https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7i8oRR1PAr7RxjuAJk4dHXoqjOTjUtdo\\_W3lKx3uF3eqrE0Ux1hM5tTf2qghr3lnqY&uniplatform=NZKPT](https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7i8oRR1PAr7RxjuAJk4dHXoqjOTjUtdo_W3lKx3uF3eqrE0Ux1hM5tTf2qghr3lnqY&uniplatform=NZKPT)
47. LIU L, Zhang GJ, Xu CY, Zhang GS. Analysis of Jiedu Sengen Decoction on anti-gastrointestinal tumor effect by treating phlegmatic and toxin with same method. *China J Tradit Chin Med Pharm* 2018;33(11):4824–4826. Chinese Available at: [https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7i8oRR1PAr7RxjuAJk4dHXoqjOTjUtdo\\_W3lKx3uF3eqrE0Ux1hM5tTf2qghr3lnqY&uniplatform=NZKPT](https://kns.cnki.net/kcms2/article/abstract?v=3uoqIhG8C44YLtIOAiTRKibYIV5Vjs7i8oRR1PAr7RxjuAJk4dHXoqjOTjUtdo_W3lKx3uF3eqrE0Ux1hM5tTf2qghr3lnqY&uniplatform=NZKPT)
  48. Yuan L, Zhou M, Wasan HS, et al. Jiedu Sengen Decoction Inhibits the Invasion and Metastasis of Colorectal Cancer Cells by Regulating EMT through the Hippo Signaling Pathway. *Evid Based Complement Alternat Med* 2019;2019:1–10. Available at: <http://doi.org/10.1155/2019/1431726>
  49. Sun LT, Zhang LY, Shan FY, Shen MH, Ruan SM. Jiedu Sengen decoction inhibits chemoresistance to 5-fluorouracil of colorectal cancer cells by suppressing glycolysis via PI3K/AKT/HIF-1 $\alpha$  signaling pathway. *Chin J Nat Med* 2021;19(2):143–152. Available at: [http://doi.org/10.1016/S1875-5364\(21\)60015-8](http://doi.org/10.1016/S1875-5364(21)60015-8)
  50. Ye P, Wu H, Jiang Y, et al. Old dog, new tricks: Polydatin as a multitarget agent for current diseases. *Phytother Res* 2022;36(1):214–230. Available at: <http://doi.org/10.1002/ptr.7306>
  51. De Gregorio A, Krasnowska EK, Zonfrillo M, et al. Influence of Polydatin on the Tumor Microenvironment In Vitro: Studies with a Colon Cancer Cell Model. *Int J Mol Sci* 2022;23(15):8442. Available at: <http://doi.org/10.3390/ijms23158442>
  52. Park SR, Kim SR, Hong IS, Lee HY. A Novel Therapeutic Approach for Colorectal Cancer Stem Cells: Blocking the PI3K/Akt Signaling Axis With Caffeic Acid. *Front Cell Dev Biol* 2020;8:585987. Available at: <http://doi.org/10.3389/fcell.2020.585987>
  53. Gu Y, Chen T, Fu S, et al. Perioperative dynamics and significance of amino acid profiles in patients with cancer. *J Transl Med* 2015;13:35. Available at: <http://doi.org/10.1186/s12967-015-0408-1>
  54. Halama A, Riesen N, Möller G, Hrabě de Angelis M, Adamski J. Identification of biomarkers for apoptosis in cancer cell lines using metabolomics: tools for individualized medicine. *J Intern Med* 2013;274(5):425–439. Available at: <http://doi.org/10.1111/joim.12117>
  55. Cheng Y, Ling Z, Li L. The Intestinal Microbiota and Colorectal Cancer. *Front Immunol* 2020;11:615056. Available at: <http://doi.org/10.3389/fimmu.2020.615056>
  56. Amin HIM, Hussain FHS, Najmaldin SK, et al. Phytochemistry and Biological Activities of Iris Species Growing in Iraqi Kurdistan and Phenolic Constituents of the Traditional Plant *Iris postii*. *Molecules* 2021;26(2):264. Available at: <http://doi.org/10.3390/molecules26020264>
  57. Rodriguez M, Du GJ, Wang CZ, Yuan CS. Letter to the Editor: Panaxadiol's Anticancer Activity is Enhanced by Epicatechin. *Am J Chin Med* 2010;38(6):1233–1235. Available at: <http://doi.org/10.1142/S0192415X10008597>
  58. Alwhaibi A, Verma A, Adil MS, Somanath PR. The unconventional role of Akt1 in the advanced cancers and in diabetes-promoted carcinogenesis. *Pharmacol Res* 2019;145:104270. Available at: <http://doi.org/10.1016/j.phrs.2019.104270>
  59. Mayer EL, Krop IE. Advances in Targeting Src in the Treatment of Breast Cancer and Other Solid Malignancies. *Clin Cancer Res* 2010;16(14):3526–3532. Available at: <http://doi.org/10.1158/1078-0432.CCR-09-1834>
  60. Hirota Y, Yamashita S, Kurihara Y, et al. Mitophagy is primarily due to alternative autophagy and requires the MAPK1 and MAPK14 signaling pathways. *Autophagy* 2015;11(2):332–343. Available at: <http://doi.org/10.1080/15548627.2015.1023047>
  61. Wei WT, Nian XX, Wang SY, et al. miR-422a inhibits cell proliferation in colorectal cancer by targeting AKT1 and MAPK1. *Cancer Cell Int* 2017;17:91. Available at: <http://doi.org/10.1186/s12935-017-0461-3>
  62. Li M, Cai O, Yu Y, Tan S. Paeonol inhibits the malignancy of Apatinib-resistant gastric cancer cells via LINC00665/miR-665/MAPK1 axis. *Phytomedicine* 2022;96:153903. Available at: <http://doi.org/10.1016/j.phymed.2021.153903>
  63. Fue M, Miki Y, Takagi K, et al. Relaxin 2/RXFP1 Signaling Induces Cell Invasion via the  $\beta$ -Catenin Pathway in Endometrial Cancer. *Int J Mol Sci* 2018;19(8):2438. Available at: <http://doi.org/10.3390/ijms19082438>
  64. Terzić J, Grivennikov S, Karin E, Karin M. Inflammation and Colon Cancer. *Gastroenterol* 2010;138(6):2101–2114.e5. Available at: <http://doi.org/10.1053/j.gastro.2010.01.058>
  65. Straus DS. TNF $\alpha$  and IL-17 cooperatively stimulate glucose metabolism and growth factor production in human colorectal cancer cells. *Mol Cancer* 2013;12:78. Available at: <http://doi.org/10.1186/1476-4598-12-78>
  66. Neradugomma NK, Subramaniam D, Tawfik OW, et al. Prolactin signaling enhances colon cancer stemness by modulating Notch signaling in a Jak2-STAT3/ERK manner. *Carcinogenesis* 2014;35(4):795–806. Available at: <http://doi.org/10.1093/carcin/bgt379>
  67. Shah K, Al-Haidari A, Sun J, Kazi JU. T cell receptor (TCR) signaling in health and disease. *Signal Transduct Target Ther* 2021;6(1):412. Available at: <http://doi.org/10.1038/s41392-021-00823-w>