Research Article

Network Pharmacology-Based Study on the Active Ingredients and Mechanism of Pan Ji Sheng Traditional Chinese Medicine Formula in the Treatment of Inflammation

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Background. Pan Ji Sheng Formula is a Chinese medicine formula that enables heat-free detoxification as well as anti-inflammatory and immune-boosting properties. This formula contains eight herbs. Its underlying mechanism is unknown. The bioactive ingredients were screened in our work, and the mechanism of this formula was investigated.

Methods. Using traditional Chinese medicine systems pharmacology database and analysis platform (TCMSP), ingredients in Pan Ji Sheng Chinese medicine formula were screened, and we selected the main bioactive ingredients for web-based research. The targets of bioactive ingredients are primarily obtained from the SwissTargetPrediction and TCMSP databases, and the text mining method is used. STRING and Cytoscape were then used to examine the protein-protein interaction (PPI) networks. To explore the biological function and related pathways, functional annotation and pathway analysis were performed.

Results. This research discovered 96 bioactive ingredients. Then, 215 potential targets of bioactive ingredients were screened. Through the analysis of the PPI network, we discovered 25 key target genes, which can be described as hub target genes regulated by bioactive ingredients. Bioactive ingredients primarily regulate CASP3, AKT1, JUN, and other proteins. The formula works synergistically to enhance immune response and anti-infection by regulating immune-related pathways, TNF signaling pathways, and apoptosis.

Conclusions. A variety of bioactive ingredients in the formula could play roles in regulating CASP3, AKT1, and other genes in immune, infection, apoptosis, and tumor-related signaling pathways. Our data point the way forward for future studies on the mechanism of action of this formula.

1. Introduction

The climate in China’s Lingnan region is standard subtropical. Summers are hot, rainy, as well as wet [1]. Furthermore, Cantonese people prefer to eat fried, dry, and hot foods. It is easy to make people “heat” and “dampness” due to the hot and humid climate, poor diet, and insufficient sleep [2, 3]. The symptoms of “heat” contain fever, thirst, sweating, fatigue, yellow urine, and yellow tongue. The common symptoms of “dampness” contain head pain, chest tightness, sluggishness, and sore or swollen joints. “Heat” and “dampness” are considered to be the cause of many inflammatory disease, cancer, and metabolic disorders [2].

Inflammation is a pathological defense response and it is also the most important protective response [4]. In modern western medicine, clinical experimental data show that the current conventional treatment for inflammation is anti-inflammatory drugs and antibiotic drugs [5, 6]. Nonsteroidal anti-inflammatory drugs (NSAIDs) are extensively used to reduce inflammation [7]. NSAIDs, such as aspirin and ibuprofen, are effective by inhibiting cyclooxygenase (COX) activity, thereby suppressing inflammatory responses [8]. Although it is effective, some anti-inflammatory drugs can lead to some side effects, such as gastrointestinal damage, gastrointestinal bleeding, and cardiovascular risk [9, 10]. The long-term use of antibiotic drugs can also lead to drug-resistance and seriously affect the treatment effect [11].

Traditional Chinese medicine (TCM) has the advantages of long efficacy and safety, so it is necessary to excavate the TCM compound formulas for treating inflammation.

The ancestors attempted to collect herbs for clearing heat and detoxification, and boiling water for drinking to...
eliminate the “heat” in order to get rid of dampness and heat and adapt to the environment. Since this type of herbal medicine was safe to drink, it gradually spread among the people [12, 13]. People gradually dig up various therapeutic properties of traditional Chinese medicine substances under the research of ancient and modern science, and make formulas with heat-clearing and detoxification features with honeysuckle, Scutellaria baicalensis, chrysanthemum, isatis root, and other traditional Chinese medicines, so as to enhance immune response and alleviate problems such as getting angry and heavy moisture caused by improper diet and lack of sleep [14, 15]. TCM (traditional Chinese medicine) is a type of traditional medicine. TCM is still a vital resource with such a long history. TCM can still influence the research of ancient and modern science, and make formulas with heat-clearing and detoxification features with honeysuckle, Scutellaria baicalensis, chrysanthemum, isatis root, and other traditional Chinese medicines, so as to enhance immune response and alleviate problems such as getting angry and heavy moisture caused by improper diet and lack of sleep [14, 15]. TCM (traditional Chinese medicine) is a type of traditional medicine. TCM is still a vital resource with such a long history. TCM can still influence the advancement of modern medicine [16, 17]. The Pan Ji Sheng formula, which contains eight different herbs, is the subject of this research: Micrococcus Foli (the leaves of Microcos paniculata), Polygonum chinense (creeping smartweed), Ecliptae Herba (false daisy), Perilla Frutescens (the leaves of Beefsteak Plant), Isatidis Radix (the dried roots of the plant Isatis indigotica Fort or Isatis tinctoria L.), Chrysanthemi Flos (the flower of Chrysanthemum indicum Linne or Chrysanthemum morifolium Ramatuelle), Glycyrrhiza uralensis (Chinese liquorice, the root of Glycyrrhiza uralensis), and Chimonanthus salicifolia (wintersweet). All of these herbs are commonly used to treat diseases by clinicians. According to published research, these Chinese herbal medicines can prevent and treat diseases by utilizing a wide range of chemical components and multiple targets [18–21]. For example, isatis root lectin can directly kill influenza viruses by blocking the expression of nuclear proteins of new influenza viruses [22]; at the same time, nucleoside components such as uridine, guanosine, and adenosine can interfere with the synthesis of viral nucleic acid and perform critical roles for influenza virus defense [23], and polysaccharides have immunomodulatory effects and play indirect roles for influenza virus defense [24].

There is, however, no systematic research report on the specific formula and network mechanism of the formula’s effects of clearing heat, detoxifying, anti-inflammatory, and enhancing immune response. Now, researchers have realized the “one key, one lock” model is insufficient for deciphering drug effects, particularly in complex diseases [25]. Network pharmacology is a new technology that uses the receptor theory and biological network technology to elucidate drug action mechanisms [26]. Its research mode of “multicomponent network target action” opens up a new research field and its compound prescriptions with multi-component and multitarget synergy [27]. Furthermore, the rapid development of biomedical data, such as the TCMSP (traditional Chinese medicine system pharmacology database and analysis platform), has facilitated such research [28]. As a result, web-based pharmacological analysis can provide us with a thorough understanding of the significance of each component, target, and pathway. Based on the research concept of traditional Chinese medicine’s multi-component and multitarget effect, this study explains the biological mechanism of clearing heat, detoxifying, anti-inflammatory, and enhancing immune response by using the network pharmacology technology and analyzing the target characteristics, biological function, and pathway of the Pan Ji Sheng formula. Our research provides a scientific basis for experimental research and product development.

2. Methods

2.1. Screening of Bioactive Ingredients. Through TCMSP, we search the relevant information about the bioactive ingredients in eight herbs in Pan Ji Sheng formula and screen the qualified compounds as the formula’s active ingredients. The screening conditions are oral bioavailability (OB) ≥ 30%, number of hydrogen bond donors (Hdon) < 5, lipid water partition coefficient (Alogp) < 5, number of hydrogen bond receptors (HACC) < 10, intestinal epithelial permeability (Caco-2) > 0, drug class (DL) ≥ 0.18, and drug half-life (HL) ≥ 4. We obtained bioactive ingredients of six herbs (Micrococcus Foli, Ecliptae Herba, Perilla Frutescens, Isatidis Radix, Chrysanthemi Flos, and Glycyrrhiza uralensis) from the TCMSP database. There is no information about Polygonum chinense and Chimonanthus salicifolia in the TCMSP database, so we search the literature for bioactive ingredients of these two herbs, then test OB ≥ 30% and DL ≥ 0.18 in TCMSP to determine the active ingredients.

2.2. Target Prediction of Bioactive Ingredients. The formula’s bioactive ingredients were imported to TCMSP to obtain information on ingredient-target interaction. Second, we use the Swiss Target Prediction online analysis tool to predict the active ingredient’s targets, screen potential targets, extract the names of the target genes, and build the chemical ingredient-target interaction network. The specific method is to convert all ingredients into standard smiles format and import the smiles format file into the Swiss Target Prediction online analysis platform [29], set the species to “Homo sapiens,” and set Probability ≥ 0.7, and export the target data in the CSV format.

The target genes were then imported to the UniProt database to confirm their gene names. Through computer research, this study obtained the list of target genes for the traditional Chinese medicine Pan Ji Sheng formula.

2.3. Construction of the Protein-Protein Interaction (PPI) Network. We import target genes into STRING [30] and set the species to “Homo sapiens (human)” and use a confidence level of 0.9 to build the target interaction network (PPI). We hide the discrete points in the network, then export the results to a TSV file and import it to Cytoscape 3.9.1 [31]. Cytoscape was then used to construct the target’s PPI network.

Then, in Cytoscape, the MCODE and Cytohubba plugins were used to extract the functional modules and top 25 hub genes of the PPI network, respectively.

2.4. Gene Ontology (GO) Functional Annotation and KEGG Pathway Analysis. All screened target genes were entered into the Metascape platform for enrichment analysis [32].
| Herbals                  | Molecule names                                                                 |
|-------------------------|--------------------------------------------------------------------------------|
| *Microctis folium*      | Isorhamnetin<br> Kaempferol<br> 4′,5-Dihydroxyflavone<br> Kaempferol<br> Quercetin |
| *Polygonum chinense*    | 3-O-Methylellagic acid<br> Kaempferol-7-O-glucoside<br> 3,3′-Di-O-methyllellagic acid<br> Protocatechuic acid<br> Isorhamnetin<br> Luteolin<br> Acacetin |
| *Ecliptae herba*        | Butin<br> 1,3,8,9-Tetrahydroxybenzofurano [3,2-c] chromen-6-one<br> 3′-O-Methylorobol<br> Pratensein<br> Demethylwedelolactone<br> Wedelolactone<br> Luteolin |
| *Perilla frutescens*    | Luteolin<br> Acacetin<br> Eupatorin<br> Dinatin<br> Quindoline<br> Hydroxyindirubin<br> Indigo<br> 2-(9-((3-Methyl-2-oxopent-3-en-1-yl) oxy)-2-oxo-1,2,8,9-tetrahydrofurano [2,3-h] quinolin-8-yl) propan-2-yl acetate<br> DFV<br> (E)-2-[(3-Indole) cyanomethylene]-3-indolinone<br> neohesperidin_qt<br> Sinensetin<br> 6-(3-Oxindolin-2-ylidene) indolo[2,1-b]quinazolin-12-one<br> (E)-3-(3,5-Dimethoxy-4-hydroxy-benzylidene)-2-indolinone<br> (E)-3-(3,5-Dimethoxy-4-hydroxy-benzylidene)-2-indolinone<br> 3-[(3,5-Dimethoxy-4-oxo-1-cyclohexa-2,5-dienylidene)methyl]-2,4-dihydro-1H-pyrrolo[2,1-b] quinazolin-9-one<br> [(1S,5S,7S)-7-Acetoxy-5-isopropenyl-2,8-dimethylene-cyclodecyl] acetate<br> Acacetin<br> Chryseryiol<br> Isorhamnetin |
| *Isatidis radix*        | Kaempferol<br> 5,7-Dihydroxy-2-(3-hydroxy-4-methoxyphenyl) chroman-4-one<br> Luteolin<br> Eupatorin<br> Diosmetin<br> Naringenin<br> Artemetin<br> Jaranol<br> Isorhamnetin<br> Formononetin |
| *Chrysanthemi flos*     |                                                                                   |
### Table 1: Continued.

| Herbals                  | Molecule names                                                                                                                                                                                                 |
|--------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
|                         | Calycosin                                                                                                                                                                                                       |
|                         | Kaempferol                                                                                                                                                                                                       |
|                         | Licochalcone a                                                                                                                                                                                                |
|                         | Inermine                                                                                                                                                                                                         |
|                         | DFV                                                                                                                                                                                                               |
|                         | Glycyrol                                                                                                                                                                                                         |
|                         | Medicarpin                                                                                                                                                                                                       |
|                         | Lupiwighteone                                                                                                                                                                                                  |
|                         | 7-Methoxy-2-methyl isoflavone                                                                                                                                                                                    |
|                         | Naringenin                                                                                                                                                                                                       |
|                         | Glyasperin B                                                                                                                                                                                                     |
|                         | Glyasperin F                                                                                                                                                                                                     |
|                         | Isotrifoliol                                                                                                                                                                                                    |
|                         | (E)-1-(2,4-Dihydroxyphenyl)-3-(2,2-dimethylchromen-6-yl) prop-2-en-1-one                                                                                                                                              |
|                         | (2S)-6-(2,4-Dihydroxyphenyl)-2-(2-hydroxypropan-2-yl)-4-methoxy-2,3-dihydrofuro [3,2-g] chromen-7-one                                                                                                                                               |
|                         | Semilicoisoflavone B                                                                                                                                                                                             |
|                         | Glepidotin A                                                                                                                                                                                                     |
|                         | Glepidotin B                                                                                                                                                                                                     |
|                         | Glypallichalcone                                                                                                                                                                                                |
|                         | 8-(6-Hydroxy-2-benzofuranyl)-2,2-dimethyl-5-chromenol                                                                                                                                                               |
|                         | Licochalcone B                                                                                                                                                                                                   |
|                         | Licochalcone G                                                                                                                                                                                                   |
|                         | Licoricone                                                                                                                                                                                                       |
|                         | Gancaonin A                                                                                                                                                                                                      |
|                         | Gancaonin B                                                                                                                                                                                                       |
|                         | 3-(3,4-Dihydroxyphenyl)-5,7-dihydroxy-8-(3-methylbut-2-enyl) chromone                                                                                                                                               |
|                         | 5,7-Dihydroxy-3-(4-methoxyphenyl)-8-(3-methylbut-2-enyl) chromone                                                                                                                                                 |
|                         | 2-(3,4-Dihydroxyphenyl)-5,7-dihydroxy-6-(3-methylbut-2-enyl) chromone                                                                                                                                               |
|                         | Licocoumarone                                                                                                                                                                                                   |
|                         | Licoisoflavone                                                                                                                                                                                                  |
|                         | Licoisoflavone B                                                                                                                                                                                                 |
|                         | Licoisoflavonane                                                                                                                                                                                                |
|                         | Shipterocarpin                                                                                                                                                                                                   |
| Licorice                  | (E)-3-[3,4-Dihydroxy-5-(3-methylbut-2-enyl)phenyl]-1-(2,4-dihydroxyphenyl) prop-2-en-1-one                                                                                                                                 |
|                         | Glyzaglabrin                                                                                                                                                                                                     |
|                         | Glabranin                                                                                                                                                                                                        |
|                         | Glabrone                                                                                                                                                                                                          |
|                         | 1,3-Dihydroxy-9-methoxy-6-benzofuran[3,2-c] chromenone                                                                                                                                                    |
|                         | 1,3-Dihydroxy-8,9-dimethoxy-6-benzofuran[3,2-c] chromenone                                                                                                                                                    |
|                         | Eurycarpin A                                                                                                                                                                                                     |
|                         | Sigmoidin-B                                                                                                                                                                                                       |
|                         | (2R)-7-Hydroxy-2-(4-hydroxyphenyl) chroman-4-one                                                                                                                                                                 |
|                         | (2S)-7-Hydroxy-2-(4-hydroxyphenyl)-8-(3-methylbut-2-enyl) chroman-4-one                                                                                                                                             |
|                         | Isoglycyrol                                                                                                                                                                                                       |
|                         | Isolicoflavonol                                                                                                                                                                                                 |
|                         | HMO                                                                                                                                                                                                               |
|                         | 1-Methoxyphaseollidin                                                                                                                                                                                            |
|                         | Quercetin der.                                                                                                                                                                                                   |
|                         | 6-Prenylated eriodictyol                                                                                                                                                                                          |
|                         | 7-Acetoxy-2-methylisoflavone                                                                                                                                                                                      |
|                         | 8-Prenylated eriodictyol                                                                                                                                                                                          |
|                         | Gancaonin G                                                                                                                                                                                                       |
|                         | Gancaonin H                                                                                                                                                                                                       |
|                         | Licoagrocarpin                                                                                                                                                                                                   |
|                         | Glyasperins M                                                                                                                                                                                                   |
|                         | Licoagroisoflavone                                                                                                                                                                                                |
|                         | Odoratin                                                                                                                                                                                                         |
|                         | Phaseol                                                                                                                                                                                                          |
|                         | Xambioona                                                                                                                                                                                                       |
| Chimonanthus salicifolius | Luteolin-5-O-glucoside                                                                                                                                                                                            |
|                         | Quercetin                                                                                                                                                                                                         |
|                         | Kaempferol                                                                                                                                                                                                       |
| No. | Target gene names | String Id   |
|-----|------------------|-------------|
| 1   | NOS2             | 9606.ENSP00000327251 |
| 2   | PTGS1            | 9606.ENSP00000354612  |
| 3   | ESR1             | 9606.ENSP00000405330  |
| 4   | AR               | 9606.ENSP0000036382   |
| 5   | PPARG            | 9606.ENSP00000287820  |
| 6   | PTGS2            | 9606.ENSP00000356438  |
| 7   | PTPN1            | 9606.ENSP00000360683  |
| 8   | ESR2             | 9606.ENSP00000343925  |
| 9   | DPP4             | 9606.ENSP00000353731  |
| 10  | MAPK14           | 9606.ENSP00000229795  |
| 11  | GSK3B            | 9606.ENSP00000324806  |
| 12  | HSP90AA1         | 9606.ENSP00000335153  |
| 13  | CDK2             | 9606.ENSP00000266970  |
| 14  | PIK3CG           | 9606.ENSP00000352121  |
| 15  | PKIA             | 9606.ENSP00000379696  |
| 16  | PRSS1            | 9606.ENSP00000308720  |
| 17  | PIM1             | 9606.ENSP00000362608  |
| 18  | CCNA2            | 9606.ENSP00000274026  |
| 19  | NCOA2            | 9606.ENSP00000399968  |
| 20  | CALM2            | 9606.ENSP00000272298  |
| 21  | PYGM             | 9606.ENSP00000164139  |
| 22  | PPARD            | 9606.ENSP00000310928  |
| 23  | CHEK1            | 9606.ENSP00000388648  |
| 24  | AKR1B1           | 9606.ENSP00000285930  |
| 25  | NCOA1            | 9606.ENSP00000385216  |
| 26  | F7               | 9606.ENSP00000364731  |
| 27  | F2               | 9606.ENSP00000308541  |
| 28  | NOS3             | 9606.ENSP00000297494  |
| 29  | AHC              | 9606.ENSP00000303211  |
| 30  | GABRA1           | 9606.ENSP00000393097  |
| 31  | MAOB             | 9606.ENSP00000367309  |
| 32  | GRIA2            | 9606.ENSP00000296526  |
| 33  | RELA             | 9606.ENSP00000384273  |
| 34  | XDH              | 9606.ENSP00000368727  |
| 35  | NCFI             | 9606.ENSP00000289473  |
| 36  | OLR1             | 9606.ENSP00000309124  |
| 37  | PGR              | 9606.ENSP00000325120  |
| 38  | CHRM1            | 9606.ENSP00000306490  |
| 39  | GABRA2           | 9606.ENSP00000421828  |
| 40  | SLC6A2           | 9606.ENSP00000219833  |
| 41  | CHRM2            | 9606.ENSP00000399745  |
| 42  | ADRA1B           | 9606.ENSP00000306662  |
| 43  | TOP2A            | 9606.ENSP00000411532  |
| 44  | IKBKB            | 9606.ENSP00000340684  |
| 45  | AKT1             | 9606.ENSP00000451828  |
| 46  | BCL2             | 9606.ENSP00000381185  |
| 47  | BAX              | 9606.ENSP00000293288  |
| 48  | CD40LG           | 9606.ENSP00000359663  |
| 49  | JUN              | 9606.ENSP00000360266  |
| 50  | AHS1A            | 9606.ENSP00000216479  |
| 51  | CASP3            | 9606.ENSP00000311032  |
| 52  | MAPK8            | 9606.ENSP00000378974  |
| 53  | MMP1             | 9606.ENSP00000322788  |
| 54  | STAT1            | 9606.ENSP00000354394  |
| 55  | CDK1             | 9606.ENSP00000378699  |
| 56  | HMOX1            | 9606.ENSP00000216117  |
| 57  | CYP3A4           | 9606.ENSP00000337915  |
| 58  | CYP1A1           | 9606.ENSP00000369050  |
| 59  | ICAM1            | 9606.ENSP00000264832  |
The hub targets were imported into the David database to clarify their function and role in signal transduction. GO biological process enrichment analysis and KEGG signal pathway analysis are carried out. The enrichment analysis results are enhanced with the R program package and displayed in the form of a bubble diagram.

2.5. Construction of the Bioactive Ingredients-Hub Target Network. Cytoscape 3.9.1 software was used to build the bioactive ingredients-hub target network. In this network, nodes represent bioactive ingredients and hub targets.

2.5.1. Hub Target-GO BP/Pathway/Disease Network. Use Cytoscape 3.9.1 to build the network model. Nodes represent hub targets, pathways, and diseases, and edges represent interactions between these nodes.

3. Results

3.1. Screening of Bioactive Ingredients of the Pan Ji Sheng Formula. The bioactive ingredients of eight Chinese herbal
medicines from the Pan Ji Sheng formula were screened from the TCMSP platform in this study. Because there is no relevant information on the TCMSP platform for Polygonum chinense and Chimonanthus salicifolia, we obtained the active components of these two herbs through literature retrieval and then tested whether they meet the standards of oral bioavailability (OB) ≥ 30 percent and drug class (DL) ≥ 0.18 in TCMSP. We obtained the active components of the other six herbs from TCMSP. In total, this study screened 96 active ingredients from eight herbs in the Pan Ji Sheng formula (Table 1).
named as key bioactive ingredients. For more information, see Table S2. The network of herbal-key bioactive ingredient-hub targets was constructed using Cytoscape 3.9.1 (Figure 5). In addition to Perilla frutescens, the other seven Chinese herbal medicines have three or more corresponding key bioactive ingredients. Some hub genes are affected by multiple bioactive ingredients at the same time. The primary targets of the active ingredients are MAPK14, HSP90AA1, PTGS2, and ESR1. These genes may be the primary targets of the formula.

3.5. GO Functional Annotation and KEGG Pathway Analysis.
To investigate the biological processes engaged in hub targets, GO enrichment analysis and KEGG enrichment analysis on 25 hub genes were analyzed in the David website. The mechanism of action of the formula can be researched, based on the biological process regulated by the hub target.

Beautify the enrichment analysis results with $R$ (Figure 6). In total, 226 GO biological process enrichment results were obtained. Negative regulation of the apoptotic process, positive regulation of the nitric oxide biosynthetic process, and positive regulation of transcription from the RNA polymerase II promoter are the top three enrichment biological processes. As shown in Figure 6(a), the top 20 GO biological processes are represented in the form of a bubble diagram, where the size of the circle represents the enrichment of relevant targets in the pathway, and the darker the color of the circle represents the degree of enrichment of targets, indicating that the formula could have physiological effects by regulating these biological processes.

For KEGG pathway enrichment analysis, 25 hub targets were mapped into the David database. The species was defined as “human,” and a total of 94 pathways were obtained. As shown in Figure 6(d), the top 20 pathways with high significance of KEGG enrichment results are closely related to the mechanism of the Pan Ji Sheng formula. The top five pathways include hepatitis B, pathways in cancer, TNF signaling pathway, toxoplasmosis, and toll-like receptor signaling pathway. The majority of these pathways are linked to the genes TP53, JUN, AKT1, MAPK14, HSP90AA1, and PTGS2.

Figure 2: Related diseases and expression patterns of all target genes. (a) The summary of enrichment analysis in Disgenet. (b) The summary of enrichment analysis in PaGenBase.
Figure 3: PPI network of all target genes. (a) PPI network, colored and in the middle are 25 hub genes. (b) Top 25 genes in the network ranked by the MCC method in "Cytohubba".

Figure 4: Clusters 1–6 in the PPI network. Among them, 25 hub genes are painted red and orange.
**Figure 5:** Herbal-key bioactive ingredient-hub target network.

**Figure 6:** GO and KEGG enrichment analysis of hub genes.
We also performed disease enrichment analysis to investigate diseases associated with hub targets. Figure 7 shows the classification of diseases enriched in hub targets. The three major categories are cancer, infection, and immune system. Our findings indicate that the formula studied in this study may primarily target these diseases.

3.5.1. Hub Target-GO BP/Pathway/Disease Class Network. In order to demonstrate the biological process of the hub target and the relationship between the hub target and the pathway more clearly, the hub target-GO BP/pathway/disease class network was built with Cytoscape 3.9.1 software (Figure 8).

The hub target is represented by the circle in the center of Figure 8. The left and right sides of Figure 8 show the top 20 enriched biological processes and pathways, respectively. We can clearly understand the relationship between the targets and biological processes or pathways. MAPK14, hSP90AA1, and PTGS2 genes are associated with apoptotic biological processes, TNF signaling pathways, toll-like receptor signaling pathways, and cancer pathways. The formula could play a significant role by regulating these pathways.

In order to demonstrate the link between the hub targets and diseases more clearly, Cytoscape 3.9.1 software was used to create a network of hub targets and diseases (Figure 9). The genes MAPK14, HSP90AA1, PTGS2, and ESR1 have been linked to cancer, infection, and immune disease.
4. Discussion

Traditional Chinese medicine formulas are typically difficult to decipher due to the action mode of traditional Chinese medicine formulas [33]. Using network pharmacology, this study explains the action mechanism of the Pan Ji Sheng Chinese medicine formula. According to the findings of this study, CASP3, AKT1, JUN, and other genes are the hub targets of the formula to enhance immune response and anti-inflammatory.

According to the active ingredient-target network, HSP90AA1, PTGS2, ESR1, and MAPK14 are the four key genes regulated by the active ingredient of the Pan Ji Sheng formula. HSP90AA1 is an inflammation-related protein that can be significantly upregulated with some inflammation-related genes in the inflammatory response [34, 35]; PTGS2 is involved in inflammation, immunity, and other processes [36, 37]; ESR1 is also involved in inflammation and immunity and is one of the key targets for the treatment of pneumonia [38, 39]; and MAPK14 is related to autophagy and plays an important role in immune response [40].

As shown in the results, 19 of the 25 hub targets were discovered to be involved in the pathways in cancer, with the pathways in cancer being the most significant pathway. This could be due to the fact that respiratory inflammation and lung disease are risk factors for cancer [41, 42]. Other top KEGG enrichment pathways include hepatitis B, the TNF signaling pathway, toxoplasmosis, and the toll-like receptor signaling pathway. A key target gene is tumor necrosis factor (TNF), a cytokine secreted by macrophages and adipocytes. It can cause IR by suppressing the activity of the PI3K/Akt signaling pathway. TNF has been shown to activate MAPK and NF-B signaling pathways, which regulate inflammatory response, oxidative stress, and apoptosis [43, 44].

The network pharmacological analysis reveals that the Pan Ji Sheng formula could regulate HSP90AA1, PTGS2, ESR1, MAPK14, and other genes, modulating pathways such as cancer pathways, TNF signaling pathways, and toll-like receptor signaling pathways to regulate inflammatory response and immune processes.

This study investigated the anti-inflammatory and immune mechanisms of Pan Ji Sheng formula. However, in vivo and in vitro experiments are needed to provide more information on the mechanism of action of the formula.

5. Conclusions

The active components of the Pan Ji Sheng formula could regulate certain proteins, including HSP90AA1, PTGS2, ESR1, and MAPK14. The Chinese herbs in the Pan Ji Sheng formula have a synergistic therapeutic effect, primarily by acting on inflammation and immune-related signal pathways. Pan Ji Sheng formula plays the functions through multicomponents, multitargets (HSP90AA1, PTGS2, ESR1, MAPK14, and other hub targets), and multipathways (inflammation and immune-related signal pathways). These findings could serve as guidelines for future research into this formula. Based on the present study, functional
experiments can be performed on animal models or human cells to validate the pharmacological mechanisms of the Pan Ji Sheng formula in the future. This research has theoretical significance for the TCM pharmacology and has application value for the development and utilization of TCMs.

Data Availability

The data used to support the findings of this study are included within the supplementary information files.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Authors’ Contributions

Qin Chen and Shiji Wu designed the experiments; Shiji Wu, Hongliang Jiang, and Zongwen Chen collected and assembled the data; data analysis was done by Shiji Wu, Hongliang Jiang, and Weinig Lu; the manuscript was written by all the authors; final approval of the manuscript was done by all the authors.

Supplementary Materials

Table S1: targets of Pan Ji Sheng Formula. Table S2: detailed information of herbal-key bioactive ingredients-top 25 hub targets. (Supplementary Materials)

References

[1] Z. Zeng, L. Li, and Y. Pang, “Analysis on climate adaptability of traditional villages in Lingnan, China-World cultural heritage site of majianglong villages as example,” Procedia Engineering, vol. 205, pp. 2011–2018, 2017.

[2] H. Rong-Rong, Y. Xin-Sheng, and K. Hiroshi, “The “Xiehuo” effect of Guangdong herbal tea and its composition,” World Science and Technology/Modernization of Traditional Chinese Medicine and Materia Medica, vol. 11, pp. 834–839, 2009.

[3] M. H. Pan, S. R. Zhu, W. J. Duan et al., “Shanghuo increases disease susceptibility: modern significance of an old TCM theory,” Journal of Ethnopharmacology, vol. 250, Article ID 112491, 2020.

[4] L. Chen, H. Deng, H. Cui et al., “Inflammatory responses and inflammation-associated diseases in organs,” Oncotarget, vol. 9, no. 6, pp. 7204–7218, 2018.

[5] L. Dall, S. Peterson, T. Simmons, and A. Dall, “Rapid resolution of cellulitis in patients managed with combination antibiotic and anti-inflammatory therapy,” Cutsis, vol. 75, no. 3, pp. 177–180, 2005.

[6] J. F. Chmiel, M. W. Konstan, and J. S. Elborn, “Antibiotic and anti-inflammatory therapies for cystic fibrosis,” Cold Spring Harbor Perspectives in Medicine, vol. 3, no. 10, Article ID a09779, 2013.

[7] G. A. Green, “Understanding NSAIDs: from aspirin to COX-2,” Clinical Cornerstone, vol. 3, no. 5, pp. 50–59, 2001.

[8] R. E. Harris, J. Beebe-Donk, H. Doss, and D. Burr Doss, “Aspirin, ibuprofen, and other non-steroidal anti-inflammatory drugs in cancer prevention: a critical review of non-selective COX-2 blockade (review),” Oncology Reports, vol. 13, no. 4, pp. 559–583, 2005.

[9] L. Laine, “Gastrointestinal effects of NSAIDs and coxibs,” Journal of Pain and Symptom Management, vol. 25, no. 2, pp. 32–40, 2003.

[10] F. Marsico, S. Paolillo, and P. P. Filardi, “NSAIDs and cardiovascular risk,” Journal of Cardiovascular Medicine, vol. 18, pp. e40–e43, 2017.

[11] A. Nande and A. L. Hill, “The risk of drug resistance during long-acting antimicrobial therapy,” medRxiv, 2021.

[12] H. Rongrong, Y. Xinxeng, and K. Hiroshi, “Studies on the “Xiehuo” effect and compositions of Guangdong herbal tea,” World Science and Technology, vol. 11, no. 6, pp. 834–839, 2009.

[13] S. Li, S. K. Li, D. P. Xu, A. N. Li, and H. B. Li, “Herbal Teas,” in Handbook of Functional Beverages and Human Health (1st ed.), F. Shahidi and C. Alasalvar, Eds., CRC Press, Boca Raton, FL, USA, 2016.

[14] R. R. He, B. Tsioi, Y. F. Li, X. S. Yao, and H. Kurihara, “The anti-stress effects of Guangdong herbal tea on immunocompromise in mice loaded with restraint stress,” Journal of Health Science, vol. 57, no. 3, pp. 255–263, 2011.

[15] Y. H. Luo, Y. Q. Huang, and H. Yang, “Research progress of Chinese herbal medical liangcha,” Stratight Pharmaceutical Journal, vol. 5, 2006.

[16] J. Xu and Y. Zhang, “Traditional Chinese medicine treatment of COVID-19,” Complementary Therapies in Clinical Practice, vol. 39, Article ID 101165, 2020.

[17] Y. Xiang, Z. Guo, P. Zhu, J. Chen, and Y. Huang, “Traditional Chinese medicine as a cancer treatment: modern perspectives of ancient but advanced science,” Cancer Medicine, vol. 8, no. 5, pp. 1958–1975, 2019.

[18] J. Yu, J. Guo, W. Tao et al., “Gancao-Gansui combination impacts gut microbiota diversity and related metabolic functions,” Journal of Ethnopharmacology, vol. 214, pp. 71–82, 2018.

[19] M. Yang, J. Luo, Q. Yang, and L. Xu, “Research on the medication rules of Chinese herbal formulas on treatment of threatened abortion,” Complementary Therapies in Clinical Practice, vol. 43, Article ID 101371, 2021.

[20] H. Yuan, S. Jiang, Y. Liu et al., “The flower head of Chrysanthemum morifolium Ramat. (Juhua): a paradigm of flowers serving as Chinese dietary herbal medicine,” Journal of Ethnopharmacology, vol. 261, Article ID 113043, 2020.

[21] L. Cheng, F. Wang, S. B. Zhang, and Q. Y. You, “Network pharmacology integrated molecular docking reveals the anti-COVID-19 and SARS mechanism of Fufang Banlangen Keli,” Natural Product Communications, vol. 16, 2021.

[22] A. Prasad, M. Muthamilarasu, and M. Prasad, “Synergistic antiviral effects against SARS-CoV-2 by plant-based molecules,” Plant Cell Reports, vol. 39, no. 9, pp. 1109–1114, 2020.

[23] Y. Mizukami, “Character of frontier orbitals of antiviral drugs: candidate drugs against COVID-19,” Open Journal of Physical Chemistry, vol. 10, no. 03, pp. 158–165, 2020.

[24] C. T. Lee, K. S. Huang, J. F. Shaw et al., “Trends in the immunomodulatory effects of cordyceps militaris: total extracts, polysaccharides and cordycepin,” Frontiers in Pharmacology, vol. 11, Article ID 575704, 2020.

[25] K. C. Hou, “Distorted key theory and its implication for drug development,” Current Proteomics, vol. 17, no. 4, pp. 311–323, 2020.

[26] Z. Zhou, B. Chen, S. Chen et al., “Applications of network pharmacology in traditional Chinese medicine research,” Evidence-Based Complementary and Alternative Medicine, vol. 2020, Article ID 1646905, 7 pages, 2020.
[27] J. Muhammad, A. Khan, A. Ali et al., “Network pharmacology: exploring the resources and methodologies,” Current Topics in Medicinal Chemistry, vol. 18, no. 12, pp. 949–964, 2018.
[28] J. Ru, P. Li, J. Wang et al., “TCMSP: a database of systems pharmacology for drug discovery from herbal medicines,” Journal of Cheminformatics, vol. 6, pp. 13–16, 2014.
[29] D. Gfeller, A. Grosdidier, M. Wirth, A. Daina, O. Michielin, and V. Zoete, “SwissTargetPrediction: a web server for target prediction of bioactive small molecules,” Nucleic Acids Research, vol. 42, no. W1, pp. W32–W38, 2014.
[30] D. Szklarczyk, A. L. Gable, K. C. Nastou et al., “The STRING database in 2021: customizable protein-protein networks, and functional characterization of user-uploaded gene/measurement sets,” Nucleic Acids Research, vol. 49, no. D1, pp. D605–D612, 2021.
[31] D. Otasek, J. H. Morris, J. Bouças, A. R. Pico, and B. Demchak, “Cytoscape automation: empowering workflow-based network analysis,” Genome Biology, vol. 20, pp. 185–215, 2019.
[32] Y. Zhou, B. Zhou, L. Pache et al., “Metascape provides a biologist-oriented resource for the analysis of systems-level datasets,” Nature Communications, vol. 10, pp. 1523–1610, 2019.
[33] R. Guo, X. Luo, J. Liu, L. Liu, X. Wang, and H. Lu, “Omics strategies decipher therapeutic discoveries of traditional Chinese medicine against different diseases at multiple layers molecular-level,” Pharmacological Research, vol. 152, Article ID 104627, 2020.
[34] A. D. Zuehlke, K. Beebe, L. Neckers, and T. Prince, “Regulation and function of the human HSP90AA1 gene,” Gene, vol. 570, no. 1, pp. 8–16, 2015.
[35] X. Xiao, W. Wang, Y. Li et al., “HSP90AA1-mediated autophagy promotes drug resistance in osteosarcoma,” Journal of Experimental & Clinical Cancer Research, vol. 37, pp. 201–213, 2018.
[36] T. Kosaka, A. Miyata, H. Ibara et al., “Characterization of the human gene (PTGS2) encoding prostaglandin-endoperoxide synthase 2,” European Journal of Biochemistry, vol. 221, no. 3, pp. 889–897, 1994.
[37] J. Li, X. Kong, J. Zhang, Q. Luo, X. Li, and L. Fang, “MiRNA-26b inhibits proliferation by targeting PTGS2 in breast cancer,” Cancer Cell International, vol. 13, pp. 7–6, 2013.
[38] D. R. Robinson, Y. M. Wu, P. Vats et al., “Activating ESR1 mutations in hormone-resistant metastatic breast cancer,” Nature Genetics, vol. 45, no. 12, pp. 1446–1451, 2013.
[39] F. Holst, P. R. Stahl, C. Ruiz et al., “Estrogen receptor alpha (ESR1) gene amplification is frequent in breast cancer,” Nature Genetics, vol. 39, no. 5, pp. 655–660, 2007.
[40] Y. Hirota, S. I. Yamashita, Y. Kurihara et al., “Mitophagy is primarily due to alternative autophagy and requires the MAPK1 and MAPK14 signaling pathways,” Autophagy, vol. 11, no. 2, pp. 332–343, 2015.
[41] G. Lee, T. C. Walser, and S. M. Dubinett, “Chronic inflammation, chronic obstructive pulmonary disease, and lung cancer,” Current Opinion in Pulmonary Medicine, vol. 15, no. 4, pp. 303–307, 2009.
[42] A. H. Wu, E. T. H. Fontham, P. Reynolds et al., “Previous lung disease and risk of lung cancer among lifetime nonsmoking women in the United States,” American Journal of Epidemiology, vol. 141, no. 11, pp. 1023–1032, 1995.
[43] G. Chen and D. V. Goeddel, “TNF-R1 signaling: a beautiful pathway,” Science, vol. 296, no. 5573, pp. 1634–1635, 2002.
[44] J. R. Bradley, “TNF-mediated inflammatory disease,” The Journal of Pathology, vol. 214, no. 2, pp. 149–160, 2008.