Transcriptional profiling and network pharmacology analysis identify the potential biomarkers from Chinese herbal formula Huosu Yangwei Formula treated gastric cancer in vivo

FANG Sheng-Quan1Δ, LIU Yue-Han2Δ, ZHAO Kun-Peng3, WANG Hong-Wei1, DENG Yu-Hai1, ZHOU Yu-Xuan1, GE Guang-Bo2, NI Hong-Mei3*, CHEN Qi-Long1,2*

1 Department of Gastroenterology, Yueyang Hospital of Integrated Traditional Chinese and Western Medicine, Shanghai University of Traditional Chinese Medicine, Shanghai 200437, China;
2 Institute of Interdisciplinary Integrative Medicine Research, Shanghai University of Traditional Chinese Medicine, Shanghai 201203, China;
3 School of Basic Medical Science, Shanghai University of Traditional Chinese Medicine, Shanghai 201203, China

Available online 20 Dec., 2021

[ABSTRACT] Huosu Yangwei (HSYW) Formula is a traditional Chinese herbal medicine that has been extensively used to treat chronic atrophic gastritis, precancerous lesions of gastric cancer and advanced gastric cancer. However, the effective compounds of HSYW and its related anti-tumor mechanisms are not completely understood. In the current study, 160 ingredients of HSYW were identified and 64 effective compounds were screened by the ADMET evaluation. Furthermore, 64 effective compounds and 2579 potential targets were mapped based on public databases. Animal experiments demonstrated that HSYW significantly inhibited tumor growth in vivo. Transcriptional profiles revealed that 81 mRNAs were differentially expressed in HSYW-treated N87-bearing Balb/c mice. Network pharmacology and PPI network showed that 12 core genes acted as potential markers to evaluate the curative effects of HSYW. Bioinformatics and qRT-PCR results suggested that HSYW might regulate the mRNA expression of DNAJB4, CALD, AKR1C1, CST1, CASP1, PREX1, SOCS3 and PRDM1 against tumor growth in N87-bearing Balb/c mice.

[KEY WORDS] Huosu Yangwei (HSYW) Formula; Network pharmacology; Protein-protein interaction (PPI) network; Gastric Cancer

[CLC Number] R965

Introduction

Gastric cancer is the fifth most frequently diagnosed malignancy and the third leading cause of cancer-related deaths worldwide [1]. As a heterogeneous disease, gastric cancer is characterized by pathological features, biological functions and genomic profiles [2]. In the past decades, despite the advances in early diagnosis and treatment, gastric cancer tends to be prevalent in younger adults [3] and remains a major cause of the global burden of cancer [4].

Chinese herbal medicine has various advantages for tumor treatment and prevention, such as multiple ingredients, multiple targets and lower adverse reactions [5, 6]. Huosu Yangwei (HSYW) formula is a traditional Chinese herbal medicine, including Huoxiang, Zisugeng, Baizhu, Zhike, Doukou, Foshou, Wumei, Shengjiang, Dazao, Gancao, Huangqi, Dihuang, Mudanpi, Tianhuafen, Dangsheng, and Pugongying. Previous studies demonstrated that HSYW has been extensively applied for the treatment of chronic atrophic gastritis and precancerous lesions of gastric cancer [7-9]. In advanced gastric cancer treatment, HSYW significantly relieved clinical symptoms, improved the quality of life, and reduced the incidence of adverse reactions after chemotherapy [10]. Furthermore, HSYW also remarkably inhibited the growth of gastric...
cancer cells in vitro. These studies suggested that HSYW may exert anti-tumor effects for gastric cancer treatment. However, in light of the multi-ingredients, multi-targets and multi-pathways of the formula, it is difficult to explore the complex mechanisms of HSYW on gastric cancer through traditional pharmacological methods. Network pharmacology is a systematic approach to disclose the mechanisms of drugs, which is a unique advantage for Chinese herbal medicine research. Therefore, more and more researchers start to utilize network pharmacology to explore the material basis of TCM, which can not only reveal the complex interactions between herbal ingredients and proteins, but also provide a proper method to evaluate the pharmacological effects of Chinese herbal medicine from multiple dimensional perspective.

In the current study, we identified 160 ingredients of HSYW using the HPLC/MS method, and screened out 64 active compounds based on ADMET evaluation. To understand the anti-tumor mechanisms of HSYW, we investigated the transcriptional profiles of HSYW treated gastric cancer in vivo, and identified several potential markers in the process of curative effects evaluation of HSYW in gastric cancer treatment.

Materials and Methods

Identification of HSYW ingredients

HSYW is a hospital preparation of Yueyang Hospital of Integrated Traditional Chinese and Western Medicine, Shanghai University of Traditional Chinese Medicine, and produced by Shanghai Baolong Pharmaceutical Co., Ltd. (trade name: Huosu Yangwei Oral Solution, batch number: Z05050328).

HSYW solution was activated by 5 mL methanol, and balanced by 60% methanol solution in advance. Then, 1 mL sample solution was passed through SPE column (Waters ACQUITY UPLC® HSS T3, 2.1 mm × 100 mm, 1.8 μm), and eluted by 5 mL 60% methanol solution. Then, 10 mL methanol was added to elute again. The above eluates were combined and concentrated at 40 °C until dryness. Furthermore, the above dry eluates were dissolved again with 1 mL methanol for centrifugation at 12 000 r·min⁻¹ for 5 min, and the resultant supernatant was collected for standard HPLC (Uitmine, Thermo, US) and fingerprint analysis. The Sciex Triple TOF 4600 LC/MS spectrometer (SCIEX, Singapore) was used to obtain the mass spectra of different compounds.

Effective compound evaluation and target prediction

First, the ADMET evaluation was used to screen the effective compounds from the identified ingredients of HSYW. The absorption, distribution, excretion and toxicity of each compound was calculated using SwissADME and pkCSM database. Second, the TCMS, TCMIP, TCMID, ZINC and SwissTargetPrediction were used to predict the potential targets of effective compounds of HSYW.

Network construction and enrichment analysis

Using the effective compounds and predicted targets, the compound-target network (CTN) of HSYW was constructed by Cytoscape software (version 3.8). The nodes represent compounds or targets, and edges represent the connection among of nodes. Furthermore, the DAVID online were used to analyzed the biological functions of potential targets of HSYW, and the significantly terms were defined as P < 0.05, for gene sets containing more than five overlapping genes.

Animals

Male Balb/c mice (four weeks old) were purchased from Shanghai Model Organisms Center (SCXK Shanghai 2014-0002), and fed in the Animal Laboratory of Shanghai University TCM (SYXK Shanghai 2014-0008). The mice were housed in polycarbonate cages at a standard air-conditioned room (23 ± 2 °C, 55% ± 10% R.H.). All animal procedures were approved by the Ethical Committee on Animal Experiments, Shanghai University of TCM. Experimental procedures were in accordance with the 3Rs principle and ARRIVE guidelines.

Model establishment of GC mice and drug treatment

Gastric cancer animal model

The human gastric cancer cell line N87 was purchased from the Institute of Biochemistry and Cell Biology (Shanghai, China). The cell line N87 was cultured in DMEM medium (Gibco, USA), supplemented with 10% fetal bovine serum (FBS) (Gibco, USA) and 100 U·mL⁻¹ penicillin-streptomycin, at 37 °C in a humidified 5% CO₂ incubator.

The mice were weighed and randomly divided into three groups (n = 10): a control group, a model group and a HSYW group. The cells at the logarithmic phase (5 × 10⁶/mL) were subcutaneously inoculated in the right axillary of the mice (100 μL per mouse).

Drug treatment

The mice were administered with the same dose of HSYW as the clinical equivalent of human (body weight, 60 kg), and the conversion coefficient was 10. Accordingly, the total dose of 20 g mouse was 60 mL/60 kg × 0.02 kg × 10.0 = 0.2 mL every day. The mice in the model group were treated with 0.1% sodium chloride solution (0.2 mL per mouse, once per day) for 25 days. One week after inoculation, the mice was weighed every three days. Tumor size was measured with a vernier caliper, and tumor volume = a × b² / 2 (a represents the longest diameter, and b represents the shortest diameter of tumor).

Pathological examination

Hematoxylin and eosin (HE) staining was performed as indicated in previous reports. The tumor tissue was immersed in 4% paraformaldehyde for 4 h, and transferred to 70% ethanol. Then, these tissues were dehydrated through a series alcohol gradient, embedded into paraffin blocks and obtained 5-μm-thick tissue sections. Furthermore, the sections were dewaxed with xylene and rehydrated by decreasing ethanol concentrations, washed in PBS buffer and then stained. After staining, the tissue sections were dehydrated by increasing ethanol concentrations and using xylene. Finally,
images were scanned by a pathological image analysis system (Olympus Corporation, Tokyo, Japan).

Detection of transcriptional profiles

Using RNA sequencing, the transcriptional profiles of HSYW treated gastric cancer mice (the HSYW Group, HSYW, n = 3) and gastric cancer mice (the Model Group, M, n = 3) were detected, respectively. Total RNA was extracted from the tumor tissues using the mirVana miRNA Isolation Kit (Ambion) following the manufacturer’s protocol. RNA integrity was evaluated using the Agilent 2100 Bioanalyzer (Agilent Technologies, Santa Clara, CA, USA). The samples with RNA Integrity Number (RIN) ≥ 7 were subjected to subsequent analysis. The libraries were constructed using TruSeq Stranded, and then sequenced on the Illumina Sequencing Platform (HiSeqTM 2500). Raw reads generated during high-throughput sequencing were fastq format sequences. The raw reads required to be further filtered, so as to get high-quality reads for later analysis.

Analysis of differential expressed mRNAs

The levels of mRNAs were calculated using FPKM (Fragments Per kb Per Million Reads) algorithm. Based on DESeq software, the mRNA levels of all samples were normalized, and the difference significance test of reads was performed by NB (Negative binomial distribution). Furthermore, R package was used to identify the differentially expressed (DE) mRNAs between the HSYW Group and Model Group (fold change > 1.5, P < 0.05). Heat-map and hierarchical analysis were performed using Cluster 3.0 and Java TreeView programs. The biological functions of DE mRNAs were analyzed using DAVID online, and the significance was defined as P < 0.05.

Protein-protein interaction (PPI) network construction

To investigate the biological functions of the mRNA profiles of HSYW treated gastric cancer, the DE mRNAs were mapped for compound-target network, and the isolated DE mRNAs were used to construct a PPI network by the IntAct, BioGRID and MINT databases. Furthermore, the consecutive parameters of the network were calculated, including betweenness centrality (BC), closeness centrality (CC), degree (De) and topological coefficient (TC). In this work, the core node of the network was defined as BC ≥ avg (BC), CC ≥ avg (CC), De ≥ avg (De), and TC ≥ avg (TC).

qRT-PCR validation

Quantitative reverse transcription-polymerase chain reaction (qRT-PCR) was carried out to determine the mRNA levels of potential markers in HSYW-treated gastric cancer mice. The mRNA was quantified by the SYBR Green PCR Master Mixture (Toyobo, Ltd., Japan). A melting curve was plotted to verify the specificity of each PCR product, and Ct was reckoned as the number of cycle requirement. All mRNAs were validated in triplicate and the mRNA levels were calculated using $2^{\Delta\Delta CT}$. The Student’s t-test was utilized to estimate the mRNA levels, and differences with P value < 0.05 were considered significant.

The primer sequences were designed in the laboratory and synthesized by Generay Biotech (Generay, PRC) based on the mRNA sequences obtained from the NCBI database (Table 1). The levels of mRNAs were normalized to ACTB.

Survival analysis of GC patients based on core genes

In this work, the GEPIA database (Gene Expression Profiling Interactive Analysis, http://gepia.cancer-pku.cn/) was used to evaluate the expression of key genes and the survival curves of these genes in predicting the prognosis of GC patients, using data concerning RNA sequencing expression of 9736 tumors and 8587 normal samples from the TCGA (the Cancer Genome Atlas) and the GTEx (Genotype-Tissue Expression) projects.

Statistical analyses

Statistical analysis was conducted by SPSS 23.0 software (IBM Analytics), and all values are presented as mean ±

<table>
<thead>
<tr>
<th>Gene name</th>
<th>Forward primer (5′- 3′)</th>
<th>Reverse primer (5′- 3′)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AKR1C1</td>
<td>ACAATAATGAGGAGGAGTT</td>
<td>AGGTAAGGGTCACATAATCC</td>
</tr>
<tr>
<td>ALDH4A1</td>
<td>GGACCTGAAAGCTATTGC</td>
<td>CGGAAGAAGTGTAGTTG</td>
</tr>
<tr>
<td>CALD1</td>
<td>CAGAGGAAATCAGGATGATA</td>
<td>TTGTGTTGTTGTTGTTG</td>
</tr>
<tr>
<td>CASP1</td>
<td>CACACCTCAGGCTCAGA</td>
<td>CAGCTTGTAGCTGCTGATT</td>
</tr>
<tr>
<td>CBR3</td>
<td>CAACGAGTTACTGCGGATA</td>
<td>TCTCCTCTTGACTGTTGCT</td>
</tr>
<tr>
<td>CST1</td>
<td>AGAGGAGAGATGATGAAATC</td>
<td>CTGGTTAATCAGCAGTGG</td>
</tr>
<tr>
<td>DNAJB4</td>
<td>GCTGATGGAGAGATGTCATA</td>
<td>ACAATGTCGCTGAATACT</td>
</tr>
<tr>
<td>FBXL16</td>
<td>CCTGGACATCGTGATGTC</td>
<td>GCTGTCAGCGATACCA</td>
</tr>
<tr>
<td>PIGR</td>
<td>TGTCAGGCTGTGTCATA</td>
<td>GGAGGCAGCATTTCTCCTT</td>
</tr>
<tr>
<td>PRDM1</td>
<td>GGATTCTGGTGTGGTGGTGA</td>
<td>AAATGGTCATTGAGTGGTGA</td>
</tr>
<tr>
<td>PREX1</td>
<td>CGGAGGAGAATGCTGAGG</td>
<td>CTGGATACGCGAGAATCT</td>
</tr>
<tr>
<td>SOCS3</td>
<td>CCACCTGGACTCTTATG</td>
<td>TTGGCTTCCTTGCTGCTGT</td>
</tr>
</tbody>
</table>
Results

Effective compound identification and potential target prediction of HSYW

In this work, 160 ingredients of HSYW were identified using the HPLC/MS method (Supplementary Table 1), while 64 effective compounds were screened out from the identified ingredients based on the ADMET analysis (Supplementary Table 2). Furthermore, a total of 2579 potential targets of 64 effective compounds were predicted from the databases, and the constructed compound-target network (CTN) was shown in Fig. 1A.

Enrichment analysis of HSYW targets

Enrichment analysis was conducted to analyze the potential targets of HSYW using DAVID online [27, 28]. In top 20 KEGG pathways, 12 terms were related to signaling pathways, such as ErbB signaling pathway, PI3K-Akt signaling pathway, HIF-1 signaling pathway, cAMP signaling pathway, chemokine signaling pathway, Rap1 signaling pathway, T cell receptor signaling pathway, Fc epsilon RI signaling pathway, TNF signaling pathway, FoxO signaling pathway, Ras signaling pathway, and VEGF signaling pathway. Seven terms were related to cancer-related pathways, including pathways in cancer, prostate cancer, chronic myeloid leukemia, acute myeloid leukemia, proteoglycans in cancer, pancreatic cancer, and central carbon metabolism in cancer. These findings suggest that the regulation of cancer and signaling-related pathways may be the principal functions in HSYW treated gastric cancer process (Fig. 1B).

Furthermore, the Gene Ontology (GO) enrichment analysis showed that the Biological Process (BP) terms were mainly associated with protein phosphorylation, MAPK cascade, response to drugs, T cell receptor signaling pathway, inflammatory response, signal transduction and cell proliferation (Fig. 1C). The Cellular Component (CC) terms were significantly related to plasma membrane, cytosol, extracellular space, extracellular region, mitochondrion, and membrane (Fig. 1D). The Molecular Function (MF) terms were mainly correlated with ATP binding, protein kinase activity, enzyme binding, receptor binding, drug binding, and protein binding (Fig. 1E).

HSYW inhibits tumor growth in N87-bearing Balb/c mice

To evaluate the effects of HSYW against tumor growth, N87-bearing Balb/c mice were used and treated with HSYW. After 25 days of HSYW treatment, the tumor size and weight results revealed that the growth of tumors was significantly decreased.
inhibited in HSYW treated N87-bearing Balb/c mice (Figs. 2A–2C). The HE staining results revealed that the nucleus and cytoplasm of the HSYW group were compact and dark stained, while the model group showed loose nucleus and pale staining (Fig. 2D). These findings indicated that the function of tumor cells in the model group was more vigorous than that in HSYW group. The abovementioned results suggested that HSYW may significantly inhibit tumor growth in vivo.

Transcriptional profile analysis

In this study, the mRNA profiles between HSYW treated (n = 3) and N87-bearing Balb/c mice (n = 3) were detected by RNA sequencing to identify the differentially expressed (DE) mRNAs. Using R package, 81 DE mRNAs were identified (fold change > 2, P < 0.05), including 33 up-regulated mRNAs and 48 down-regulated mRNAs in HSYW-treated mice (Fig. 2E). Enrichment analysis showed that the Gene Ontology (GO) terms (top 10) were mainly associated with regulation of neutrophil degranulation, regulation of neutrophil activation, integrin-mediated signaling pathway, superoxide metabolic process, retinoid metabolic process, collagen metabolic process, diterpenoid metabolic process, myeloid leukocyte migration, lymphocyte differentiation, and T cell differentiation (Fig. 2F). The KEGG pathways (top 10) were mainly associated with pertussis, hedgehog signaling pathway, Staphylococcus aureus infection, metabolism of xenobiotics by cytochrome P450, complement and coagulation cascades, rheumatoid arthritis, arginine and proline metabolism, amoebiosis, Legionellosis, and drug metabolism (Fig. 2G).

Compound-DEG network and PPI network construction

To screen the important targets of HSYW, the DE mRNAs were mapping to the compound-target network (CTN). As shown in Fig. 3A, the Compound-DEG network was isolated from CTN, including 47 compounds and 39 DE mRNAs. Furthermore, the 39 DE mRNAs related protein-protein interaction (PPI) network was constructed by the IntAct, BioGRID and MINT databases (Fig. 3B). In the PPI network, the consecutive parameters were calculated, respectively, and the core nodes were defined as BC ≥ avg (BC), CC ≥ avg (CC), De ≥ avg (De), and TC ≥ avg (TC). Finally, 12 core clusters were isolated from PPI network, including 6 up-regulated genes (DNAJB4, CALD1, AKR1C1, CST1, ALDH4A1, and FBXL16) and 6 down-regulated genes (PRDM1, CBR3, PIGR, SOCS3, CASP1, and PREX1) (Fig. 3C).

Furthermore, qRT-PCR was performed to validate the expression of 12 core mRNAs between the Model and HSYW groups. As showed in Fig. 3D, the transcriptional expression of these core genes were significantly regulated between the Model and HSYW groups (P < 0.05). These results...
ults suggest that the 12 mRNAs are the potential markers during the process of curative effects evaluation of HSYW, and play critical roles in gastric cancer treatment.

**Survival analysis for potential markers**

Using GEPIA, the expression of 12 core genes between gastric cancer \( (n = 408) \) and normal \( (n = 36) \) was calculated, respectively. The results revealed that the expression of DNAJB4, CALD1 and AKR1C1 significantly decreased, while the expression of CST1, CASP1 and PREX1 significantly increased in tumor tissues (Fig. 4A). Furthermore, we also explored the prognostic values of these 12 core genes in gastric cancer patients based on the overall survival (OS) calculation (Supplementary Fig. 1). The results showed that the high mRNA levels of SOCS3 and PRDM1 have statically significance \( (P = 0.0067 \) and \( 0.02) \) of OS in gastric cancer patients (Fig. 4B).

However, there was no difference in the levels of SOCS3 and PRDM1 between gastric cancer and normal tissue. To explore this phenomenon, we further analyzed the levels of SOCS3 and PRDM1 among different tumor grades in gastric cancer. The results showed that SOCS3 and PRDM1 had differential expression between tumor grade 1 and grade 3 (Fig. 5). In fact, differential expression revealed that these genes were associated with tumorigenesis, while the survival rate was correlated with tumor progression. This phenomenon suggested that SOCS3 and PRDM1 are highly correlated with cancer progression, rather than gastric carcinogenesis.

**Discussion**

As a traditional Chinese herbal medicine, Huosu Yang-wei (HSYW) formula exerted anti-tumor effects for advanced gastric cancer treatment \( [10] \). However, the molecular mechanisms of HSYW treated gastric cancer are still unclear. In this work, we first identified the main compounds of HSYW based on a HPLC/MS method. Then, the transcriptional profiles of HSYW treated N87-bearing Balb/c mice were detected by RNA sequencing. The KEGG pathways revealed that the hedgehog signaling pathway and metabolism of xenobiotics by cytochrome P450 might be more important for HSYW treated gastric cancer. The hedgehog signaling pathways involved in the biological processes are shown in Fig. 3.
pathway plays an important role during inflammation and carcinogenesis of gastric epithelial cells [11], and can mediate PD-L1 expression and promote tumor proliferation in gastric cancer [30]. Furthermore, the hedgehog signaling pathway is important in the maintenance of CD44 (+) cells, and acts to reverse chemotherapy resistance in these cells and may be beneficial in gastric cancer patients whose tumors express high levels of CD44 [30]. Cytochrome P450 enzyme family plays a critical role in the metabolism of various xenobiotics [33], especially in chemical carcinogenesis by activating or inactivating carcinogens, which impacts the initiation and promotion of tumors [33]. In gastric cancer, the expression of cytochrome P450 1A1 (CYP1A1) significantly increased in cancerous tissue [34], and cytochrome P450 2A6 (CYP2A6) polymorphisms were associated with the efficacy of S-1 in the adjuvant setting for gastric cancer [35]. Our previous study also demonstrated that HSYW strongly inhibited a range of human CYPs in a reversible manner [36]. These findings demonstrated that the hedgehog signaling pathway and metabolism of xenobiotics by cytochrome P450 might be high correlated with gastric cancer.

Furthermore, 12 core genes were selected based on compound-DEG network and PPI network. qRT-PCR validation showed that DNAJB4, CALD1, AKR1C1, CST1, ALDH4A1 and FBXL16 was up-regulated, while PRDM1, CBR3, PIGR, SOCS3, CASP1 and PREX1 was down-regulated in HSYW treated mice. According to GEPIA analysis, the expression of DNAJB4, CALD1 and AKR1C1 were down-regulated, while CST1, CASP1 and PREX1 were up-regulated in tumor tissues. The overall survival (OS) analysis showed that the high mRNA levels of SOCS3 and PRDM1 indicate poor prognosis in gastric cancer patients. Although no difference was found in SOCS3 and PRDM1 between gastric cancer and normal tissues, the significant difference among tumor grades revealed that SOCS3 and PRDM1 are high correlated with cancer progression, but not associated with gastric carcinogenesis. Interestingly, our studies showed that HSYW significantly down-regulated the expression of SOCS3 and
PRDM1 in gastric cancer mice (Fig. 3D). This phenomenon indicated that HSYW can down-regulate the expression of SOCS3 and PRDM1 and improve the survival rate of patients with gastric cancer.

Based on qRT-PCR and GEPIA results, DNAJB4, CALD1, AKR1C1, CST1, CASP1, PREX1, SOCS3 and PRDM1 were obtained from the 12 core genes, which were considered to be highly correlated with gastric cancer progression. In these eight genes, DNAJB4 belongs to the DNAJ (HSP40) family of heat shock proteins [39] and was considered as a tumor suppressor in various tumors [39]. In gastric cancer, DNAJB4 was likely to act as a sensor of E-cadherin structural features and contribute to tumor progression [40].

CALD1 is a calmodulin- and actin-binding protein that was revealed to display opposite roles in cancer and invasion [41], and associated with cell proliferation and migration in gastric cancer [42]. AKR1C1 is a member C1 of the human aldo-keto reductase 1 family, and up-regulated AKR1C1 was associated with a variety of cancers [43]. In gastric cancer, activation of the Nrf2/AKR1C axis possibly contributed to oxaliplatin resistance [43].

CASP1 is a member of the cysteine-aspartic acid protease (caspase) family, CASP1 was involved in diverse cellular processes regulation [48, 49], and higher CASP1 mRNA expression was associated with better overall survival (OS) in gastric cancer patients [50]. Phosphatidylinositol-3, 4, 5-trisphosphate dependent Rac exchange factor 1 (PREX1), was frequently up-regulated in many tumors [51], and high expression of PREX1 revealed poor prognosis in advanced gastric cancer patients [52]. SOCS3, suppressors of cytokine signaling 3, is a negative regulator of the JAK-STAT signaling pathway [53]. In gastric cancer, SOCS3 was identified to be the best predicator of lymph node metastasis [54], and high expression of SOCS3 inhibited cell proliferation, arrested cell cycle and facilitated apoptosis [55]. Notably, PRDM1 functioned as a tumor suppressor and played differential prognostic impacts among different cancers [56]. These findings suggested that DNAJB4, CALD1, AKR1C1, CST1, CASP1, PREX1, SOCS3 and PRDM1 are highly correlated with gastric cancer progression.

In conclusion, we identified 160 ingredients from HSYW and obtained 64 effective compounds based on ADMET analysis. Animal experiments demonstrated that HSYW significantly inhibited tumor growth in vivo. Transcriptional profiles and network pharmacology showed that 12 core genes acted as potential markers to evaluate the curative effects of HSYW. Bioinformatics analysis revealed that DNAJB4,
CALD1, AKR1C1, CST1, CASP1, PREX1, SOCS3 and PRDM1 were highly related with gastric cancer progression, and HSYW might regulate the expression of these genes against tumor growth in N87-bearing Balb/c mice.

References


Cite this article as: FANG Sheng-Quan, LIU Yue-Han, ZHAO Kun-Peng, ZHANG Hui-Xing, WANG Hong-Wei, DENG Yu-Hai, ZHOU Yu-Xuan, GE Guang-Bo, NI Hong-Mei, CHEN Qi-Long. Transcriptional profiling and network pharmacology analysis identify the potential biomarkers from Chinese herbal formula Huosu Yangwei Formula treated gastric cancer in vivo [J]. Chin J Nat Med, 2021, 19(12): 944-953.