A Network Pharmacology Approach to Explore the Potential Mechanisms of Huangqin-Baishao Herb Pair in Treatment of Cancer

ABCEF Tian Xu
E Qingguo Wang
AEG Min Liu

Corresponding Author: Min Liu, e-mail: liumin78@126.com
Source of support: Min Liu was supported by the Basic Scientific Research in Beijing University of Chinese Medicine (2018-JYB-ZDSYS002) and National Natural Science Foundation of China (No. 81774375)

Background: The aim of this study was to identify the bioactive ingredients of Huangqin-Baishao herb pair and to reveal its anti-cancer mechanisms through a pharmacology approach.

Material/Methods: Detailed information on compounds in the HQ-BS herb pair was obtained from the Traditional Chinese medicine systems pharmacology (TCMSP) and screened by the criteria of OB ≥30% and DL ≥0.18. A systematic drug targeting model (SysDT) was used for compound targets prediction, and then the targets were analyzed for Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment. The protein-protein interaction (PPI) network of HQ-BS targets was constructed, after identifying core networks through Cytoscape plugins.

Results: We found 47 bioactive compounds of HQ-BS and 107 human-derived targets. A compound target network and a target signal pathway network were constructed and used for topological analysis. Kaempferol, beta-sitosterol, stigmasterol, wogonin, and oroxylin-a were identified as core compounds and pathways in cancer. The calcium signaling pathway, PI3K-Akt signaling pathway, TNF signaling pathway, chemical carcinogenesis, estrogen signaling pathway, proteoglycans in cancer, HIF-1 signaling pathway, thyroid hormone signaling pathway, VEGF signaling pathway, small cell lung cancer, prostate cancer, colorectal cancer, NOD-like receptor signaling pathway, and T cell receptor signaling pathway were found to be potential signals of HQ-BS in treating cancer. Through PPI network analysis, TNF signaling pathway, tryptophan metabolism, proteoglycans in cancer, cell cycle, and chemical carcinogenesis sub-networks were obtained.

Conclusions: HQ-BS contains various bioactive compounds, including flavonoids, phytosterols, and other compounds, and these compounds can inhibit or activate multiple targets and pathways against cancer.

MeSH Keywords: Computer Communication Networks • Medicine, Chinese Traditional • Molecular Mechanisms of Pharmacological Action • Protein Interaction Maps • Ethnopharmacology

Full-text PDF: https://www.medscimonit.com/abstract/index/idArt/923199
Background

Cancer is an important public health problem in developing and as well as developed countries, and is now a leading cause of death worldwide. According to newly published cancer statistics data, the cancer death rate rose until 1991, and then fell continuously through 2017 in the USA [1]. Although there have been huge advances in cancer treatment in recent decades, discovering treatments with better efficacy and less toxicity is still an important area of cancer research.

As a major complementary and alternative medicine system, traditional Chinese medicine (TCM) has provided anti-cancer therapy for a long time because of its anti-inflammatory activities and use of abundant ingredients with tumor toxicity and microenvironment regulation. Huang Qin (HQ) refers to the dried root of Scutellaria baicalensis and Bai Shao (BS) is derived from Paeonia lactiflora Pall in the Chinese Pharmacopoeia (2015). As recorded in the Treatise on Febrile and Miscellaneous Diseases, the 2 herbs are used together at a crude weight ratio of 3:2 in Huangqin decoction to treat gastrointestinal ailments. Recent research focusing on the anti-cancer effect of Huangqin decoction found Huangqin decoction could ameliorate chemotherapy-induced gastrointestinal toxicity and enhance the therapeutic efficacy of antitumor drugs [2,3].

According to these experimental data, deletion of either HQ or BS eliminated Huangqin decoction’s synergistic activity, and deletion of either GC and DZ did not eliminate Huangqin decoction’s synergistic activity, indicating HQ and BS are essential in this formula and may have synergistic effects as an herb pair. Furthermore, based on a Chinese patent medicines study, HQ and BS are considered as 2 typical pathogen-eliminating and health-strengthening herbs, which are widely used against cancer [4]. Recent studies showed that HQ inhibited tumor growth and targeted apoptotic pathways, tumor-associated macrophages, MAPK pathway, and PI3K-Akt-mTOR signaling pathway [5]. BS is approved to induce apoptosis in HL-60 leukemic cells [6] and is recognized as a therapeutic agent against cancer cachexia [7].

To understand the anti-cancer mechanisms of HQ-BS herb pair, a network pharmacology approach was employed. Particularly, network pharmacology can help elucidate the interactive relationship between multiple components and multiple targets and investigate multiple molecular mechanisms of HQ-BS herb pair at a network level. Meanwhile, the relationships among compounds, targets, and signal pathways were also investigated. Finally, the multitarget and multipathway mechanisms of the cancer signal pathway were determined for HQ-BS against cancer.

Material and Methods

Active ingredients identification

The Traditional Chinese Medicine Systems Pharmacology Database (TCMSP, http://ibts.hkbu.edu.hk/LSP/tcmsp.php) [8] was used for bioactive ingredients identification of HQ-BS. Pharmacokinetic absorption, distribution, metabolism, and excretion (ADME) parameters of each compound in HQ-BS were determined. Bioactive ingredients were screened based on threshold values of OB ≥30% and DL ≥0.18, as recommended by the TCMSP database (Table 1).

Prediction of putative targets of HQ-BS

To identify the potential targets of bioactive ingredients in HQ-BS, a systematic drug targeting model (SysDT) based on RF and SVM methods was proposed, as previously described [9]. The gene name of each target was obtained from the UniProt Knowledgebase (http://www.uniprot.org/).

PPI network construction

Protein-protein interactions (PPI) data were obtained from STRING (https://string-db.org/cgi/input.pl). We filtered the STRING with threshold 0.4 and constructed a PPI network. Only interactions with weight above the threshold were selected for the newly constructed PPI network.

GO and KEGG pathway enrichment analysis

The gene ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analyses were conducted using the functional annotation tool of DAVID Bioinformatics Resources (http://david.abcc.ncifcrf.gov/). Official gene symbols were uploaded and annotations were limited by Homo sapiens and we set the background to be Homo sapiens. With p<0.05, we applied a hypergeometric test to obtain enriched GO terms and KEGG pathways.

Network construction

We constructed networks using Cytoscape (version 3.7.1) as follows: (1) the “HQ-BS bioactive compound target network” was built by connecting the HQ-BS ingredients and their targets, and (2) the “compound target signal pathway network” was built by connecting the ingredient targets and relevant signal pathways, and (3) the “PPI network” was constructed by linking targets to other human proteins interacting with them. CytoHubba, a Cytoscape plugin, was applied for calculating topological features of the PPI network, and another Cytoscape plugin, MCODE, was used to identify sub-networks.
Table 1. Detailed information on active ingredients in HQ-BS herb pair.

| Mol ID    | Molecule Name                          | Pubchem Cid  | MW     | OB (%) | DL | Herb      |
|-----------|----------------------------------------|--------------|--------|--------|----|-----------|
| MOL001689 | Acacetin                                | 5280442      | 284.28 | 34.97  | 0.24 | Huangqin  |
| MOL000173 | Wogonin                                 | 5281703      | 284.28 | 30.68  | 0.23 | Huangqin  |
| MOL000228 | (2R)-7-hydroxy-5-methoxy-2-phenylchroman-4-one | 821279      | 376.34 | 33.82  | 0.2  | Huangqin  |
| MOL002908 | 5,8,2'-Trihydroxy-7-methoxylavone       | 156992       | 302.28 | 38.04  | 0.23 | Huangqin  |
| MOL002909 | 5,7,2,5-tetrahydroxy-8,6-dimethoxylavone | 44258628     | 376.34 | 33.82  | 0.2  | Huangqin  |
| MOL009210 | Carthamidin                             | 188308       | 376.34 | 41.15  | 0.24 | Huangqin  |
| MOL002911 | 5,8,2'-Trihydroxy-6'-methoxylavone      | N/A          | 302.28 | 69.04  | 0.22 | Huangqin  |
| MOL002913 | Dihydrobaicalin_qt                      | 14135323     | 302.28 | 41.37  | 0.23 | Huangqin  |
| MOL002914 | Eriodyctiol (flavanone)                 | 373261       | 284.28 | 41.37  | 0.23 | Huangqin  |
| MOL002915 | Salvigenin                              | 161271       | 328.34 | 49.07  | 0.33 | Huangqin  |
| MOL002917 | 5,2',6'-Trihydroxy-7,8-dimethoxylavone  | 5322059      | 330.31 | 45.05  | 0.33 | Huangqin  |
| MOL002925 | 5,7,2',6'-Tetrahydroxylavone            | 5321865      | 286.25 | 37.01  | 0.24 | Huangqin  |
| MOL002926 | Dihydroxyroxylin A                      | 5316733      | 286.3  | 38.72  | 0.23 | Huangqin  |
| MOL002927 | Skullcapflavone II                     | 124211       | 374.37 | 69.51  | 0.44 | Huangqin  |
| MOL002928 | Oroxylin a                              | 5320315      | 284.28 | 41.37  | 0.23 | Huangqin  |
| MOL002931 | Panicolin                                | 5320399      | 314.31 | 76.26  | 0.29 | Huangqin  |
| MOL002932 | Panicolin                                | 5320399      | 314.31 | 76.26  | 0.29 | Huangqin  |
| MOL002933 | 5,7,4'-Trihydroxy-8-methoxylavone       | 5322078      | 300.28 | 36.56  | 0.27 | Huangqin  |
| MOL002934 | Neobaicalin                             | 124211       | 374.37 | 104.34 | 0.44 | Huangqin  |
| MOL002937 | Dihydroxyroxylin                        | 25721350     | 286.3  | 66.06  | 0.23 | Huangqin  |
| MOL000525 | Norwogonin                              | 5281674      | 302.28 | 39.4   | 0.21 | Huangqin  |
| MOL000552 | 2,6,2',4'-Tetrahydroxy-6'-methoxylavone | 159029       | 336.39 | 43.83  | 0.76 | Huangqin  |
| MOL000073 | Ent-Epicatechin                          | 182232       | 290.29 | 48.96  | 0.24 | Huangqin  |
| MOL000449 | Stigmasterol                            | 5280794      | 412.77 | 43.83  | 0.76 | Huangqin  |
| MOL001458 | Coptisine                               | 72322        | 320.34 | 30.67  | 0.86 | Huangqin  |
| MOL001490 | Bis[(2S)-2-ethylhexyl] benzene-1,2-dicarboxylate | 7057920 | 390.62 | 43.59  | 0.35 | Huangqin  |
| MOL001506 | Supraene                                | 638072       | 410.8  | 33.55  | 0.42 | Huangqin  |
| MOL002897 | Epiberberine                            | 160876       | 336.39 | 43.09  | 0.78 | Huangqin  |
| MOL008206 | Moslosoflavone                          | 188316       | 298.31 | 44.09  | 0.25 | Huangqin  |
| MOL010415 | 11,13-Eicosadienoic acid, methyl ester  | 5365674      | 322.59 | 39.28  | 0.23 | Huangqin  |
| MOL012245 | 5,7,4'-Trihydroxy-6-methoxylavone       | 26213330     | 302.3  | 36.63  | 0.27 | Huangqin  |
| MOL012246 | 5,7,4'-Trihydroxy-8-methoxylavone       | 42608119     | 302.3  | 74.24  | 0.26 | Huangqin  |
Results

Identification of bioactive compounds

From the TCMSP database, 143 compounds of Huangqin and 85 compounds of Baishao were obtained. A total of 47 compounds were identified by ADME-related pharmacokinetic parameters, OB and DL (Table 1), and the screening criteria were OB ≥30% and DL ≥0.18. In detail, 34 compounds were only in Huangqin, 11 compounds were only in Baishao, and 2 compounds were both in Huangqin and Baishao (Figure 1A).

Construction of compound target network

After converting the corresponding gene name into Gene Symbol through the UniProt database, deleting duplicate targets, 107 human-derived targets were obtained. Deleting non-target compound, we constructed a visualized compound target network containing 146 nodes and 871 edges (Figure 1B). Through topological analysis, some compounds were characterized as important molecules in compound target network of HQ-BS pair, including kaempferol (Degree: 55), beta-sitosterol (Degree: 50), stigmasterol (Degree: 44), wogonin (Degree: 36), (2R)-7-hydroxy-5-methoxy-2-phenylchroman-4-one (Degree: 36), oroxylin-a (Degree: 35), baicalein (Degree: 33), 5,2’-Dihydroxy-6,7,8-trimethoxyflavone (Degree: 32), Skullcap flavone II (Degree: 32), and rivularin (Degree: 31).

Construction of target signal pathway network

To evaluate the potential mechanisms of HQ-BS herb pair pharmacological effects, we used the DAVID web server to analyze target-related signal pathways. The threshold of P<0.05 was regarded as a significant signal pathway, which was then used to construct a target signal pathway network (Figure 2A). This network contained 68 nodes and 142 edges. Sorted by degree value, pathways in cancer were identified as the most important signal pathway (Degree: 21), indicating an anti-cancer effect of HQ-BS herb pair. Other related signal pathways included Calcium signaling pathway (Degree: 16), PI3K-Akt signaling pathway (Degree: 13), TNF signaling pathway (Degree: 10), Chemical carcinogenesis (Degree: 9), Estrogen signaling pathway (Degree: 9), Proteoglycans in cancer (Degree: 9), HIF-1 signaling pathway (Degree: 8), Thyroid hormone signaling pathway (Degree: 8), VEGF signaling pathway (Degree: 7), Small cell lung cancer (Degree: 7), Prostate cancer (Degree: 7), Colorectal cancer (Degree: 6), NOD-like receptor signaling pathway (Degree: 6), and T cell receptor signaling pathway (Degree: 6). Some targets were involved in multiple signals. PIK3CG was involved in 12 pathways and TP53 was involved in 7 pathways, suggesting they are potential targets against cancer.

Table 1 continued. Detailed information on active ingredients in HQ-BS herb pair.

| Mol ID   | Molecule Name                                  | Pubchem Cid | MW   | OB (%) | DL    | Herb          |
|---------|------------------------------------------------|-------------|------|--------|-------|---------------|
| MOL012266 | Rivularin                                      | 1389022     | 344.34 | 39.74  | 0.37  | Huangqin      |
| MOL001910 | 11alpha,12alpha-epoxy-3beta-23-dihydroxy-30-norolean-20-en-28,12beta-olide | N/A         | 470.71 | 64.77  | 0.38  | Baishao       |
| MOL001918 | Paeoniflorgenone                               | 70698143    | 318.35 | 87.59  | 0.37  | Baishao       |
| MOL001919 | (3S,5R,8R,9R,10S,14S)-3,17-dihydroxy-4,4,8,10,14-pentamethyl-2,3,5,6,7,9-hexahydro-1H-cyclopenta[a]phenanthrene-15,16-dione | 9841735     | 358.52 | 43.56  | 0.53  | Baishao       |
| MOL001921 | Lactiflorin                                    | 14605198    | 462.49 | 49.12  | 0.33  | Baishao       |
| MOL001924 | Paeoniflorin                                   | 442534      | 480.51 | 53.87  | 0.79  | Baishao       |
| MOL001925 | Paeoniflorin_qt                                | 11973336    | 318.35 | 68.18  | 0.33  | Baishao       |
| MOL001928 | Albiflorin_qt                                  | 134761887   | 318.35 | 66.64  | 0.33  | Baishao       |
| MOL001930 | Benzoylpaeoniflorin                            | 21631106    | 584.62 | 31.27  | 0.75  | Baishao       |
| MOL000211 | Mairin                                         | 64971       | 456.78 | 55.38  | 0.78  | Baishao       |
| MOL000422 | Kaempferol                                     | 5280863     | 286.25 | 41.88  | 0.24  | Baishao       |
| MOL000492 | (+)-catechin                                   | 9064        | 290.29 | 54.83  | 0.24  | Baishao       |
| MOL000358 | Beta-sitosterol                                 | 222284      | 414.79 | 36.91  | 0.75  | Huangqin and Baishao |
| MOL000359 | Sitosterol                                      | 12303645    | 414.79 | 36.91  | 0.75  | Huangqin and Baishao |

Tian X. et al.: Network pharmacology on Huangqin-Baishao against cancer © Med Sci Monit, 2020; 26: e923199

This work is licensed under Creative Common Attribution-NonCommercial-NoDerivatives 4.0 International (CC BY-NC-ND 4.0)
Figure 1. Bioactive compounds screening and compound target network construction. (A) The number of bioactive compounds in Huang Qin and Bai Shao are shown in a Venn diagram. (B) Compound target network of Huang Qin-Bai Shao herb pair constructed for topological analysis.
To understand the multiple target effect of HQ-BS herb pair, an ideogram was constructed using the KEGG mapper server (Figure 2B). Red labels represented HQ-BS herb pair-related targets or signal pathways, green labels represented other proteins in cancer pathway, and white labels represented other related signal pathways. HQ-BS herb pair influenced upstream, midstream, and downstream targets of the cancer signal pathway, and also influenced Calcium signaling pathway, PI3K-Akt signaling pathway, HIF-1 signaling pathway, and Estrogen signaling pathway, which may crosstalk with cancer signal.

Analysis of target protein interaction network (PPI)

We used the STRING online server to construct a PPI network of HQ-BS targets with the combined score $>0.4$. A network containing 102 nodes and 769 edges was obtained (Figure 3). As shown in Figure 3, each solid circle represented a target protein, and the center of the dot shows the protein structure. In the PPI network, the linkage of each node represented protein homology, gene co-expression, and gene co-evolution. Based on degree values, the top 10 targets in the network were identified: IL6 (degree: 46), TP53 (degree: 44), VEGFA (degree: 43), PTGS2 (degree: 38), JUN (degree: 37), TNF (degree: 37), NOS3 (degree: 37), ESR1 (degree: 36), CAT (degree: 36), and MAPK8 (degree: 35). The more positive the degree value of a target, the more prominent the role of this target in the network, and these may be the key targets for HQ-BS anti-cancer effects.

Identification of PPI sub-network

A Cytoscape plugin, MCODE, was used to identify sub-networks to understand the modulation of HQ-BS herb pair at a PPI network level. As shown in Figure 4, 5 sub-networks were identified. Sub-network 1 contained 19 nodes and 160 edges and
was involved in TNF signaling pathway. Sub-network 2 contained 9 nodes and 22 edges and was involved in tryptophan metabolism. Sub-network 3 contained 7 nodes and 14 edges and was involved in proteoglycans in cancer. Sub-network 4 contained 5 nodes and 8 edges and was involved in cell cycle. Sub-network 5 contained 4 nodes and 6 edges and was involved in chemical carcinogenesis.

**Discussion**

In ancient times, Chinese doctors always used a single herb to treat a specific disease, which was recorded by Shen Nong’s Materia Medica. Later, people started to realize that the therapeutic effect of a single herb may be not enough and could have various clinical adverse effects in. Therefore, multi-herb therapy characterized by the combination of 2 or more herbs with more efficacies and less adverse effects have been utilized for thousands of years in China. Herb pairs, which are the most fundamental, simplest, and the most basic composition units of multi-herb therapy, are unique combinations of 2 relatively fixed herbs [10]. Studying the biological mechanisms of herb pairs helps understand compatibility theory of TCM and improves its clinical usage. HQ and BS are 2 typical pathogen-eliminating and health-strengthening herbs which are widely used against cancer. Based on the HQ-BS herb pair, many formulae, including Huangqin decoction [11], are consequently formed, which are all used to treat cancer. Clinical and experimental evidence show that HQ-BS herb pair induces inflammation and oxidative stress of precancerous lesions of colorectal cancer [12,13]. In this study, we employed network pharmacology to reveal the potential biological mechanisms of HQ-BS herb pair against cancer.

Through ADME screening, 47 compounds were recognized as bioactive compounds of HQ-BS herb pair, indicating a multi-component effect. We constructed a compound target network and calculated the degree value of each node. Based on degree value, some ingredients were identified as important in this network, including kaempferol, beta-sitosterol, stigmasterol, wogonin, and oroxylin-a. Many in vivo and in vitro studies have found that these compounds possess potent anti-cancer pharmacological activity. For example, Kaempferol has a powerful anti-cancer effect against various cancer cell lines [14,15].

**Figure 2.** Multiple targets and multiple signal pathways of Huang Qin-Bai Shao (HQ-BS) herb pair against cancer. (A) Target KEGG signal pathway network. (B) Modulation by HQ-BS herb pair of pathways in cancer. Red labels represent HQ-BS herb pair-related targets or signal pathways, green labels represent other proteins in cancer pathway, and white labels represent other related signal pathways.
Figure 3. Protein-protein interaction (PPI) networks of Huang Qin-Bai Shao herb pair.
Beta-sitosterol suppresses tumor growth without toxicity in AGS xenograft mouse models and induces apoptosis in human gastric adenocarcinoma cells [16]. Stigmasterol was reported to reduce tumor growth, macrophage recruitment, and tumor angiogenesis in a cholangiocarcinoma xenograft model [17]. Wogonin significantly suppresses inflammation-associated carcinogenesis and tumor development [18]. Oroxylin-a has been reported to have multifunctional roles in anti-cancer effects, and a recent study found that long-term exposure inhibited cell migration via the CCL2 pathway in OSCC cells, without cytotoxic effects [19]. Baicalein was reported to promote MCF-7 breast adenocarcinoma cells apoptosis without toxic properties on normal breast epithelial cells [20]. Rivularin was found to gather in human liver cancer (HepG2) cells, indicating an anti-carcinoma effect [21]. Skullcap flavone II suppressed cell proliferation in a variety of cancer cell lines, such as HeLa, PC-3, and LNCaP [22, 23]. Salvigenin was reported to reduce tumor volume and modulate splenic T regulatory cells [24]. Most of these proven anti-cancer compounds were flavonoids, except for 2 phytosterol compounds – beta-sitosterol and stigmasterol. Kaempferol is derived from BS, beta-sitosterol is derived from both BS and HQ, and other compounds are derived from HQ.

KEGG signal pathway enrichment analysis showed that HQ-BS affects multiple targets and multiple related signal pathways in cancer. Various specific cancer signals were obtained based on P-value, including pathways in cancer, small cell lung cancer, prostate cancer, and colorectal cancer, indicating an anti-cancer effect of HQ-BS herb pair. To evaluate the anti-cancer effect of HQ-BS targets at a network level, we constructed a network according to PPI data and found 5 sub-networks related to TNF signaling pathway, tryptophan metabolism, proteoglycans in cancer, cell cycle, and chemical carcinogenesis, respectively. Tumor necrosis factor (TNF) mediates a variety of cell processes such as inflammation, differentiation, proliferation, and apoptosis. Clinical evidence shows that TNF polymorphisms are associated with susceptibility to cancer, including hepatocellular carcinoma [25], multiple myeloma [26], cervical cancer [27], and colorectal cancer [28]. Tryptophan (Trp) is an essential amino acid that is obtained exclusively from diet and used for the production of neurotransmitters and neuromodulators. Over 95% of free Trp is a substrate for the kynurenine pathway. Evidence indicates that Trp metabolism is involved in tumor progression by suppressing antitumor immune responses and increasing the malignant properties of cancer cells [29]. Some endogenous tryptophan metabolites, such as melatonin, kynurenines, and serotonin, and bacterial
tryptophan metabolites, including tryptamine, skatole, indole, and indolic acid were proved to play an important role in regulating the cancer immune system based on data obtained from SPF mice [30]. Our data showed HQ-BS can influence tryptophan metabolism by modulating a network containing 9 proteins. Future studies should assess whether HQ-BS influences tryptophan metabolism and its role in intestinal microbiota homeostasis. Proteoglycans are proteins that are attached by a specific linear carbohydrate chain of the glycosaminoglycan type and a part of the extracellular matrix and cell surfaces. Due to of their interactions with other ECM proteins, growth factors, and receptors, they can activate important cell signaling pathways (e.g., NF-κB, MAPK/β-catenin, Wnt, Hedgehog, TNF, IFN, Erk, FGF, and TGF-β) and their targets are associated with proliferation, angiogenesis, and cell motility [31]. Cell surface proteoglycans are involved in tumor-derived exosome biogenesis through the SDC1-syntenin-alix pathway [32], which mediates interactions of tumor cells and microenvironment, and stimulated tumor growth and development through specific signaling pathways related to metastasis, therapeutic resistance, and immunosuppression [33]. CDK2 is a core regulator of cell cycle through late G1-phase and S-phase. CDK2 is thought to be strongly linked to development of cancer, and accumulating evidence shows that inhibition of CDK2 induces cancer cell apoptosis without normal cell damage [34,35]. TOP2A is a marker of proliferation and chemotherapy resistance in cancer [36]. Several studies have reported that higher expression levels of TOP2A are related to poor cancer prognosis [36,37], and modulating TOP2A resulted in cancer cell apoptosis [38]. Our data show that HQ-BS herb pair can modulate cell cycle via a network containing CDK2 and TOP2A. HQ-BS herb pair also regulated chemical carcinogenesis pathway, indicating an anti-cancer effect.

Conclusions

HQ-BS herb pair contains various bioactive compounds, including flavonoids (e.g., kaempferol and wogonin) and phytosterols (e.g., beta-sitosterol and stigmasterol), and these compounds can interact with multiple targets and pathways. More importantly, HQ-BS herb pair can regulate the TNF signaling pathway, tryptophan metabolism, proteoglycans in cancer, cell cycle, and chemical carcinogenesis pathways to treat cancer. The present study provides evidence and promotes understanding of the multi-compounds and multitarget synergy of traditional Chinese medicine. However, in vivo and in vitro experiments and clinical investigations should be performed to verify the mechanism of HQ-BS herb pair against specific types of cancer in future studies.

Conflict of interest

None.

References:

1. Siegel RL, Miller KD, Jemal A: Cancer statistics, 2020. Cancer J Clin, 2020; 70(1): 7–30
2. Cui DN, Wang X, Chen JQ et al: Quantitative evaluation of the compatibility effects of huangqin decoction on the treatment of irinotecan-induced gastrointestinal toxicity using untargeted metabolomics. Front Pharmacol, 2017; 8: 211
3. Lam W, Jiang Z, Guan F et al: PHY906(KD018), an adjuvant based on a specific E3 ubiquitin ligases in cancer-bearing mice. J Ethnopharmacol, 2020; 246: 112222
4. Wu M, Lu P, Shi L, Li S: Traditional Chinese patent medicines for cancer treatment: Recent progress and perspectives in biomedical and clinical studies. Int J Chin Med, 2018; 46(1): 25–54
5. Kwon KB, Kim EK, Han MJ et al: Induction of apoptosis by Radix Paoniae Alba extract through cytochrome c release and the activations of caspase-9 and caspase-3 in HL-60 cells. Biol Pharm Bull, 2006; 29(6): 1082–86
6. Bae T, Jang J, Lee H et al: Paonia lactiflora root extracts suppresses cancer cachexia by down-regulating muscular NF-κB signalling and muscle-specific E3 ubiquitin ligases in cancer-bearing mice. J Ethnopharmacol, 2020; 246: 112222
7. Ru J, Li P, Wang J et al: TCMSP: A database of systems pharmacology for drug discovery from herbal medicines. J Cheminform, 2014; 6: 13
8. Zhang W, Xue K, Gao Y et al: Systems pharmacology dissection of action mechanisms of Diplacis Radix for osteoporosis. Life Sci, 2019; 235: 116820
9. Chen G, Yang Y, Hu C et al: Protective effects of Huangqin Decoction against ulcerative colitis and associated cancer in mice. Oncotarget, 2016; 7(38): 61643–55
10. Liu L: Clinical study on Huangqin Baishao decoction against ulcerative colitis. Asia-Pacific Traditional Medicine, 2017; 13(12): 118–19
11. Chen G, Yang Y, Hu C et al: Protective effects of Huangqin Decoction against ulcerative colitis and associated cancer in mice. Oncotarget, 2016; 7(38): 61643–55
12. Lu L: Clinical study on Huangqin Baishao decoction against ulcerative colitis. Asia-Pacific Traditional Medicine, 2017; 13(12): 118–19
13. Liu Y, Li LT, Ji XQ et al: Study on the effect and mechanism of huangqin-baishao herb pair on mice ulcerative colitis. China Pharmacy, 2018; 29(03): 356–60
14. Kim TW, Lee SY, Kim M et al: Kaempferol induces autophagic cell death via AMPK/PTEN/HSP90 axis in AGS human gastric adenocarcinoma cells and xenograft mouse models. Biochim Pharmacol, 2018; 152: 60–70
15. Budsan L, Gulei D, Jur A et al: Inhibitory effect of CAPE and kaempferol in colon cancer cell lines-possible implications in new therapeutic strategies. Int J Mol Sci, 2019; 20(5): 1199
16. Shin EJ, Choi HK, Sung MJ et al: Anti-tumour effects of beta-sitosterol are mediated by AMPK/PTEN/HSP90 axis in AGS human gastric adenocarcinoma cells and xenograft mouse models. Biochim Pharmacol, 2018; 152: 60–70
17. Kangsamsakin T, Chalhongyt S, Wootthichairangs C et al: Lupeol and stigmasterol suppress tumor angiogenesis and inhibit cholangiocarcinoma growth in mice via downregulation of tumor necrosis factor-α. PLoS One, 2017, 12(12): e0189628
18. Yao J, Zhao L, Zhao Q et al: NF-κB and Nrf2 signaling pathways contribute to wogonin-mediated inhibition of inflammation-associated colorectal carcinogenesis. Cell Death Dis, 2014; 5(6): e1283
19. Ku WT, Tung JJ, Lee TJ, Lai KC: Long-term exposure to oroxin A inhibits metastasis by suppressing CCL2 in oral squamous cell carcinoma cells. Cancers (Basel), 2019; 11(3): 353
20. Liu ZH, Yang CX, Zhang L et al: Baicalein, as a prooxidant, triggers mitochondrial apoptosis in MCF-7 human breast cancer cells through mobilization of intracellular copper and reactive oxygen species generation. Onco Targets Ther, 2019; 12: 10749–61
21. Jia D, Chen X, Cao Y et al: On-line comprehensive two-dimensional HepG2 cell membrane chromatographic analysis system for charactering anti-hepatoma components from rat serum after oral administration of Radix scutellariae: A strategy for rapid screening active compounds in vivo. J Pharm Biomed Anal, 2016; 118: 27–33
22. Tayarani-Najarani Z, Asili J, Parsaee H et al: Wogonin and neobaicalein from Scutellaria litwinowii roots are apoptotic for HeLa cells. Revista Brasileira de Farmacognosia, 2012; (22)2: 268–76
23. Bonham M, Posakony J, Coleman I et al: Characterization of chemical constituents in Scutellaria baicalensis with antiandrogenic and growth-inhibitory activities toward prostate carcinoma. Clin Cancer Res, 2005; 11(10): 3905–14
24. Noori S, Hassan ZM, Yaghmaei B, Dolatkhah M: Antitumor and immunomodulatory effects of salvigenin on tumor bearing mice. Cell Immunol, 2013; 286(1): 16–21
25. Yue X, Jiang X, Zou H et al: Association of hepatocellular carcinoma risk with polymorphisms in tumour necrosis factor alpha gene in a Chinese Han population. Int J Immunogenet, 2020 [Epub ahead of print]
26. Zmorzyński S, Popek-Marciniec S, Szudy-Szczyrek A et al: The association of GSTT1, GSTM1, and TNF-α polymorphisms with the risk and outcome in multiple myeloma. Front Oncol, 2019; 9: 1056
27. Wang Y, Yang J, Huang J, Tian Z: Tumor necrosis factor-α polymorphisms and cervical cancer: Evidence from a meta-analysis. Gynecol Obstet Invest, 2020; 85(2): 153–58
28. Huang X, Qin S, Liu Y et al: Associations of tumor necrosis factor-α polymorphisms with the risk of colorectal cancer: A meta-analysis. Biosci Rep, 2019; 39(1): BS20181750
29. Platten M, Nollen EAA, Röhrig UF et al: Tryptophan metabolism as a common therapeutic target in cancer, neurodegeneration and beyond. Nat Rev Drug Discov, 2019; 18(5): 379–401
30. Gao J, Xu K, Liu H et al: Impact of the gut microbiota on intestinal immunity mediated by tryptophan metabolism. Front Cell Infect Microbiol, 2018; 8: 13
31. Espinoza-Sánchez NA, Götte M: Role of cell surface proteoglycans in cancer immunotherapy. Semin Cancer Biol, 2020; 62: 48–67
32. Cerezo-Magála M, Bang-Rudemstam A, Belting M: The pleiotropic role of proteoglycans in extracellular vesicle mediated communication in the tumor microenvironment. Semin Cancer Biol, 2020; 62: 99–107
33. Tang Z, Li D, Hou S, Zhu X: The cancer exosomes: Clinical implications, applications and challenges. Int J Cancer, 2020; 146(11): 2946–59
34. Liang JW, Wang MY, Wang S, Li SL, Li WQ, Meng FH: Identification of novel CDK2 inhibitors by a multistage virtual screening method based on SVM, pharmacophore and docking model. J Enzyme Inhib Med Chem, 2020; 35(1): 235–44
35. Tadesse S, Anshabo AT, Portman N et al: Targeting CDK2 in cancer: Challenges and opportunities for therapy. Drug Discov Today, 2020; 25(2): 406–13
36. Jain M, Zhang L, He M et al: TOP2A is overexpressed and is a therapeutic target for adrenocortical carcinoma. Endocr Relat Cancer, 2013; 20(3): 361–70
37. Depowski PL, Rosenthal SI, Brien TP et al: Topoisomerase I alpha expression in breast cancer: Correlation with outcome variables. Mod Pathol, 2000; 13(5): 542–47
38. Chen T, Sun Y, Ji P et al: Topoisomerase IIC in chromosome instability and personalized cancer therapy. Oncogene, 2015; 34(31): 4019–31