Network Pharmacology-Based Approach to Investigate the Mechanisms of Piwei-Peiyan Decoction in the Treatment of Gastric Carcinoma

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Abstract

Purpose

Piwei-Peiyuan Decoction is a traditional Chinese medicine decoction, which has shown promising treatment in gastric carcinoma. However, the molecular mechanisms of gastric carcinoma (GC) have not been systematically revealed yet. In this work, a network pharmacology research was conducted to analyze mechanisms of Piwei-Peiyuan Decoction, aiming to provide a basis for the development and application of new drugs in treatment of GC.

Methods

In this study, a network pharmacology approach was used to predict targets, construct network maps and analyze relevant signaling pathways. We discovered active chemical ingredients and their targets in the Piwei-Peiyuan Decoction by the TCMSP and ADME database. Then we found main targets of GC by GeneCards (<https://www.genecards.org>), OMIM (<http://www.omim.org>) and DRUGBANK (<https://go.drugbank.com>) databases. According to the information of ingredients and GC, a PPI network was performed by the String (<https://string-db.org>) database. And a targets-pathways interaction network was constructed by Cytoscape (version 3.7.2), a bioinformatics software used for data visualization and integration.

Results

Our result indicated that the main active ingredients of Piwei-Peiyuan Decoction in treating GC are Isorhamnetin, Kaempferol, Quercetin and Luteolin, and main active targets are CASP-3, MAPK1, MYC, AKT1, P53. We found the mechanism of Piwei-Peiyuan Decoction for treating GC is mainly to regulate PI3K-Akt signaling pathway, MAPK signaling pathway, Ras signaling pathway, IL-17 signaling pathway, HIF-1 α signaling pathway to inhibit cell proliferation, promote apoptosis and then exert anti-cancer effects.

Conclusion

This study preliminarily analyzed the multi-component, multi-target and multi-path mechanism of Piwei-Peiyuan Decoction in the treatment of gastric carcinoma, providing a theoretical basis for its clinical application.

1. Background

As a type of malignancy that origin from epithelial cells of gastric mucosa. Almost 95% of GC is adenocarcinoma. GC is a malignancy with the highest morbidity and mortality in China after lung cancer,

breast cancer and colon cancer, its 5-year survival rate is only 29%. Although the morbidity of GC in China has declined in recent years, the mortality has not decreased significantly^{1 2}. The morbidity of GC still ranks second and fifth among all malignancy among men and women, and the mortality ranks third and second in China^{3 4}. At present, surgery is main treatment of GC. Perioperative chemotherapy and radiotherapy are main treatments to improve the surgical resection rate^{5 6}. However, since the high recurrence rate after surgery and the complications of other treatments, the final prognosis of patients is poor^{7 8}.

Traditional Chinese medicine has been widely used in clinical treatment of cancer in Asian countries, especially China and Japan⁹. As many clinical trials on comprehensive cancer treatment continue to be conducted, we have found that TCM not only relieves the discomfort caused by surgery and other treatments, but also alleviates the clinical symptoms of patients and improves their immune function and postoperative survival rate¹⁰⁻¹². The results of a Meta-analysis on TCM treatment improves the survival rate of GC patients in Taiwan¹³ showed that the median survival time of patients who received TCM treatment was longer than that of patients who did not receive TCM treatment, suggesting that TCM treatment improves the medium overall survival rate of GC patients. TCM has its unique advantages in treating cancer, which can effectively inhibit tumor spread and metastasis, reduce toxicity of chemotherapy drugs and enhance immunity. For thousands of years, TCM has been widely used to treat various diseases because of its multi-target, low price and few side effects¹⁴. TCM also helps to improve the quality of life among GC patients¹⁵. Piwei-Peyuan Decoction is a prescription developed by Professor Xuejun Li on the basis of Dongheng Li's theory of spleen and stomach diseases, combined with the clinical experience of Professor Ma Jun, a national famous TCM doctor. This prescription consists of six herbs including Bai Zhu, Huang Qi, Xiang Fu, Gui Zhi, Bai Shao and Liu Jinu. In our hospital, Piwei-Peyuan Decoction has shown outstanding efficacy in treatment of GC, which can effectively reduce the clinical symptoms of GC patients. However, its pharmacological mechanisms and compounds are still unclear. It is necessary for us to discover the compounds and key targets of Piwei-Peyuan Decoction in treatment of GC.

Based on genomic, proteomic and pharmacological information, a “disease-gene” biological network and a “drugs-targets” biological network can be constructed through a network pharmacology approach. After analysis, we can understand the mechanisms of drugs in treatment of diseases systematically and comprehensively^{16 17}. Through the network pharmacology method, the main targets of Piwei-Peyuan Decoction can be identified, which is helpful to further investigate the mechanisms of Piwei-Peyuan Decoction in treating GC and provide an important theoretical basis for its clinical treatment.

2. Methods

2.1 Predicting Targets of Piwei-Peyuan Decoction

To obtain the targets of Piwei-Pei yuan Decoction, TCMSP database (<https://tcmbspw.com/tcmbsp.php>) were used. TCMSP¹⁸ contains 499 Chinese herbal medicines registered in the Chinese Pharmacopoeia, including information of 29,384 ingredients, 3,311 targets and 837 related diseases. TCM are mostly oral medication, according to oral bioavailability (OB) and drug-likeness point (DL), active ingredients and related targets can be selected^{19 20}. We took $OB \geq 30\%$ and $DL \geq 0.18$ as a standard, discovered the active ingredients and related targets. Then the names of those identified targets were sent to UNIPROT database (<http://www.uniprot.org>) for normalization.

2.2 Collecting Targets Related to Gastric Carcinoma

To obtain the disease-related gene, Genecards database (<https://www.genecards.org>) and OMIM database (<http://www.omim.org>) were used. GeneCards²¹ is a comprehensive and authoritative database of human gene information that has been widely used for nearly 26 years. Its information is automatically discovered and integrated from more than 80 digital sources, including more than 73,000 human genes. We set "Gastric Carcinoma" as keywords, selected targets with hit scores greater than median (1.55) and deleted the repeated targets. Finally, GC-related targets were obtained.

2.3 Protein-Protein-Interaction Analysis

Protein-Protein-Interaction (PPI) is fundamental for most biological processes in a living cell and is crucial for understanding cell physiology in normal and disease states. Our PPI network mapping was performed on obtained bioactive ingredients and disease targets using STRING database (<http://string-db.org>) with the species limited to "homo sapiens". A targets-pathways interaction network were constructed by Cytoscape (version 3.7.2), a bioinformatics software used for data visualization and integration^{22 23}. Cytoscape is the most powerful tool when large amounts of information about the interrelationship of DNA, protein and signal pathways need to be analyzed.

2.4 GO and KEGG Pathway Enrichment

The Gene Ontology (GO) provides information for functional genomics and defines the concepts relating to gene functions²⁴. The Kyoto Encyclopedia of Genes and Genomes (KEGG) is a database that is famous for its pathway information²⁵. It is a powerful database for systematic analysis of gene function, which links genomic information to biological function information. In order to investigate the biological effects of Piwei-Pei yuan Decoction, GO analysis and KEGG pathway enrichment analysis were conducted and calculated by STRING database (<http://string-db.org>). The enriched GO terms and pathways having a corrected P value of less than 0.01 were selected for further analysis.

2.5 Molecular Docking

Based on the information from network maps of Piwei-Pei yuan Decoction and GC, we selected main ingredients and targets of Piwei-Pei yuan Decoction for molecular docking experiment. The three-dimensional (3D) structure diagrams of these compounds of Piwei-Pei yuan Decoction were downloaded

through the TCMSP database and imported into the AutodockTools1.5.6 software for hydrogenation and energy optimization, then the mol2 format files were saved. We add the charge and display rotatable keys of these compounds and the files were saved in pdbqt format. Next, the protein crystal structures corresponding to the target genes were downloaded from the PDB database, imported into PyMOL software to remove water molecules and heteromolecules, imported into AutoDockTools-1.5.6 software to add hydrogen atoms, saved to pdbqt format. Finally, the compound is used as a ligand, and the protein corresponding to the target gene is used as a receptor for molecular docking. Autodock vina 1.1.2 and PyMol software were used to analyze and interpret the results.

3. Results

3.1 Ingredients and Targets Analysis

In our study, the TCMSP database was used to predicted the ingredients of Piwei-Peyuan Decoction initially. We obtained 55 ingredients for Bai Zhu, 87 ingredients for Huang Qi, 104 ingredients for Xiang Fu, 220 ingredients for Gui Zhi, 85 ingredients for Bai Shao and 46 ingredients for Liu Jinu. Then we deleted the active ingredients without matched targets and selected the active ingredients from 6 herbs according to oral bioavailability (OB) and drug likeness (DL) values ($OB \geq 30\%$ and $DL \geq 0.18$). Eventually, 67 active ingredients such as quercetin, isorhamnetin, luteolin and kaempferol were discovered (**Table.1**), and they all have hepatoprotective, anti-inflammatory, antibacterial, analgesic and antitumor effects ²⁶⁻²⁹.

Table 1. Active ingredients of Piwei-Peyuan Decoction

Medicine	MOL ID	MOL NAME	OB(%)	DL
Bai Zhu	MOL000033	(3S,8S,9S,10R,13R,14S,17R)-10,13-dimethyl-17-[(2R,5S)-5-propan-2-yloctan-2-yl]-2,3,4,7,8,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta[a]phenanthren-3-ol	36.23	0.78
Bai Zhu	MOL000020	12-senecioid-2E,8E,10E-atractylentriol	62.4	0.22
Bai Zhu	MOL000021	14-acetyl-12-senecioid-2E,8E,10E-atractylentriol	60.31	0.31
Bai Zhu	MOL000022	14-acetyl-12-senecioid-2E,8Z,10E-atractylentriol	63.37	0.30
Bai Zhu	MOL000049	3 β -acetoxylatractylone	54.07	0.22
Bai Zhu	MOL000072	8 β -ethoxy atractylenolide	35.95	0.21
Bai Zhu	MOL000028	α -Amyrin	39.51	0.76
Huang Qi	MOL000438	(3R)-3-(2-hydroxy-3,4-dimethoxyphenyl)chroman-7-ol	67.67	0.26
	MOL000033	(3S,8S,9S,10R,13R,14S,17R)-10,13-dimethyl-17-[(2R,5S)-5-propan-2-yloctan-2-yl]-2,3,4,7,8,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta[a]phenanthren-3-ol	36.23	0.78
Huang Qi	MOL000380	(6aR,11aR)-9,10-dimethoxy-6a,11a-dihydro-6H-benzofurano[3,2-c]chromen-3-ol	64.26	0.42
	MOL000442	1,7-Dihydroxy-3,9-dimethoxy pterocarpene	39.05	0.48
	MOL000371	3,9-di-O-methylnissolin	53.74	0.48
	MOL000374	5'-hydroxyiso-muronulatol-2',5'-di-O-glucoside	41.72	0.69
	MOL000378	7-O-methylisomucronulatol	74.69	0.30
	MOL000379	9,10-dimethoxypterocarpan-3-O- β -D-glucoside	36.74	0.92
	MOL000387	Bifendate	31.10	0.67
Huang Qi	MOL000417	Calycosin	47.75	0.24
	MOL000433	FA	68.96	0.71
	MOL000392	Formononetin	69.67	0.21
Huang Qi	MOL000296	Hederagenin	36.91	0.75
	MOL000398	Isoflavanone	109.99	0.30
Huang Qi	MOL000439	Isomucronulatol-7,2'-di-O-glucoside	49.28	0.62
	MOL000354	Isorhamnetin	49.60	0.31
Huang Qi	MOL000239	Jaranol	50.83	0.29
	MOL000422	Kaempferol	41.88	0.24

Bai Shao	MOL000211	Mairin	55.38	0.78
Bai Shao	MOL001910	11alpha,12alpha-epoxy-3beta-23-dihydroxy-30-norolean-20-en-28,12beta-olide	64.77	0.38
Bai Shao	MOL001928	albiflorin_qt	66.64	0.33
Bai Shao	MOL001925	Paeoniflorin_qt	68.18	0.40
	MOL001918	Paeoniflorgenone	87.59	0.37
Bai Shao				
Bai Shao				
Bai Shao				
Xiang Fu	MOL003044	Chryseriol	35.85	0.27
Xiang Fu	MOL000354	Isorhamnetin	49.60	0.31
Xiang Fu	MOL003542	8-Isopentenyl-kaempferol	38.04	0.39
Xiang Fu	MOL000358	Beta-sitosterol	36.91	0.75
Xiang Fu	MOL000359	Sitosterol	36.91	0.75
Xiang Fu	MOL004027	1,4-Epoxy-16-hydroxyheneicos-1,3,12,14,18-Pentaene	45.10	0.24
	MOL004053	Isodalbergin	35.45	0.20
Xiang Fu	MOL004058	Khell	33.19	0.19
Xiang Fu	MOL004059	Khellol glucoside	74.96	0.72
Xiang Fu	MOL010489	Resivit	30.84	0.27
Xiang Fu	MOL004068	Rosenonolactone	79.84	0.37
Xiang Fu	MOL004071	Hyndarin	73.94	0.64
Xiang Fu	MOL004074	Stigmasterol glucoside_qt	43.83	0.76
Xiang Fu	MOL004077	Sugeonyl acetate	45.08	0.20
Xiang Fu	MOL000422	Kaempferol	41.88	0.24
Xiang Fu	MOL000449	Stigmasterol	43.83	0.76
Xiang Fu	MOL000006	Luteolin	36.16	0.25
Xiang Fu	MOL000098	Quercetin	46.43	0.28
Gui Zhi	MOL000073	ent-Epicatechin	48.96	0.24

Gui Zhi	MOL000492	(+)-Catechin	54.83	0.24
Gui Zhi	MOL001736	(-)-Taxifolin	60.51	0.27
Gui Zhi	MOL004576	Taxifolin	57.84	0.27
Gui Zhi	MOL000359	Sitosterol	36.91	0.75
Gui Zhi	MOL011169	Peroxyergosterol	44.39	0.82
Liu Ji'nu	MOL001733	EUPATORIN	30.23	0.37
Liu Ji'nu	MOL000358	Beta-sitosterol	36.91	0.75
Liu Ji'nu	MOL000006	Luteolin	36.16	0.25
Liu Ji'nu	MOL008127	Ermanin	58.95	0.30
Liu Ji'nu	MOL008135	3,4-di-O-caffeoylquinic acid	49.62	0.69

3.2 Targets of Gastric Carcinoma and Piwei-Peiyan Decoction

9877 targets of GC were obtained by Genecards database. We selected targets with hit scores greater than median 1.55. Finally, after integrating the target information in the database, 4938 targets were saved.

3.3 Construction of Interaction Network Maps

Based on the targets of Piwei-Peiyan Decoction and GC, 181 common targets shown in the venn diagram were recognized as targets of Piwei-Peiyan Decoction in treatment of GC (**Figure 1**). A PPI network was constructed based on STRING database (**Figure 2**), and a Targets-Pathways Interaction Network was built based on Cytoscape 3.7.1 (**Figure 3**). The obtained targets, such as MAPK1, TP53, IL-1, Fox and Bax, are all involved in anti-inflammatory, cell proliferation-promoting, angiogenic, and anti-tumor processes³⁰⁻³³.

3.4 GO and KEGG Pathway Enrichment Analysis

GO enrichment analysis consists three parts: Biological Process, Molecular Function and Cellular Component. We sent 181 targets information to the STRING database for GO analysis. Finally, 2311 biological process terms, 139 cellular component terms and 259 molecular function terms were found (**Figure 4**). Based on the correct P value, we found the top 5 terms in cellular component were cell (174), intracellular (166), organelle (151), intracellular organelle (150), membrane-bounded organelle (149). And top 5 terms in biological processes were cellular process (178), biological regulation (174), response to stimulus (173), regulation of biological process (168), regulation of cellular process (165). Top 5 terms in molecular funtions were binding (173), protein binding (145), ion binding (111), catalytic activity (96), organic cyclic compound binding (92).

To further uncover the potential pharmacological mechanisms of Piwei-Peiyan Decoction against GC, pathway analysis was conducted to explore the potential pathways affected by Piwei-Peiyan Decoction. 190 pathways were found by KEGG database, we selected top 20 pathways and draw a bubble diagram as follows (**Figure 5**). Combining the pathogenesis of GC, the pathways which have no association with GC were removed. Finally, 6 remarkable terms were found to be the related pathways in treatment of GC. Results demonstrated that “Pathway in cancer”, “PI3K-Akt signaling pathway”, “MAPK signaling pathway”, “Ras signaling pathway”, “IL-17 signaling pathway”, “HIF-1alpha signaling pathway” and “TNF signal pathway” were obviously enriched. According to the KEGG analysis, We found “Pathway in cancer” is the signal pathway that contains the most targets (72 targets), the network of “Pathway in cancer” is shown below (**Figure 6**). These signaling pathways are closely related to cell differentiation, proliferation, apoptosis and angiogenesis, most of which play a key role in the development and progression of cancer. The molecular functions and biological processes were closely related to the occurrence and development of GC, which indicated that Piwei-Peiyan Decoction can treat GC through multiple targets and pathways. Our research shows that Piwei-Peiyan Decoction acts as a treatment for GC mainly through the coordinated regulation of cancer-related signal pathways. For example, PI3K-Akt signaling pathway is a key pathway that regulates cell proliferation, differentiation and metastasis during the development of cancer³⁴, and activated AKT can lead to apoptosis by participating in the regulation of cellular protein expression, then ultimately inhibiting cell proliferation³⁵. It has been shown that the abnormal expression of P13K-Akt signaling pathway in GC tissues is closely related to the development and prognosis of tumor, P13K-AKT signal pathway plays an important role in regulating the proliferation, invasion and metastasis of tumor cells. Meanwhile, the high expression of P13K-Akt signaling pathway in GC is related to the degree of tumor differentiation, its expression level was positively correlated with the malignancy of GC³⁶⁻³⁸. MAPK signal pathway is expressed in almost all eukaryotes. It participates in cellular activities such as gene expression, cell proliferation and apoptosis, playing a key role in cellular activities³⁹. ERK signaling pathway is a significant part of MAPK signal pathway, the gene mutations or abnormal activation in ERK signal pathway can lead to development of cancer⁴⁰. Ras is a binding kinase in the upstream of ERK/MAPK signal pathway, its point mutations can lead to dysregulation of ERK/MAPK signal pathway and abnormal cellular activity, which in turn lead to migration and invasion of cancer cells and ultimately induces the development of tumor^{41 42}. IL-17 is an inflammatory factor with great anti-inflammatory effects. It participates in the processes of cell proliferation and differentiation, immune regulation, and tumor growth. A study⁴³ showed that the median serum IL-17 level in GC patients was significantly higher than that in controls (9.04 VS 8.07 pg/ml, $p=0.01$), which shows that serum IL-17 level can be used as a new potential indicator for GC diagnosis. Xu J et al⁴⁴ found that IL-17-mediated downregulation of LCN2 expression inhibited the proliferation, migration, and invasion of GC cells by regulating SLPI. In addition to IL-17A, a member of the IL-17 family, several experiments have confirmed that IL-17B/IL-17 receptor B (IL-17RB) can inhibit growth and progression of tumor. A recent study⁴⁵ by Bastid J et al showed that IL-17B promotes tumor progression by promoting the secretion of chemokines and cytokines and thereby dramatically altering the tumor microenvironment. Furthermore, in GC patients, higher serum IL-17B levels have been shown to be strongly associated with poor prognostic outcomes.

Therefore, it is clear that IL-17 levels are closely associated with the development of GC. However, few pharmacological findings have been reported for the treatment of GC with Piwei-Peiyuan Decoction. Therefore, more experiments should be conducted to validate our findings in the future.

3.5 Results of Molecular Docking Experiment

Molecular docking was used for verification of interaction between ingredient and its target gene. The results of molecular docking scores of the ingredients and target genes we selected in this study were shown in **Table.2**, and the optimal schematic of molecular docking was shown in **Figure.7**.

From the results, the lowest binding free energy of caspase-3 and kaempferol is -6.1 kcal/mol. There is one hydrogen bond between amino acid THR62 and kaempferol. The lowest binding free energy of caspase-3 and luteolin is -6.1 kcal/mol. There are hydrophobic interactions between amino acid THR62, SER251 and luteolin. In addition, the lowest binding free energy of MAPK1 and luteolin is -7.9 kcal/mol. There are hydrophobic interactions between amino acid LYS54, GLN105, MET108, ASP167 and luteolin. The lowest binding free energy of caspase-3 and quercetin is -6.0 kcal/mol. There are two hydrophobic bond forces between amino acid THR62 and quercetin. Moreover, The lowest binding free energy of MAPK1 and quercetin is -8.1 kcal/mol. There are two hydrophobic bond forces between amino acid ASP106, MET108 and quercetin. All compounds showed a compact binding pattern to the protein active pocket, and these interactions enabled the proteins to form stable complexes with all compounds. In this study, molecular docking experiment gives explanation for the way of protein-compound interactions and lays the theoretical foundation for further studies of Piwei-Periyuan Decoction in treatment of GC.

Table.2 Scores of Molecular Docking

Receptor_Name	Ligand_Name	Scores (kcal/mol)
CASP3	kaempferol.pdbqt	-6.1
CASP3	luteolin.pdbqt	-6.1
CASP3	quercetin.pdbqt	-6.0
MAPK1	luteolin.pdbqt	-7.9
MAPK1	quercetin.pdbqt	-8.1

Note: A higher absolute value of scores means that the binding of receptor protein and active ingredient is more stable.

4. Conclusion

In this study, we investigated the mechanism of Piwei-Peiuyan Decoction in treatment of GC using a network pharmacology approach. The results showed that Piwei-Peiuyan Decoction exerted its excellent pharmacological effects in treatment of GC through various pathways, such as regulation of cell cycle, promotion of tumor cell apoptosis and immunomodulation. Our study also provided a theoretical basis for the clinical application of Piwei-Peiuyan Decoction, which is of clinical value to further explore the role of TCM in prevention and treatment of cancer in the future.

5. Discussion

GC is one of the most common cancers in the world. At present, the mechanism of GC is not fully understood. However, according to the patient's pathogenesis and clinical symptoms, in the theory of TCM, GC is caused by weakness of body, bad dietary habits and emotional disorders. Several studies have shown that surgery, radiotherapy and chemotherapy for GC are ineffective, while TCM treatment has its own advantages. It has great potential in improving immunity of body and alleviating symptoms caused by GC⁴⁶. Although Piwei-Peiuyan Decoction has been widely used in treatment of GC, there is no research on its specific mechanisms in treatment of GC. In our study, a network pharmacology method was used to predict and discover the potential mechanisms of Piwei-Peiuyan Decoction in treatment of GC. From this experiment, it can be seen that quercetin, isorhamnetin, luteolin and kaempferol may be the key components of Piwei-Peiuyan Decoction, and cell cycle regulation-related proteins play an important role in the development of GC. Quercetin is a bioflavonoid with anti-inflammatory, antiviral, and anti-tumor effects, and it has been shown to exert inhibitory activity on growth of GC cells through various mechanisms⁴⁷⁻⁴⁹. Quercetin can regulate the expression of caspases family and Bcl-2 family proteins to induce cell apoptosis through MAPK, ERK, PI3K, PKC and other signaling pathways⁵⁰⁻⁵³. Shang HS et al⁵⁴ found that quercetin could induce apoptosis in AGS cells by causing changes in cell morphology and thus reduce the overall survivability of GC cells. Kaempferol is an important flavonoid widely found in vegetables and fruits, which induces cell cycle arrest and promotes cell apoptosis⁵⁵⁻⁵⁹. It has been shown to inhibit the proliferation and metastasis of a variety of tumor cells. Yang L et al⁶⁰ found that kaempferol treats GC mainly through three key targets, ESR1, EGFR and SRC. The expression levels of EGFR and SRC were differentially elevated in GC tissues, and high expression of these targets could directly affect the prognostic survival of GC patients. It was found that apoptosis and G2/M phase cell cycle arrest were observed in tumor cells after kaempferol treatment, and the expression levels of G2/M cell cycle regulators, cyclin B1, Cdk1 and Cdc25C were significantly reduced. In addition, after kaempferol treatment, Bcl-2 expression levels were significantly reduced while Bax expression levels were increased, these changes led to the upregulation of caspase-3 and caspase-9, which promoted apoptosis and finally inhibited the development of GC⁶¹. Isorhamnetin is one of the most important active ingredients in hippophae rhamnoides fruits and ginkgo biloba leaves, with a wide range of pharmacological activity, which can exert anti-inflammatory, anti-tumor and antioxidant effects through the regulation of PI3K/AKT/PKB, NF- κ B, MAPK signaling pathways and the expression of related cytokines and kinases⁶². Ramachandran L et al⁶³ found that isorhamnetin can reduce the migratory and invasive properties of GC cells by regulating peroxisome proliferator-activated receptor γ (PPAR- γ), thereby inhibiting GC cell

proliferation and inducing cell apoptosis. In addition, the combination of isorhamnetin and chemotherapy drugs can enhance the inhibition of tumor cell proliferation. Luteolin is a common flavonoid in plants, which significantly inhibits cell cycle progression, proliferation, migration, invasion and promotes cell apoptosis⁶⁴. Luteolin exerts its anti-tumor, antioxidant, and anti-inflammatory effects mainly through the regulation of signaling pathways such as Notch1, PI3K, AKT and ERK signal pathway. A study has been found that high expression of Notch1 was closely associated with low overall survival rate of GC patients⁶⁵. However, Luteolin could inhibit GC progression by inhibiting Notch1 signaling pathway and reversing epithelial-mesenchymal transition (EMT). It suggests that luteolin may be an effective anti-tumor compound in treatment of GC. KEGG pathway enrichment analysis showed that the target genes of Piwei-Peiyan Decoction for the treatment of GC were mainly involved in Pathway in cancer, PI3K-Akt signaling pathway, MAPK signaling pathway, IL-17 signaling pathway, Ras signaling pathway, HIF-1 signaling pathway and TNF signaling pathway. In summary, it can be seen that the mechanism of Piwei-Peiyan Decoction in treatment of GC is to inhibit cell proliferation and promote cell apoptosis by interfering with tumor-related signaling pathways, thus exerting anti-tumor effects.

Abbreviations

AKT
Protein Kinase B
Bai Shao
Radix Paeoniae Alba
Bai Zhu
Rhizoma Atractylodis Macrocephalae
DL
Drug Likeness
EGFR
Epidermal Growth Factor Receptor
ESR
Estrogen Receptor
GC
Gastric Carcinoma
GO
Gene Ontology
Gui Zhi
Ramulus Cinnamomi
HIF
Hypoxia Inducible Factor
Huang Qi
Radix Astragali
IL

Interleukin
KEGG
Kyoto Encyclopedia of Genes and Genomes
Liu Jinu
Herba Artemisiae Anomala
MAPK
Mitogen-activated Protein Kinase
OB
Oral Bioavailability
OMIM
Online Mendelian Inheritance in Man
PKB
Protein Kinase B
PPI
Protein-Protein Interaction Analysis
TCM
Traditional Chinese Medicine
TCMSP
Traditional Chinese Medicine Systems Pharmacology Database and Analysis Platform
TNF
Tumor Necrosis Factor
Xiang Fu
Cyperus Rotundus

Declarations

Contributions

XL conceived and designed research. BP, ZW and YZ conducted experiments and collected data. BP analyzed data and wrote the manuscript. All authors read and approved the manuscript.

Besides, we declare that all data were generated in-house and that no paper mill was used.

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Ethics Approval

Not applicable.

Conflicts of Interest

There are no conflicts of interest to declare.

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Figures

Figure 1

Venn diagram. Note: We found 4938 targets genes of GC and 236 targets of Piwei-Peiyan Decoction. 181 common targets are common targets.

Figure 3

Targets-Pathways Interaction Network.

Gene Ontology Analysis

Figure 4

Gene Ontology Analysis of Targets Genes

Figure 5

KEGG Pathway Analysis of Target Genes

Figure 6

Network of Pathway in Cancer Signal Pathway

CASP3-Kaempferol

(a)

CASP3-Luteolin

(b)

CASP3-Quercetin

(c)

MAPK1-Luteolin

(d)

MAPK1-Quercetin

(e)

Figure 7

Molecular Docking of Key Targets and Ingredients. Note: Docking concept and binding site analysis of Kaempferol (a). Docking concept and binding site analysis of Luteolin (b, d). Docking concept and binding site analysis of Quercetin (c, e).