

Study on the Action Mechanism of *Isodon suzhouensis* Stems in the Treatment of Prostate Cancer Based on UPLC-Q-TOF-MS/MS Combined with Network Pharmacology and Molecular Docking

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Abstract [**Objectives**] This study was conducted to explore the action mechanism of chemical components from stems of *Isodon suzhouensis* on prostate cancer based on UPLC-Q-TOF-MS/MS technique, network pharmacology, and molecular docking validation. [**Methods**] UPLC-Q-TOF-MS/MS was applied to search active components in stems of *I. suzhouensis*, and SwissTargetPrediction and PharmaMapper databases were used to predict their potential targets. The GeneCards database was adopted to screen the targets of prostate cancer, and the targets of the active components and the targets of prostate cancer were intersected to obtain common targets. A protein-protein interaction network was constructed, and also, an "active component-target-disease-pathway" network was constructed using Metascape for gene ontology function and Kyoto Encyclopedia of Genes and Genomes pathway enrichment analysis. Moreover, AutoDock was adopted to perform molecular docking verification on key active components and core targets. [**Results**] Sixty three active components were screened from stems of *I. suzhouensis*, and they had 233 common targets with prostate cancer, mainly involving biological processes such as protein phosphorylation, response to hormones, active regulation of cell movement, cell response to lipids, and response to oxidative stress, as well as pathways in cancer, prostate cancer, micro ribonucleic acids in cancer, p53 signaling pathway, NF- κ B signaling pathway and other signaling pathways, exerting anti-tumor effects. Molecular docking showed that the top 5 key active components had good binding ability with the top 5 core targets. [**Conclusions**] Active components from stems of *I. suzhouensis* can exert therapeutic effects on prostate cancer through multiple targets and pathways.

Key words Stem of *Isodon suzhouensis*; UPLC-Q-TOF-MS/MS; Network pharmacology; Molecular docking; Prostatic cancer

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Isodon suzhouensis is a perennial herbaceous plant in the Labiatae family, belonging to the genus *Isodon*. It is one of few medicinal and edible Chinese herbal plants in the genus *Isodon*^[1]. *I. suzhouensis* is pungent in nature and bitter in taste, and attributed to the liver, lung, stomach and kidney meridians. Its functions are clearing away heat and toxic materials, inducing diuresis and reducing edema, and promoting blood circulation and removing blood stasis. *I. suzhouensis* is used for damp-heat jaundice, sore throat, lung abscess, epigastric pain, stranguria, edema, arthralgia, amenorrhea, mammary abscess, hemorrhoid, traumatic injury, and venomous snake bites, and it is used for treating various diseases such as malignant sores, tumors, venomous snake bites, tuberculosis, nephritis, pharyngitis, colds, and acute infectious hepatitis in the folk^[2].

Prostate cancer is the most common male malignant tumor in Europe and America, and its incidence rate is the first among male malignant tumors in the United States, and its mortality rate is second only to lung cancer^[3]. With the development and progress of

our society and the extension of the average life span of the population, the incidence of prostate cancer is increasing significantly. At present, the early diagnosis of prostate cancer mainly depends on the level of serum prostate-specific antigen (PSA)^[4]. However, as an early diagnostic marker, PSA still lacks the best specificity and sensitivity. Therefore, finding new specific molecular diagnostic markers for prostate cancer to assist or eventually replace PSA has been a hot topic in recent years. Studies on the occurrence of various cancers at the cellular and molecular levels show that RecQ helicases serve as an important family of DNA helicases, which plays a key role in the process of nucleic acid metabolism^[5].

At present, the action mechanism of active components in stems of *I. suzhouensis* with prostate cancer is still uncertain. Therefore, based on liquid chromatography-mass spectrometry (LC-MS) combined with network pharmacology and molecular docking techniques, the active components of *I. suzhouensis* were analyzed, and their active targets and action mechanism for treating prostate cancer were analyzed and predicted to explore the pharmacologic material basis of *I. suzhouensis*. This study provides reference for the quality control of *I. suzhouensis*, and also some theoretical reference for exploring the treatment of prostate cancer with stems of *I. suzhouensis*.

Materials and Methods

Experimental materials

Medicinal materials The medicinal materials were purchased

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from Bozhou medicinal materials market in Anhui Province and kept in the pharmaceutical laboratory of People's Hospital of Anshun City. They were identified as *I. suzhouensis* of *Isodon* in Labiatae by Yan Chen from the pharmaceutical laboratory of People's Hospital of Anshun City.

Samples and reagents Sample treatment: First, 50 mg of *I. suzhouensis* stems was extracted by refluxing with 10 times of ethanol for 3 times, 3 h each time, and the extracts were combined. After filtration, the filtrate was concentrated under reduced pressure to recover ethanol and obtain a thick paste. Next, 10 mg of the sample was dissolved in 70% methanol water, and filtered through a 0.22 μm organic microporous filter membrane to obtain a sample solution of *I. suzhouensis* as the sample for LC-MS analysis.

Reagents: 95% ethanol; methanol (chromatographically pure); acetonitrile (chromatographically pure).

Instruments and conditions Chromatographic conditions: ACE Ultracore2.5 SuperC18 (100 mm \times 2.1 mm); mobile phase: acetonitrile (0.1% formic acid)-0.1% formic acid water; gradient elution (0–2 min, 5% acetonitrile; 2–47 min, 95% acetonitrile; 47–50 min, 5% acetonitrile); volume flow rate: 0.3 ml/min; column temperature: 40 $^{\circ}\text{C}$.

Mass spectrometry conditions: HESI-II ion source; ion source voltage: 3.0 kV (+)/2.5 kV (-); capillary heating temperature: 320 $^{\circ}\text{C}$; sheath gas flow rate: 35 arb; auxiliary gas flow rate: 10 arb; ion source temperature: 350 $^{\circ}\text{C}$; mass spectrometry scanning method: Full MS dds2; scanning range: 100–1500 m/z; primary resolution of Full MS: 70000, and secondary resolution: 17500.

Data analysis: Searching was conducted with Compound discover software from Thermo Fisher Scientific.

Methods

Search and screening of active components The UPLC-Q-TOF-MS/MS method was used to retrieve active components from stems of *I. suzhouensis*. Names and structural information of the compounds were obtained from PubChem database^[6] (<https://pubchem.ncbi.nlm.nih.gov>), and the 2D structures were derived and saved as sdf structure. The compound structures were uploaded into Swiss Target Prediction database^[7] (<http://swisstargetprediction.ch/>), and the "probability" value was set as "probability" > 0.01. The PharmMapper database^[8–10] (<http://www.lilab-ecust.cn/pharmmapper/>) was used to supplement the target information of compounds that had not been collected, and the Norm Fit value was set as Norm Fit > 0.9 to predict and analyze target information of compounds. All the target information was summarized, and the Uniprot (<https://www.uniprot.org>) database^[11] was used to correct names of target genes for chemical components, so as to standardize the protein target information.

Prediction of prostate cancer targets and acquisition of drug-disease common targets In the GeneCards database platform^[19], related genes were collected using "Prostate cancer" as the keyword and setting Relevance scores > 15, and duplicate

values were deleted. The active component targets of *I. suzhouensis* stems and prostate cancer-related targets were intersected using Venny (<http://www.liuxiaoyuuan.cn/>) to obtain common targets.

Construction of protein-protein interaction (PPI) network

The obtained common targets of prostate cancer and active components from *I. suzhouensis* stems were imported into the String online platform^[20] (<https://cn.string-db.org/>) for analysis. The results were saved in the TSV format and imported into Cytoscape 3.7.0 software for visual analysis. The function of "Network Analyzer" was applied to perform network topology analysis on the PPI results, and a PPI network was constructed. Key targets were screened by CytoNCA, a plugin in Cytoscape 3.7.0 software.

GO and KEGG pathway analysis The TSV format file saved in the String database was processed. node1 and node2 were merged, and duplicate values were deleted. Next, the data were imported into Metascape database^[14] (<http://metascape.org/gp/index.html#/main/step1>) to perform analysis by selecting GO Biological Processes, GO Cellular Components, GO Molecular Functions and KEGG Pathway, respectively. And the Bioinformatics online platform (<http://www.bioinformatics.com.cn/>) was used for visual mapping analysis.

Construction of *I. suzhouensis* stem active component-target-disease-pathway network diagram

The collected common targets from active components of *I. suzhouensis* stems and prostate cancer, the active components, prostate cancer, the top 20 KEGG pathways and their related target data were integrated into Excel files. Next, the Excel files were imported into Cytoscape 3.7.0 software to construct an "active component-target-disease-pathway" network.

Molecular docking The top five key active components in the "active ingredient-target-disease-pathway" network were molecularly docked with the top five core targets in the PPI network. Appropriate protein structures were screened from Uniprot database, and then, the protein structures were downloaded from PDB (<https://www.rcsb.org/>) database^[15]. Small molecule structure files were downloaded from TCMSp (<https://old.tcmsp-e.com/tcmsp.php>) database^[16], and Pymol software was used to delete water from protein receptors and solvent molecules from small molecules. Autodock software was used to add all hydrogens to proteins and small molecules, respectively. Next, molecular docking was performed after setting as receptors and ligands and setting twisted bonds, and results were obtained by calculation.

Results and Analysis

Extraction of active components from stems of *I. suzhouensis*

The total ion current (TIC) diagram of positive and negative ion modes was obtained from rapid analysis using UPLC-Q-TOF-MS/MS, as shown in Fig. 1. Based on the compound information generated by mass spectrometry combined with secondary fragment ions and relevant references, qualitative analysis was conducted on chemical components of *I. suzhouensis* stems, and a total of 63

compounds were identified from stems of *I. szechouensis*. The retained components, molecular formulas, compound names,

fragment ion information and other data of chemical components are shown in Table 1.

Table 1 Results of composition identification of *I. szechouensis* stems

No.	Compound	Chemical formula	Molecular weight	Ion product	Parent ion peak
1	Nepetin	C ₁₆ H ₁₂ O ₇	316	[M - H] ⁻¹	315.1
2	Kahweol	C ₂₀ H ₂₆ O ₃	314	[M + H] ⁺¹	315.2
3	Rutin	C ₂₇ H ₃₀ O ₁₆	610	[M - H] ⁻¹	609.1
4	Vicenin II	C ₂₇ H ₃₀ O ₁₅	594	[M - H] ⁻¹	593.1
5	Caffeic acid	C ₉ H ₈ O ₄	180	[M - H] ⁻¹	179.0
6	Hydroxygenkwanin	C ₁₆ H ₁₂ O ₆	300	[M - H] ⁻¹	299.1
7	Jaceosidin	C ₁₇ H ₁₄ O ₇	330	[M - H] ⁻¹	329.1
8	Isoquercitrin	C ₂₁ H ₂₀ O ₁₂	464	[M - H] ⁻¹	463.1
9	Ethyl caffeate	C ₁₁ H ₁₂ O ₄	208	[M - H] ⁻¹	207.1
10	Tectoridin	C ₁₆ H ₁₂ O ₆	300	[M + H] ⁺¹	301.1
11	Salvianic acid A	C ₉ H ₁₀ O ₅	198	[M - H] ⁻¹	197.0
12	Salvianolic acid C	C ₂₆ H ₂₀ O ₁₀	492	[M - H] ⁻¹	491.1
13	Isosteviol	C ₂₀ H ₃₀ O ₃	336	[M + H - H ₂ O] ⁻¹	319.2
14	Ligustilide	C ₁₂ H ₁₄ O ₂	190	[M + H] ⁺¹	191.1
15	Esculetine	C ₉ H ₆ O ₄	178	[M - H] ⁻¹	177.0
16	Echinocystic acid	C ₃₀ H ₄₈ O ₄	472	[M - H] ⁻¹	471.3
17	Apigenin	C ₁₅ H ₁₀ O ₅	270	[M + H] ⁺¹	271.1
18	Rosmarinic acid	C ₁₈ H ₁₆ O ₈	360	[M - H] ⁻¹	359.1
19	L-Valine	C ₅ H ₁₁ NO ₂	117	[M + H] ⁺¹	118.1
20	Tsugaric acid A	C ₃₂ H ₅₀ O ₄	498	[M - H] ⁻¹	497.4
21	Morin	C ₁₅ H ₁₀ O ₇	302	[M + H] ⁺¹	303.0
22	Kaempferol 3-rutinoside	C ₂₇ H ₃₀ O ₁₅	594	[M - H] ⁻¹	593.1
23	D-Glyceric acid	C ₃ H ₆ O ₄	166	[M - H] ⁻¹	165.0
24	Lithospermic acid	C ₂₇ H ₂₂ O ₁₂	538	[M - H] ⁻¹	537.1
25	Protocatechualdehyde	C ₇ H ₆ O ₃	138	[M - H] ⁻¹	137.0
26	Lupenone	C ₃₀ H ₄₈ O	424	[M + H] ⁺¹	425.4
27	4-Dicaffeoylquinic Acid	C ₁₆ H ₁₈ O ₉	354	[M - H] ⁻¹	353.1
28	α-Linolenic acid	C ₁₈ H ₃₀ O ₂	278	[M + H] ⁺¹	279.2
29	5-Hydroxymethylfurfural	C ₆ H ₆ O ₃	126	[M + H] ⁺¹	127.0
30	Rutecarpine	C ₁₈ H ₁₃ N ₃ O	287	[M + H] ⁺¹	288.1
31	7-Hydroxycoumarin	C ₉ H ₆ O ₃	162	[M - H] ⁻¹	161.0
32	Genkwanin	C ₁₆ H ₁₂ O ₅	284	[M - H] ⁻¹	283.1
33	Salicylic acid	C ₇ H ₆ O ₃	138	[M - H] ⁻¹	137.0
34	Orsellinic acid	C ₈ H ₈ O ₄	168	[M - H] ⁻¹	167.0
35	Methyl hexadecanoate	C ₁₇ H ₃₄ O ₂	316	[M - H] ⁻¹	315.3
36	Luteolin	C ₁₅ H ₁₀ O ₆	304	[M - H - H ₂ O] ⁻¹	285.0
37	Lithospermic acid	C ₂₇ H ₂₂ O ₁₂	538	[M - H] ⁻¹	537.1
38	Senkyunolide H	C ₁₂ H ₁₆ O ₄	206	[M + H] ⁺¹	207.1
39	Astragalin	C ₂₁ H ₂₀ O ₁₁	448	[M - H] ⁻¹	447.1
40	Dehydrodiisoeugenol	C ₂₀ H ₂₂ O ₄	294	[M + H] ⁺¹	295.1
41	Bayogenin	C ₃₀ H ₄₈ O ₅	488	[M - H] ⁻¹	487.3
42	Methyl rosmarinate	C ₁₉ H ₁₈ O ₈	374	[M - H] ⁻¹	373.1
43	3-Acetyl-11-keto-beta-boswellic acid	C ₃₂ H ₄₈ O ₅	512	[M + H] ⁺¹	513.4
44	Roburicacid	C ₃₀ H ₄₈ O ₂	440	[M + H] ⁺¹	441.4
45	N-Butylidenephthalide	C ₁₂ H ₁₂ O ₂	188	[M + H] ⁺¹	189.1
46	Oleanonic acid	C ₃₀ H ₄₆ O ₃	454	[M + H] ⁺¹	455.4
47	Glycyrrhetic acid	C ₃₀ H ₄₆ O ₄	470	[M + H] ⁺¹	471.3
48	Kurarinone	C ₂₆ H ₃₀ O ₆	438	[M + H] ⁺¹	439.2
49	Leucinic acid	C ₆ H ₁₂ O ₃	132	[M - H] ⁻¹	131.1

(Continued)

(Table 1)

No.	Compound	Chemical formula	Molecular weight	Ion product	Parent ion peak
50	Ferulic acid	C ₁₀ H ₁₀ O ₄	194	[M - H] ⁻¹	193.0
51	Handelin	C ₃₂ H ₄₀ O ₈	514	[M + Na] ⁺¹	553.4
52	Kaempferol	C ₁₅ H ₁₀ O ₆	286	[M + H] ⁺¹	287.1
53	Gentisuric acid	C ₇ H ₆ O ₄	154	[M - H] ⁻¹	153.0
54	Nootkatone	C ₁₅ H ₂₂ O	218	[M + H] ⁺¹	219.2
55	Pectolarigenin	C ₁₇ H ₁₄ O ₆	314	[M - H] ⁻¹	313.1
56	Chrysin	C ₁₅ H ₁₀ O ₄	254	[M + H] ⁺¹	255.1
57	Tetramethylcurcumin	C ₂₅ H ₂₈ O ₆	424	[M - H] ⁻¹	423.2
58	Perillartine	C ₁₀ H ₁₅ NO	165	[M + H] ⁺¹	166.1
59	Gamma-Aminobutyric acid	C ₄ H ₉ NO ₂	103	[M + H] ⁺¹	104.1
60	L(+)-Leucinol	C ₆ H ₁₅ NO	117	[M + H] ⁺¹	118.1
61	Polygalacic acid	C ₂₉ H ₄₄ O ₆	488	[M - H] ⁻¹	487.3
62	Wangzaozin A	C ₂₀ H ₃₀ O ₄	334.0	[M - H] ⁻¹	333.2
63	Glaucocalyxin B	C ₂₀ H ₂₈ O ₄	332.0	[M - H] ⁻¹	331.2

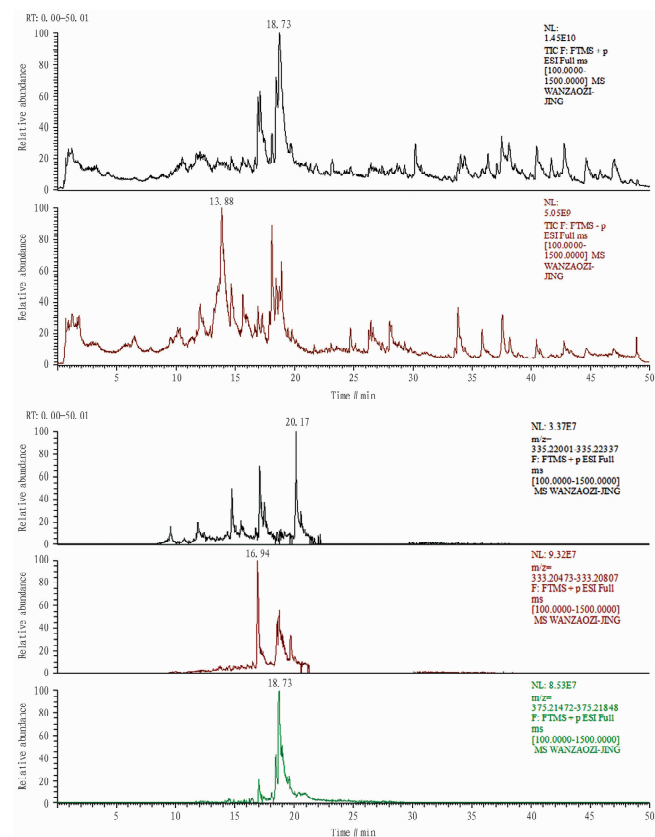


Fig. 1 Total ion current (TIC) diagram

Common targets from active components in stems of *I. suzhouensis* and prostate cancer

Sixty three chemical components were obtained by UPLC-Q-TOF-MS/MS, and 62 active components were obtained by Swiss Target Prediction and PharmMapper, while wzz-47 acquired no active targets. Totally, 684 targets were obtained after deduplication. Potential targets of prostate cancer were collected in GeneCards database, and 1 565 targets were obtained after deduplication. The targets of active components and those of prostate cancer were intersected, resulted in 233 common targets, which were the

potential anti-prostate cancer targets of the active components in stems of *I. suzhouensis*, as shown in Fig. 2.

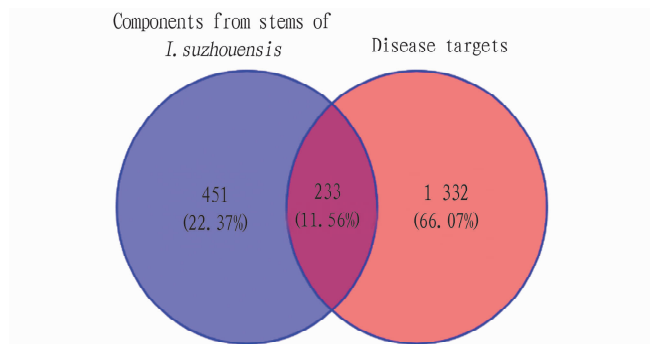
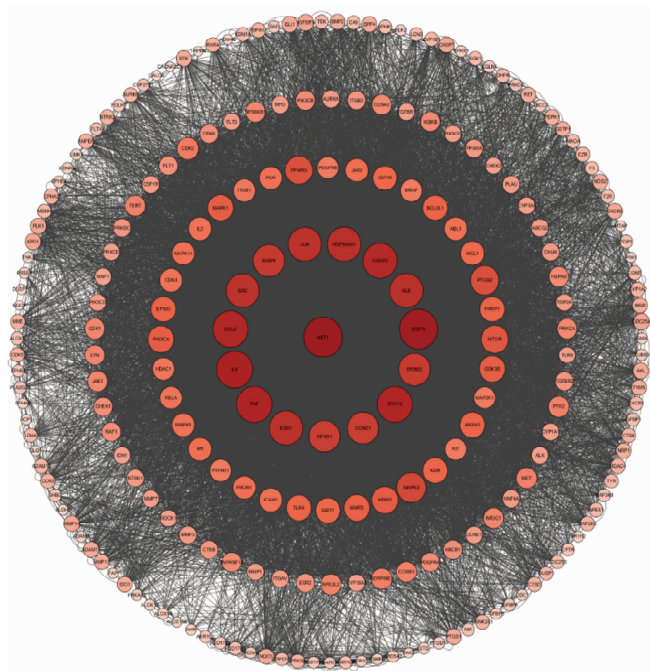
Fig. 2 Potential anti-prostate cancer targets of active components from stems of *I. suzhouensis*

Fig. 3 PPI network

Construction of PPI network

The common targets were imported into the String database to obtain a PPI network using Cytoscape software, involving a total of 233 nodes and 10 914 edges, as shown in Fig. 3. Higher degree values and larger nodes indicated that the nodes were more important in the network. The top 5 targets sorted by degree values were selected as core targets: AKT1, EGFR, IL6, TNF, and MMP9.

The enrichment results of KEGG pathways showed that 198 pathways were involved, and the bubble diagram of the top 20 KEGG pathways was drawn using LogP values, as shown in Fig. 5. The results showed that active components in stems of *I. szechouensis* for prevention and treatment of prostate cancer were mainly distributed in the cancer pathway, prostate cancer, micro ribonucleic acids in cancer, AGE-RAGE signal pathway in diabetes complications, toxoplasmosis, bladder cancer, axonal guidance, steroid hormone biosynthesis, transcriptional disorders in cancer, platinum resistance, *etc.* These pathways were closely related to the treatment of prostate cancer.

Construction of "active component-target-disease-pathway" network

Cytoscape software was employed to construct an "active component-target-disease-pathway" network, as shown in Fig. 6. The CytoNCA plugin and the "Network Analyzer" function were used for topology analysis, and the degree centrality (DC), betweenness centrality (BC) and closeness centrality (CC) were calculated. The top 5 active components were obtained as key active components: wzz-21 (morin), wzz-36 (luteolin), wzz-7 (jaceosidin), wzz-32 (genkwarin) and wzz-1 (nepetin), as shown in Table 2.

Table 2 Topological analysis of key active components

	Degree	Betweenness	Closeness
wzz-21	56.0	4 150.086 4	0.075 077 21
wzz-36	55.0	2 916.264	0.075 041 555
wzz-7	53.0	2 926.052	0.074 970 34
wzz-32	53.0	4 112.955	0.074 970 34
wzz-1	52.0	2 607.138 2	0.074 934 79

Molecular docking

Molecular docking was conducted to analyze the top 5 core targets (AKT1, EGFR, IL6, TNF and MMP9) in PPI and the top 5 key active components (morin, luteolin, jaceosidin, genkwarin and nepetin) analyzed by CytoNCA plugin in the "active component-target-disease-pathway" network. The results are shown in Table 3. The binding energy of the 5 key active components and the 5 core targets was all less than -5 kJ/mol, indicating that the stem activity of *I. szechouensis* had good binding ability with the core targets, thereby exerting anti-prostate cancer effects. Pymol was employed to visualize some docking results, as shown in Fig. 7.

Table 3 Binding energy of key active components and core targets

	AKT1 (1UNQ)	EGFR (1XKK)	IL6 (1ALU)	TNF (2E7A)	MMP9 (1ITV)
wzz-21	-6.78	-6.28	-5.77	-6.58	-7.01
wzz-36	-6.65	-7.26	-5.79	-6.86	-6.69
wzz-7	-6.04	-6.33	-6.00	-7.10	-6.25
wzz-32	-6.10	-6.81	-5.97	-7.53	-6.38
wzz-1	-5.95	-6.48	-5.79	-7.12	-6.45

Conclusions and Discussion

I. szechouensis, also known as Wangsaozi, Wenchongbuchi and Baibingbusheng, is a plant in the genus *Isodon* of the Labiatae family^[17]. *I. szechouensis* contains various effective medicinal ingredients, and has functions such as anti-tumor, antithrombotic, antibacterial, anti-inflammatory, and antioxidant. It is a rare Chinese herbal medicine that can be used for both food and medicine, but its mechanism of action is still unclear. In this study, we preliminarily explored the anti-prostate cancer mechanism and correlation of the active components from stems of *I. szechouensis*, through methods such as LC-MS, network pharmacology and molecular docking.

In this study, the potential mechanism of *I. szechouensis* stems in treating prostate cancer was analyzed by network pharmacology. Sixty three chemical components were quickly analyzed using UPLC-Q-TOF-MS/MS technique, and 62 active components were obtained through Swiss Target Prediction and PharmaMapper, resulting in a total of 684 targets. A total of 1 565 potential targets for prostate cancer were collected in the GeneCards database. A total of 233 common targets were obtained through intersection. Cytoscape software was employed to construct a PPI network, and the top 5 targets were selected as core targets: AKT1, EGFR, IL6, TNF and MMP9. An "active component-target-disease-pathway" network was constructed, and the top 5 active components were obtained as key active components: wzz-21 (morin), wzz-36 (luteolin), wzz-7 (jaceosidin), wzz-32 (genkwarin) and wzz-1 (nepetin). Molecular docking was performed on key active components and core targets, and their binding energy was all less than -5 kJ/mol, indicating good binding activity between the two.

The above 5 key active components all belong to flavonoids. In recent years, the anti-tumor effects of flavonoids have gradually attracted attention, such as the anti-tumor activity of quercetin, which is related to its antioxidant effect, reduction of drug resistance in tumor cells, induction of tumor cell apoptosis, and estrogen like effects^[18-22]. The anti-tumor activity of silymarin is related to its antioxidant effect, inhibition of related-enzyme activity, and induction of cell cycle arrest^[23-24]. AMP, as a kind of flavonoid, has a preventive effect on the growth and metastasis of prostate cancer cells. AMP has stronger inhibitory activity on androgen-sensitive LNCaP proliferation, but weaker inhibitory activity on proliferation of androgen-independent PC-3 human prostate cancer cell line, *in vitro*, mainly by inducing apoptosis related to downregulation of bcl-2. The inhibitory activity of AMP on the proliferation of normal prostate epithelial cells is significantly lower than that on prostate cancer cells. AMP can also inhibit the migration and invasion of PC-3 cells, which is related to the downregulation of CXCR4 expression^[25].

Zhang *et al.*^[26] used immunohistochemical methods to detect and analyze MMP9 in 51 surgical specimens of prostate cancer and 10 surgical specimens of prostate hyperplasia. The results showed that MMP9 expression was positive in 2 out of 10 cases of prostate hyperplasia, which was significantly different from that in prostate

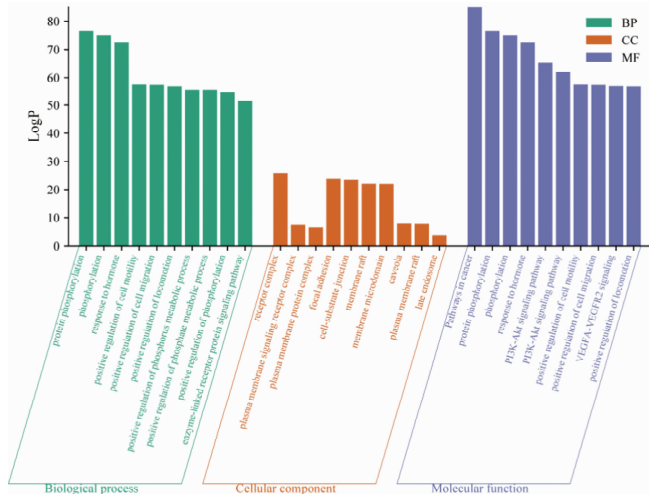


Fig. 4 GO biological function analysis

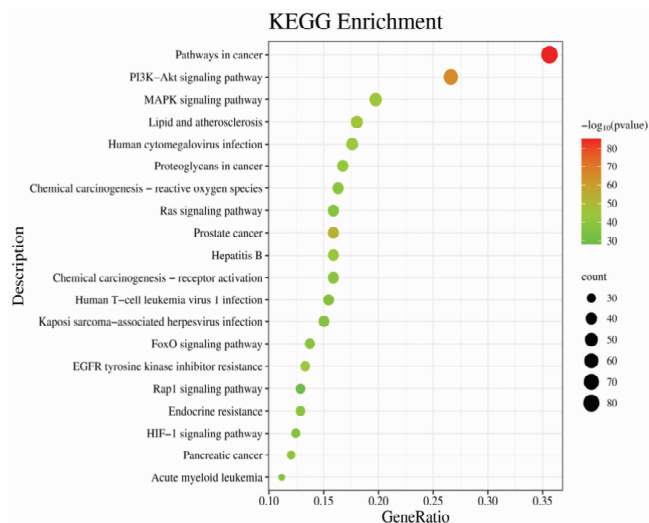


Fig. 5 KEGG pathway enrichment analysis

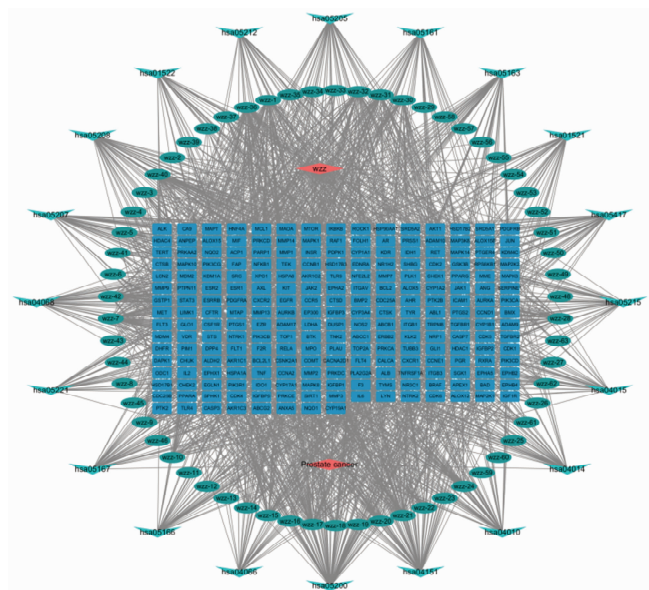


Fig. 6 Active component-target-disease-pathway network

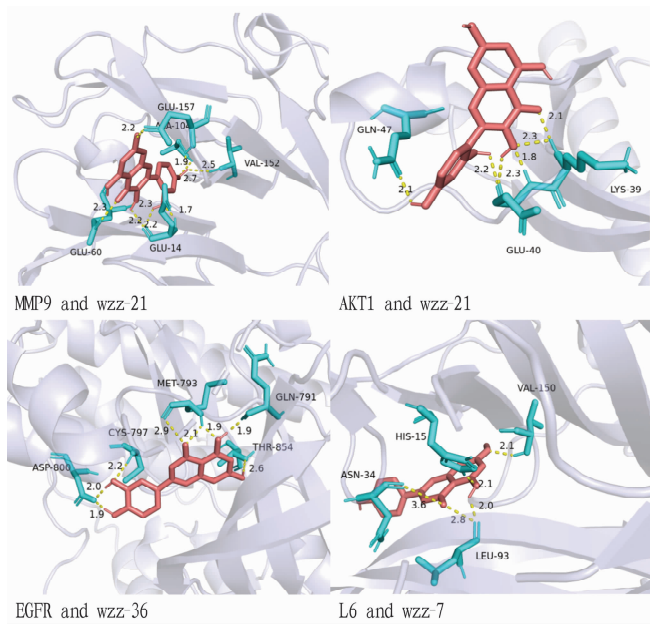


Fig. 7 Molecular docking of some key active components and core target molecules

cancer. Moreover, the expression of MMP9 was closely related to the pathological grading, Gleason score, and metastasis of prostate cancer. MMP9 expression may play an important role in the development and metastasis of prostate cancer. Multiple studies have confirmed that upregulation of MMP9 gene expression is an effective marker of tumor metastasis.

RecQ helicase is an important member of the DNA helicase family, and BLM helicase is one of the five RecQ helicases currently found in the human body. Studies on the occurrence of multiple cancers at the cellular and molecular levels have found that RecQ helicases are involved in the occurrence of leukemia, breast cancer and other cancers, and their functional abnormalities determine the multiple characteristics of related diseases in terms of cytogenetics and clinical manifestations. Multiple studies have shown that RecQ helicases are associated with the development of prostate cancer and play a certain regulatory role [27–29].

In summary, in this study, a preliminary study was conducted on the mechanism of *I. Suzhouensis* stems in treating prostate cancer. Through network pharmacology and molecular docking, it was confirmed that stems of *I. Suzhouensis* may be related to pathways in cancer, prostate cancer, and MicroRNAs in cancer signaling pathways, thereby exerting a therapeutic effect on prostate cancer. Next, we will investigate the relationship between helicases of the RecQ family and MMP9 expression. We aim to determine the correlation between their expression and the severity of prostate cancer lesions, as well as the correlation with MMP9 expression, by detecting the expression of helicase genes in the RecQ family at the protein and transcriptional levels. We will further determine the correlation between helicase genes of the RecQ family and prostate cancer cell metastasis, and explore the feasibility of using RecQ helicase expression as diagnostic markers based on quantitative detection for the development and progression of prostate cancer, hoping to provide a theoretical reference for exploring the treatment of

prostate cancer with stems of *I. szechouensis*.

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