

Antidepressant mechanism of Guipi Decoction revealed by network pharmacology and molecular docking

Fangyuan Wei

Shenyang Pharmaceutical University

Jianxin Song

Shenyang Pharmaceutical University

Xiaoya Pan

Shenyang Pharmaceutical University

Xiangrong Dr. Zhang (✉ xrzhang@syphu.edu.cn)

Shenyang Pharmaceutical University <https://orcid.org/0000-0003-4783-171X>

Research Article

Keywords: Ziziphi Spinosa Semen, Poria cocos, Longan, cytokines, protein kinases, depression

Posted Date: July 27th, 2023

DOI: <https://doi.org/10.21203/rs.3.rs-3012304/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License. [Read Full License](#)

Abstract

Guipi Decoction is a famous Traditional Chinese Medicine formulae, which can be used to treat insomnia and depression. But its specific mechanism is still unclear. In this study, the active ingredients, targets and mechanisms of the Ziziphi Spinosae Semen-Poria cocos-Longan (ZPL) in Guipi Decoction was investigated by network pharmacology analysis and molecular docking. A total of 32 active ingredients, 344 intersection targets and 13 key targets were obtained. The result showed (S)-Coclaurine, coumestrol, n-trans-feruloyltyramine, ellagic acid, ellipticine and tartaric acid were the key compounds for depression. These compositions could reduce inflammatory response, inhibit pro-inflammatory cytokines, reduce neuronal apoptosis, and then modulate depression by the key targets of RAC-alpha serine/threonine-protein kinase (AKT1), tumor necrosis factor (TNF), interleukin (IL6), mitogen activated protein kinase 3 (MAPK3). Molecular docking results showed that the binding energy of n-trans-feruloyltyramine with PPARG was the lowest, -9.513 kcal/mol and the binding energy of (S)-Coclaurine and ESR1 was -9.336. Upregulation of AKT1 gene inhibits apoptosis. Downregulation of TNF- α , MAPK and CTNNB1 genes reduces the expression of inflammatory factors and decreases the inflammatory response, which plays an important role in the treatment of depression. In conclusion, the active component of ZPL binds stably with AKT1, MAPK3, ESR1 and CTNNB1 and controlled the onset of depression by regulation of genes expression.

1 Introduction

Depression is a mood psychiatric disorder with a variety of clinical features including negative attitude, reduced interest, decreased ability to learn and think, sleep disturbances, loss of appetite, and suicidal tendencies. It seriously influences the psychological function and quality of life of patients (Li et al., 2016; Zuo et al., 2022; Wang et al., 2021). In recent years, with the fast-paced life and the high-pressure work, mental stress significantly rises especially for the people suffering from depression. Indeed, depression is a major cause of disability and a contributor to the overall global burden of disease (Yan et al., 2017). As a popular theory, depression is caused by the monoamine-deficiency hypothesis. Therefore, most of the commercially available antidepressants are based on blocking norepinephrine transport. Studies have confirmed the herbal compound therapeutic presents significant effects on depressed mice (Huang et al., 2020). Hence, it is necessary to find more herbal formulations for depressed patients for more treatment options.

“Guipi Decoction” is a famous Traditional Chinese Medicine formulae, recorded in ‘Ji Sheng Fang’, a medical classic written by Yan Yonghe in the Chinese Song Dynasty. It consists of 12 medicinal herbs, including Ginseng, Atractylodis Rhizoma, Poria cocos and Longan which is commonly used to treat anxiety, neurosis, insomnia or anemia (Araki et al., 2021). Therefore, the herbs of Ziziphi Spinosae Semen-Poria cocos-Longan (ZPL) from “Guipi Decoction” were selected to investigate the targets and mechanisms of action on depression.

Pharmacological studies have found that Ziziphi Spinosae Semen contain active ingredients such as jujuboside, magnoflorine, and coclaurine, which contribute to hypnotic and sedative effects (Du et al., 2020). Moreover, Ziziphi Spinosae Semen has commonly used in Chinese doctors’ prescriptions for its unique effects on insomnia, calming nerves and insomnia (Gong et al., 2021). Poria, known as Fu ling in China, is the dried nucleus of the fungus Poria cocos, which is from the ‘Shen Nong Ben Cao Jing’ (Xu et al., 2022). It was found that P. cocos water extract treatment alleviated depression-like symptoms in rats, which is related to modulation of neurotransmitter systems and reduced expression of inflammation in the brain (Huang et al., 2020). Moreover, by the regulation of neurotransmitter levels, promotion of neuronal cell regeneration and regulation of NLRP3 inflammatory vesicle, acidic polysaccharides in Poria cocos can improve depressive behavior in rats (Chen et al., 2021). According to Chinese medicine, Longan is sweet and warm. In the ‘Shen Nong Ben Cao Jing’, it is recorded that it can nourish the heart and spleen, and benefit the Qi and blood. Studies have pointed out that Longan fruit extract can enhance memory, the BDNF upregulation and neurogenesis, and contribute to memory performance (Se et al., 2010). Furthermore, Longan extract also has excellent effects of cardiovascular and anxiety (Okuyama et al., 1999). Since they are successfully used in the formulation of Guipi Decoction, there is a compatibility relationship among them. Based on this, Ziziphi Spinosae Semen-Poria cocos-Longan (ZPL) were selected to investigate their therapeutic effects on depression.

The thinking of “one gene, one target, one disease” has influenced many aspects of drug discovery strategies (Hopkins et al., 2007). However, with increased understanding, it has been discovered that many drugs can act on multiple targets rather than individual targets. Network pharmacology, as a multidisciplinary notion rooted in systems biology and multidisciplinary pharmacology, represents a new network model of “multi-target, multi-component, disease” (Zhang et al., 2013). In addition, molecular docking has been widely applied to recognize the extent of receptor-ligand binding. The advantage of network pharmacology combined with molecular docking is to explain the mechanism of action between the target and the active ingredient (Ding et al., 2021). In this study, we intend to study the antidepressant active components, action targets and action mechanism of ZPL through network pharmacology. Molecular docking technique was used to investigate the interaction between the components and the target.

2 Methods

2.1 Establishment of active ingredient database and target of action of ZPL

The ingredients of Ziziphi Spinosae Semen-Poria cocos-Longan were obtained from the TCMS database (<https://old.tcmsp-e.com/>), and then the ingredients were supplemented with literature and the HERB database (<http://herb.ac.cn/>), ETCM database (www.tcmip.cn/ETCM) and the

Batman-TCM database (<http://bionet.ncpsb.org/batman-tcm/>). Each component was entered into the TCMSP database to determine molecules to obtain Mol ID and the oral bioavailability values, and drug-likeness values. Eligible active ingredients were screened on the value of oral availability (OB) $\geq 30\%$ and the value of drug-likeness (DL) ≥ 0.18 (Ru et al., 2014). In addition, the HERB database was supplemented with ingredients that did not pass the drug-like screening but had been validated by high-throughput experiments with pharmaceutical activity. The SMILE format and 2D structure of the active components were found on the PubChem database (<https://pubchem.ncbi.nlm.nih.gov>), the resulting SMILE format was entered into the Swiss Target Prediction database (<http://www.swisstargetprediction.ch>), the species was selected as "*Homo sapiens*", and then the targets of the components were predicted.

2.2 Establishment of the disease target database

The keyword "Depression" was searched in the GeneCards database (<https://www.genecards.org/>) and DrugBank database (<https://go.drugbank.com>). Species selected "*Homo sapiens*" to obtain disease targets about depression. The resulting targets were searched and corrected on the Uniprot database (<https://www.uniprot.org/>) to identify the correct gene names and delete the non-compliant targets.

2.3 The construction of the protein-protein interaction network

The intersection targets of ZPL and depression were obtained by inputting component and disease targets via the online tool Venny 2.1, respectively (<https://bioinfogp.cnb.csic.es/tools/venny/>). The resulting intersection targets were input to String (<https://string-db.org>) for protein-protein interaction (PPI) analysis. The obtained relationship diagrams were saved in tab-separated value format and imported into Cytoscape 3.7.1 software for analysis. The top thirteen targets were chosen for diagrams by sorting and filtering based on the freedom degree centrality, betweenness centrality and closeness centrality.

2.4 Enrichment pathway analysis

Enrichment analysis of component-disease intersection targets was carried out by the Metascape database (<https://metascape.org>). After input the intersection targets, species were selected as "*Homo sapiens*" for analysis. Select Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analysis, Gene Ontology (GO) enrichment analysis respectively in the analysis options. GO enrichment analysis includes GO CC, GO BP and GO MF. Metascape database is a powerful gene function annotation analysis tool that helps users to apply current popular bioinformatics analysis methods for bulk gene and protein analysis. Gene function annotation (GO and KEGG and the like) can be implemented. In terms of database types, Metascape integrates several authoritative data resources such as GO, KEGG, STRING, UniProt and DrugBank, making it possible to do not only pathway enrichment and biological process annotation, but also gene-related protein network analysis and involved drug analysis (Zhou et al., 2019). The obtained analysis results were organized into tables and then imported into the online graphing tool Bioinformatics (<http://www.bioinformatics.com.cn/>) for graphing.

2.5 Molecular docking screening of targets

Molecular docking was further screened for target proteins using Schrodinger software. At first, the 3D structure of the active ingredient was downloaded from PubChem and optimized by the Chem3D software. The structures of target proteins were found and downloaded from the PDB database (<https://www.rcsb.org>). The protein structure download was based mainly on the selection of molecules less than 2A, newer date, and the species "*Homo sapiens*". Secondly, the target proteins were imported into Schrodinger software, which processed the proteins, removed water, minimized energy, etc. Then, the 3D molecular structures were introduced and the molecules were processed. Finally, using software operations, the ligands were found and lattice points were established for molecular docking. Molecular docking results were used to evaluate the interaction between compounds and target proteins. The more negative the score value, the more stable the binding. A docking score less than -5kcal/mol indicates good binding activity and stable ligand-acceptor binding. The workflow of the current study is shown in Fig. 1.

3 Results

3.1 Screening of the active components of the ZPL

ZPL was composed of three edible traditional Chinese medicines of Ziziphi Spinosae Semen, Poria cocos and Longan. The active components were searched in TCMSP database, ETCM database, Batman-TCM database and HERB database for herbal medicines separately to find literature supplement. Then, the collected data were organized and the obtained component names, MOL ID numbers and structures were compared one by one to remove some inaccurate information. After screening, a database containing 32 active ingredients, 10 from Ziziphi Spinosae Semen, 19 from Poria cocos and 3 from Longan (Table 1).

Table 1
Active compounds of ZPL

Herb name	MOL ID	Molecule name	OB/%	DL
Ziziphi Spinosae Semen	MOL001521	ceanothic acid	33.41	0.77
	MOL001522	(S)-Coclaurine	42.35	0.24
	MOL001525	Daucosterol	36.91	0.75
	MOL001532	phytosterol	36.91	0.75
	MOL001542	swertisin	31.83	0.75
	MOL001546	zizyphusine	41.53	0.55
	MOL000211	Mairin	55.38	0.78
	MOL012976	coumestrol	32.49	0.34
	MOL008647	n-trans-feruloyltyramine	86.71	0.26
	MOL012940	Spiradine A	113.52	0.61
Poria cocos	MOL000273	(2R)-2-[(3S,5R,10S,13R,14R,16R,17R)-3,16-dihydroxy-4,4,10,13,14-pentamethyl-2,3,5,6,12,15,16,17-octahydro-1H-cyclopenta[a]phenanthren-17-yl]-6-methylhept-5-enoic acid	30.93	0.81
	MOL000275	trametenolic acid	38.71	0.8
	MOL000279	Cerevisterol	37.96	0.77
	MOL000280	(2R)-2-[(3S,5R,10S,13R,14R,16R,17R)-3,16-dihydroxy-4,4,10,13,14-pentamethyl-2,3,5,6,12,15,16,17-octahydro-1H-cyclopenta[a]phenanthren-17-yl]-5-isopropyl-hex-5-enoic acid	31.07	0.82
	MOL000282	ergosta-7,22E-dien-3beta-ol	43.51	0.72
	MOL000283	Ergosterol peroxide	40.36	0.81
	MOL000285	(2R)-2-[(5R,10S,13R,14R,16R,17R)-16-hydroxy-3-keto-4,4,10,13,14-pentamethyl-1,2,5,6,12,15,16,17-octahydrocyclopenta[a]phenanthren-17-yl]-5-isopropyl-hex-5-enoic acid	38.26	0.82
	MOL000287	3beta-Hydroxy-24-methylene-8-lanostene-21-oic acid	38.7	0.81
	MOL000289	pachymic acid	33.63	0.81
	MOL000291	Poricoic acid B	30.52	0.75
	MOL000292	poricoic acid C	38.15	0.75
	MOL000296	hederagenin	36.91	0.75
	MOL000300	dehydroeburicoic acid	44.17	0.83
	MOL009149	Cheilanthifoline	46.68	0.72
	MOL004492	Chrysanthemaxanthin	38.72	0.58
	MOL001002	ellagic acid	43.06	0.43
	MOL009135	ellipticine	30.82	0.28
	MOL005890	pachypodol	75.06	0.4
	MOL009136	Peraksine	82.58	0.78
	Longan	MOL001987	β -sitosterol	33.94
MOL001886		Tartaric Acid	66.38	0.02
MOL001788		Adenine	62.81	0.03

3.2 Identification of component targets and disease targets

By the database screening, a total of 747 component targets were obtained from ZPL, including 523 targets of Ziziphi Spinosae Semen, 532 targets of Poria cocos, and 174 targets of Longan. A total of 2409 targets depression were obtained by combining GeneCards and DrugBank databases after removing duplicate genes. The Venn diagram of drug and disease targets (Fig. 2), were a total of 344 drug-disease intersecting

targets. Figure 3 showed the relationship between ZPL, active ingredient, intersecting targets and depression. From the Fig. 3, it can be seen how ZPL can treat depression through the interaction between the ingredients and the targets.

3.3 PPI network

The intersecting targets were imported into the String database to get the relationship graph between the targets. Figure 4a showed the relationship map after removing the free target genes. There were 344 intersecting genes and 57 free genes. The topological index node degree of the remaining 287 genes was calculated, which represented the degree of cross-linking between individual genes and other genes, and the strength of their roles. The mean value of the topological index node degree of 287 genes was 8.43, and the genes greater than the mean value were selected to generate 95 core targets with relatively important networks of interactions with other genes, as shown in Fig. 4b (Wu et al., 2021). Cytoscape software was used to draw and analyze the PPI network, and it was found that there were strong interactions among the targets (Fig. 4c).

3.4 GO pathway enrichment analysis

The results of pathway and enrichment showed that 344 common targets had the same biological function. This work analyzed from three aspects: biological process (BP), molecular function (MF) and cellular component (CC). Figure 5 represented the top ten enrichment result plots of BP, CC and MF. The graph showed that the focus of the BP analysis was mainly performed with cellular response to nitrogen compound, cellular response to organonitrogen compound and synaptic signaling. The CC terms were associated with dendrite, dendritic tree, postsynapse, synaptic membrane and cell body. While MF terms involved phosphotransferase activity, alcohol group as acceptor, kinase activity and protein kinase activity. The *p*-value determined the outcome of the correlation test (Jafari et al, 2019), and the pathway data values according to log*P* values were ranked (Fig. 5).

3.5 KEGG pathway enrichment analysis

The enrichment analysis of common targets by KEGG yielded a total of 219 signaling pathways. The results were sorted according to the enrichment scores, and the top ten pathways were obtained (Fig. 6). From the graph, it can be seen that the pathways associated with depression include Neuroactive ligand-receptor interaction, Pathways in cancer, Calcium signaling pathway, cAMP signaling pathway and Pathways of neurodegeneration-multiple diseases. In general, KEGG pathway enrichment analysis can provide further insight into the potential pathways of ZPL for depression (Gong et al., 2021).

3.6 Molecular docking results

Based on the PPI network analysis (Fig. 4), these 13 targets were selected as the main target proteins and docked with the active ingredient molecules. The specific results of molecular docking were presented in Table 2. When the molecular docking binding energy is less than -5 kcal/mol, it indicates the ligand molecule binds well to the receptor protein (Li et al., 2022). Therefore, the stability of the conformation is judged by the magnitude of the binding force values. A total of 22 molecules were able to the core targets and had good binding stability. From the table, we could see that *n*-trans-feruloyltyramine had the lowest binding energy with PPARG, -9.513 kcal/mol. Followed by (S)-Coclaurine and ESR1 with the binding energy of -9.336. *N*-trans-feruloyltyramine formed hydrogen bonds with GLN283 and hydrophobic action with PHE282 (Fig. 7a). (S)-Coclaurine formed hydrogen bonds with PRO82, MET105 and ASN135 (Fig. 7b). Enoic acid formed hydrogen bonds with GLN79, THR82 and ASN204 (Fig. 7c). Poricoic acid B formed hydrogen bonds with ARG288 and TYR473 (Fig. 7d). In conclusion, a total of 7 molecules were successfully docked with 13 core targets, and AKT1, MAPK3, ESR1 and CTNNB1 targets bounded well to each molecule (Table 2).

Table 2
The binding energy of active components with target genes

Mol ID	Binding Energy (kcal/mol)												
	AKT1	ALB	TNF	IL6	SRC	TP53	EGFR	MAPK3	CTNNB1	JUN	ESR1	IL1B	PPARG
MOL001521	-4.209	/	/	/	-2.123	/	/	/	-3.853	-2.623	/	/	/
MOL001522	-7.934	-5.766	-3.664	-5.401	-3.217	-4.768	-7.233	-7.001	-6.468	-6.075	-9.336	-4.783	-7.662
MOL001525	-8.231	/	/	/	/	/	-6.429	-4.874	-3.808	/	-3.004	/	/
MOL001532	-6.331	/	-1.598	-5.143	-1.85	-3.888	-5.072	-4.44	-3.58	-3.303	-5.336	-2.6	/
MOL001542	-7.462	-4.424	/	-3.225	-3.866	/	-6.009	-5.291	-5.416	-5.428	-6.091	-3.813	-7.516
MOL001546	-6.533	-3.175	-3.299	/	-3.384	-3.172	-5.234	-6.473	-6.206	-5.016	-5.007	-2.405	-7.524
MOL000211	-5.192	-3.839	/	/	/	/	/	-3.608	-4.494	-2.647	/	/	/
MOL012976	-7.64	-4.459	-5.163	-6.509	-4.357	-5.005	-6.674	-7.909	-6.032	-6.31	-8.696	-4.334	-7.757
MOL008647	-6.432	-4.799	-3.247	-6.809	-4.55	-3.912	-6.778	-6.177	-6.682	-6.992	-7.129	-3.828	-9.513
MOL000279	-6.944	-4.752	-2.582	-3.756	-1.207	/	-5.406	-5.579	-4.288	-3.211	-3.843	-2.763	-8.245
MOL000282	-6.493	-4.008	-2.393	-4.016	-1.377	/	-5.549	-4.678	-5.283	-4.101	-5.06	-2.667	-8.092
MOL000283	-5.926	-2.835	-3.338	-4.756	/	/	-5.496	-5.718	-4.173	-4.061	-3.389	-2.574	-7.924
MOL000285	-8.658	-1.078	-2.237	-2.53	-0.414	/	-5.463	-4.95	-4.482	-2.26	-4.164	-1.755	/
MOL000291	-6.515	-4.226	-2.125	-3.427	/	/	-4.58	-4.981	-3.279	-3.851	-1.82	-2.252	-8.938
MOL000292	-6.088	-2.549	-2.336	-4.151	-3.442	/	/	-3.409	-1.453	-3.848	-3.628	-2.419	-6.462
MOL009149	-7.500	-3.693	-3.103	-4.509	-2.842	/	-6.162	-5.414	-6.636	-7.996	-7.684	-3.195	-8.329
MOL004492	-5.823	/	/	/	/	/	-5.549	/	-4.627	/	/	/	/
MOL001002	-8.502	-4.809	-3.994	-5.643	-4.015	-0.945	-6.021	-7.973	-5.305	-5.711	-5.85	-4.306	-7.021
MOL009135	-7.533	-3.641	-3.984	-5.312	-3.569	-4.408	-6.051	-7.091	-6.013	-4.598	-8.598	-4.041	-7.079
MOL005890	-7.294	-3.859	-4.149	-4.792	-3.047	/	-6.763	-7.287	-6.181	-5.039	-8.263	-4.219	-7.901
MOL001886	-4.577	-4.631	-3.746	-3.619	-6.842	-3.118	-4.895	-3.574	-4.048	-3.669	-4.039	-3.879	-4.999
MOL001788	-5.539	-4.574	-4.879	-5.466	-4.186	-5.122	-5.578	-6.264	-5.968	-5.958	-8.086	-4.676	-6.812

4 Discussion

Depression is a major health problem affecting over 20% of the global population (Geoffroy et al., 2014; Geoffroy et al., 2017; Stubbs et al., 2016; Guichard et al., 2020). "Guipi Decoction" is a well-known formulation which has excellent effect on treating poor memory, fatigue and palpitations. Ziziphi Spinosae Semen, Poria cocos and Longan were selected from the Guipi Decoction to explore the treatment mechanism and treatment effect of depression. A compound formulae composed including Ziziphi Spinosae Semen, Poria cocos. It can nourish the heart and tranquilize the mind (PRC PCO,2020). Longan can nourish blood and essence, and it was selected to investigate its effect on depression and its mechanism of action. In the study, the active constituents and targets of ZPL were identified by network pharmacological method, and correlated them with depression-related targets to construct a network relationship map between drug-ingredient-target-disease. The protein interaction network of the main target was obtained by analyzing 344 intersecting targets. Then, the molecular mechanism of antidepressant effect of ZPL was analyzed by enrichment analysis of GO and KEGG pathways. Finally, the binding stability of the core targets to the active ingredients was determined by molecular docking.

The PPI network diagram was analyzed and the core targets were determined based on three topological parameters, using Cytoscape software (Fig. 4c). These 13 core targets may have a strong connection with depression treatment. Through the metaspape platform, the path analysis of multiple targets was carried out to find the relationship between key targets and paths. Therefore, the relevance of ZPL to depression was closely related to the signaling pathways involved in the core targets. The biological functions of ZPL for depression were mainly related to the ligand receptor action of neural activity, synaptic signaling, and inflammatory response. Studies have indicated that in patients with depression, IL-1 β , IL-6, TNF- α and other pro-inflammatory cytokines increased significantly (Dannehl et al., 2014; Zhang et al., 2018). In addition, the injection of TNF- α could induce depression in mice (Katz et al., 1981), thus confirming the involvement of inflammatory cytokines in the depressed state (Vishnu et

al., 2021). Therefore, the decrease of inflammatory response also plays an important role in the treatment of depression. In addition, a possible explanation for the pro-inflammatory theory has been proposed for the pathogenesis of various mood disorders (e.g., depression) (Milenkovic et al., 2019; Miller et al., 2016).

Through the study of GO functional and BP enrichment analysis, the gene expression information can be obtained and the protein transport pattern in the biological pathway can be elucidated (Gong et al., 2021). ZPL-depression related pathways were analyzed in detail by GO and KEGG enrichment analysis. For example, PI3K-AKT signaling pathway and MAPK signaling pathway. The PI3K-AKT pathway plays an important role in the regulation of cell proliferation, survival, differentiation and protein translation (Xing et al., 2020). PI3K-AKT plays an important role in the formation and development of depression (Gong et al., 2021). The MAPK pathway is the key pathway of inflammatory cytokine production and can regulate the expression of inflammatory genes at post-transcription level (Zhou et al., 2017). Some studies have shown that neuronal apoptosis plays an important role in the development of depression (Shen et al., 2018). In the pathway enrichment analysis, there were several pathways about neural cell bodies, synaptic conduction, with high enrichment scores (Fig. 5). Hence, ZPL had an inhibitory effect on depression by acting on neurons in multiple ways. As a serine/threonine kinase, AKT1 plays an important role in intracellular signaling pathways. AKT1 is an important intermediate molecule in the PI3K/AKT signaling pathway, which takes part in the metabolism of a variety of intracellular substances and plays an important role in the regulation of a variety of cellular responses, closely related to oxidative stress, apoptosis and protein synthesis (Abeyrathna et al., 2015; Larson-Casey et al., 2016; Singh et al., 2017; Saatloo et al., 2019). A novel mechanism for activating AKT1 phosphorylation to attenuate apoptosis, AKT1 inhibits apoptosis to repair cerebrospinal fluid (Li et al., 2022). Excess TNF can lead to a variety of inflammatory diseases (Atrekhany et al., 2020). TNF- α is an essential inflammatory cytokine that promotes balance in the body by regulating inflammation, cell proliferation, differentiation, survival and death (Chen and Goeddel, 2002; Walczak, 2011). The cytokine IL-6 is an important neuroimmune factor produced mainly by astrocytes in the central nervous system (Lin et al., 2017). Some studies have suggested the use of IL-6 as a confirmatory marker for major depression in the clinical (Khandaker et al., 2014, Roohi et al., 2021). SRC family kinases can be expressed in many cell types, but are particularly abundant in neural tissue. Primarily, they can regulate the ability of various neuronal channel activities (Wang et al., 2004). In the mouse experiments of Komarova et al. TP53 was found to be strongly expressed during mouse brain development and involved in neurodevelopment (Komarova et al., 1997). Patients with stage IV non-small cell lung cancer carrying EGFR mutations have been shown to exhibit elevated pro-inflammatory marker TNF- α , and present the less depression level than those without EGFR mutations (Jacobs et al., 2017). It was shown by immunoblot analysis that MAPK3 may be an important junction and Camk2b-Mapk3 may serve as a new potential pathway affecting synaptic protein expression in hippocampal neurons (Yang et al., 2022). The CTNNB1 gene encodes the intracellular scaffolding protein β -linked protein. The protein is a key regulator of inflammatory cytokine expression and controls the infiltration of immune cells (Kato, 2018; Li et al., 2019; Sumida et al., 2018; Xia et al., 2021). As a ligand-activated transcription factor, ESR1 influences hundreds of genes, including the regulation of synthesis and metabolism of various neurotransmitters in the brain, and then affects depression (Różycka et al., 2016). Generally, the key targets related to the development of depression make affects through inflammatory responses, while some ones work through the regulation of synaptic expression in neurons. Furthermore, the specific experiments were needed to confirm the effectiveness of these targets in the future.

5 Conclusions

In this study, both 32 active ingredients of ZPL and 13 key targets related to depression were investigated based on network pharmacology and molecular docking research methods. GO and KEGG pathways were analyzed to verify the binding stability of important active ingredients and targets. In conclusion, the key targets can reduce the inflammatory response and inhibit the elevation of pro-inflammatory cytokines such as IL 6 and TNF- α to alleviate depression. It also can have the modulatory effects on neurons, reduce neuronal apoptosis and regulate the expression of synaptic proteins. Network pharmacology combined with molecular docking has a broad application prospect, and through this approach can provide a theoretical basis for the treatment of depression by ZPL from Guipi Decoction.

Declarations

Funding: This work was funded by the People's Livelihood Plan Project of Department of Science and Technology of Liaoning Province (2021JH2/10300069) and the Department of Education of Liaoning Province (LJKZ0918). Shenyang Pharmaceutical University Scientific Research Foundation (GGJJ2015102).

Competing Interests: The authors declared that no potential conflicts of interest with respect to the research, author-ship, and publication of this article.

Author Contributions: **Fangyuan Wei:** Data curation; Formal analysis; Investigation; Methodology; Validation; Writing-original draft. **Jianxin Song:** Writing-review & editing. **Xiaoya Pan:** Methodology; Visualization. **Xiangrong Zhang:** Project administration; Supervision.

Data availability: The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

Code availability: Not applicable

Ethics approval: Not applicable

Consent to participate: Not applicable

Consent for publication: All the authors listed have approved the manuscript that is enclosed.

References

1. Abeyrathna P, Su Y (2015) The critical role of Akt in cardiovascular function. *Vascul Pharmacol* 74:38–48
2. Araki R, Tachioka H, Kita A, Fujiwara H, Toume K, Matsumoto K, Yabe T (2021) Kihito prevents corticosterone-induced brain dysfunctions in mice. *J Tradit Compl Med* 11(6):513–519
3. Atrekhany KN, Gogoleva VS, Drutskaya MS, Nedospasov SA (2020) Distinct modes of TNF signaling through its two receptors in health and disease. *J Leukoc Biol* 107(6):893–905
4. Chen E, Goeddel DV (2002) TNF-R1 signaling: a beautiful pathway. *Science* 296(5573):1634–1635
5. Chen K, Chen S, Ren J, Lin S, Xiao M, Cheng L, Ye X (2021) Antidepressant effect of acidic polysaccharides from *Poria* and their regulation of neurotransmitters and NLRP3 pathway. *Zhongguo Zhong yao za zhi* 46(19):5088–5095
6. Dannehl K, Rief W, Schwarz MJ, Hennings A, Riemer S, Selberdinger V, Stapf T, Euteneuer F (2014) The predictive value of somatic and cognitive depressive symptoms for cytokine changes in patients with major depression. *Neuropsychiatric Dis Treat* 10:1191–1197
7. Ding S, Wang W, Song X, Ma H (2021) Based on network pharmacology and molecular docking to explore the underlying mechanism of Huangqi Gegen decoction for treating diabetic nephropathy. *Evidence-Based Complementary and Alternative Medicine*, 2021, 9928282
8. Du C, Yan Y, Shen C, Cui X, Pei X, Qin X (2020) Comparative pharmacokinetics of six major compounds in normal and insomnia rats after oral administration of *Ziziphi Spinosae Semen* aqueous extract. *J Pharm Anal* 10(4):385–395
9. Geoffroy PA, Boudebessé C, Bellivier F, Lajnef M, Henry C, Leboyer M, Scott J, Etain B (2014) Sleep in remitted bipolar disorder: a naturalistic case-control study using actigraphy. *J Affect Disord* 158:1–7
10. Geoffroy PA, Micoulaud Franchi JA, Lopez R, Poirot I, Brion A, Royant-Parola S, Etain B (2017) [How to characterize and treat sleep complaints in bipolar disorders?]. *Encephale* 43(4):363–373
11. Gong F, Xie L, Chen B, Yang S, Wu W, Li J, Gong Q, Zhong L, Wu J (2021) Exploration of *Ziziphi Spinosae Semen* in Treating Insomnia Based on Network Pharmacology Strategy. Hindawi Limited
12. Guichard K, Geoffroy PA, Taillard J, Micoulaud-Franchi JA, Royant-Parola S, Poirot I, Brion A, d'Ortho M-P, Gagnadoux F, Schroder C, Philip P, Bioulac S (2020) Stratégies de gestion de l'impact du confinement sur le sommeil: une synthèse d'experts. *Med Sommeil* 17(2):108–112
13. Hopkins AL (2007) Network pharmacology. *Nat Biotechnol* 25(10):1110–1111
14. Huang Y, Hsu N, Lu K, Lin Y, Lin S, Lu Y, Liu W, Chen M, Sheen L (2020) *Poria cocos* water extract ameliorates the behavioral deficits induced by unpredictable chronic mild stress in rats by down-regulating inflammation. *J Ethnopharmacol* 258:112566
15. Jacobs JM, Traeger L, Eusebio J, Simon NM, Sequist LV, Greer JA, Temel JS, Pirl WF (2017) Depression, inflammation, and epidermal growth factor receptor (EGFR) status in metastatic non-small cell lung cancer: A pilot study. *J Psychosom Res* 99:28–33
16. Jafari M, Ansari-Pour N (2019) Why, when and how to adjust your P values? *Cell J* 20(4):604–607
17. Kato M (2018) Multilayered prevention and treatment of chronic inflammation, organ fibrosis and cancer associated with canonical WNT/betacatenin signaling activation (Review). *Int J Mol Med* 42(2):713–725
18. Katz RJ, Roth KA, Carroll BJ (1981) Acute and chronic stress effects on open field activity in the rat, implications for a model of depression. *Neurosci Biobehavioral Reviews* 5(2):247–251
19. Khandaker GM, Pearson RM, Zammit S, Lewis G, Jones PB (2014) Association of serum interleukin 6 and C-reactive protein in childhood with depression and psychosis in young adult life: a population-based longitudinal study. *JAMA Psychiatry* 71(10):1121
20. Komarova EA, Chernov MV, Franks R, Wang K, Armin G, Zelnick CR, Chin DM, Bacus SS, Stark GR, Gudkov AV (1997) Transgenic mice with p53-responsive lacZ: p53 activity varies dramatically during normal development and determines radiation and drug sensitivity in vivo. *EMBO J* 16:1391–1400
21. Larson-Casey JL, Deshane JS, Ryan AJ, Thannickal VJ, BrentCarter A (2016) Macrophage Akt1 kinase-mediated mitophagy modulates apoptosis resistance and pulmonary fibrosis. *Immunity* 44(3):582–596
22. Li E, Yan R, Yan K, Huang R, Zhang R, Wen Y, Li S, Li P, Ma Q, Liao B (2022) Erxian decoction inhibits apoptosis by activating Akt1 and repairs spinal cord injury in rats. *Heliyon* 8(11):11279
23. Li H, Linjuan L, Wang Y (2016) G-CSF improves CUMS-induced depressive behaviors through downregulating Ras/ERK/MAPK signaling pathway. *Biochem Biophys Res Commun* 479:827–832

24. Li J, Wang Y, Wang R, Wu M, Shan J, Zhang Y, Xu H (2022) Study on the molecular mechanisms of tetrandrine against pulmonary fibrosis based on network pharmacology, molecular docking and experimental verification. *Heliyon* 8(8):10201
25. Li X, Xiang Y, Li F, Yin C, Li B, Ke X (2019) WNT/ β -Catenin signaling pathway regulating T cell-inflammation in the tumor microenvironment. *Front Immunol* 10:2293
26. Lin Y, Liu L, Jiang H, Zhou J, Tang Y (2017) Inhibition of interleukin-6 function attenuates the central sensitization and pain behavior induced by osteoarthritis. *Eur J Pharmacol* 811:260–267
27. Milenkovic VM, Stanton EH, Nothdurfter C, Rupprecht R, Wetzel CH (2019) The Role of Chemokines in the Pathophysiology of Major Depressive Disorder. *Int J Mol Sci*, 20 (9)
28. Miller AH, Raison CL (2016) The role of inflammation in depression: from evolutionary imperative to modern treatment target. *Nat Rev Immunol* 16(1):22–34
29. Okuyama E, Ebihara H, Takeuchi H, Yamazaki M (1999) Adenosine, theanxiolytic-like principle of the Arillus of *Euphoria longana*. *Planta Med* 65:115–119
30. PRC PCO (2020) Pharmacopoeia of the People's Republic of China. China Medical Science Press, Beijing, China
31. Roohi E, Jaafari N, Hashemian F (2021) On inflammatory hypothesis of depression: what is the role of IL-6 in the middle of the chaos? *J Neuroinflamm* 18:45
32. Różycka A, Słopeń R, Słopeń A, Dorszewska J, Seremak-Mrozikiewicz A, Lianeri M, Maciukiewicz M, Warenik-Szymankiewicz A, Grzelak T, Kurzawińska G, Drews K, Klejewski A, Jagodziński PP (2016) The MAOA, COMT, MTHFR and ESR1 gene polymorphisms are associated with the risk of depression in menopausal women. *Maturitas* 84:42–54
33. Ru J, Li P, Wang J, Zhou W, Li B, Huang C, Li P, Guo Z, Tao W, Yang Y, Xu X, Li Y, Wang Y, Yang L (2014) TCMSp: a database of systems pharmacology for drug discovery from herbal medicines. *J Cheminform*, 6 (13).
34. Saatloo MV, Aghbali AA, Koohsoltani M, Khosroushahi AY (2019) Akt1 and Jak1 siRNA based silencing effects on the proliferation and apoptosis in head and neck squamous cell carcinoma. *Gene* 714:143997
35. Se J, Dong H, Dong H, Seungjoo L, Byung H, Won Y, Kyung T, Jae H, Jong H (2010) The memory-enhancing effects of *Euphoria longan* fruit extract in mice. *J Ethnopharmacol* 128:160–165
36. Singh AK, Kashyap MP, Tripathi VK, Singh S, Garg G, Rizvi SI (2017) Neuroprotection through rapamycin-induced activation of autophagy and PI3K/Akt1/mTOR/CREB signaling against amyloid-beta-induced oxidative stress, synaptic/ neurotransmission dysfunction, and neurodegeneration in adult rats. *Mol Neurobiol* 54(8):5815–5828
37. Stubbs B, Vancampfort D, Veronese N, Solmi M, Gaughran F, Manu P, Rosenbaum S, Hert MD, Fornaro M (2016) The prevalence and predictors of obstructive sleep apnea in major depressive disorder, bipolar disorder and schizophrenia: A systematic review and meta-analysis. *J Affect Disord* 197:259–267
38. Sumida T, Lincoln MR, Ukeje CM, Rodriguez DM, Akazawa H, Noda T, Naito AT, Komuro I, Dominguez-Villar M, Hafler DA (2018) Activated beta-catenin in Foxp3(+) regulatory T cells links inflammatory environments to autoimmunity. *Nat Immunol* 19(12):1391–1402
39. Vishnu N, Thakare SH, Lakade MP, Mahajan YP, Kulkarni VD, Dhakane, Minal T, Harde, Bhoomika M, Patel (2021) Protocatechuic acid attenuates chronic unpredictable mild stress induced-behavioral and biochemical alterations in mice. *Eur J Pharmacol* 898:173992
40. Walczak H (2011) TNF and ubiquitin at the crossroads of gene activation, cell death, inflammation, and cancer. *Immunol Rev* 244(1):9–28
41. Wang H, Liu H, Wang L, Min L, Chen B, Li H (2021) Uncovering the active components, prospective targets, and molecular mechanism of Baihe Zhimu decoction for treating depression using network pharmacology-based analysis. *J Ethnopharmacol* 281:114586
42. Wang K, Hackett JT, Cox ME, Hoek MV, Lindstrom JM, Parsons SJ (2004) Regulation of the Neuronal Nicotinic Acetylcholine Receptor by Src Family Tyrosine Kinases. *J Biol Chem* 279(10):8779–8786
43. Wu J, Ye X, Yang S, Yu H, Zhong L, Gong Q (2021) Systems pharmacology study of the anti-liver injury mechanism of *Citri reticulatae* pericarpium. *Front Pharmacol* 12:618846
44. Xia CY, Xu JK, Li L, Lian WW, Yan Y, Ma BZ, He J, Zhang WK (2021) Identifying the mechanism underlying antidepressant-like effects of loganin by network pharmacology in combination with experimental validation. *J Ethnopharmacol* 281:114526
45. Xing N, Wang Y, Wang W, Zhong R, Xia T, Ding Z, Yang Y, Zhong Y, Shu Z (2020) Cardioprotective effect exerted by Timosaponin B through the regulation of endoplasmic stress-induced apoptosis. *Phytomedicine* 78:153288
46. Xu T, Zhang H, Wang S, Xiang Z, Kong H, Xue Q, He M, Yu X, Li Y, Sun D, Gao P, Cong Z (2022) A review on the advances in the extraction methods and structure elucidation of *Poria cocos* polysaccharide and its pharmacological activities and drug carrier applications. *Int J Biol Macromol* 217:536–551
47. Yan T, He B, Wan S, Xu M, Yang H, Feng X, Bi K, Jia Y (2017) Antidepressant-like effects and cognitive enhancement of *Schisandra chinensis* in chronic unpredictable mild stress mice and its related mechanism. *Sci Rep* 7(1):6903–6918. 10.1038/s41598-017-07407-1
48. Yang C, Zhang M, Li S, Yi F, Huang H, Xie H, Liu H, Huang R, Zhou J (2022) Effects of Camk2b Overexpression and Underexpression on the Proteome of Rat Hippocampal Neurons. *Neuroscience* 503:58–68

49. Zhang G, Li Q, Chen Q, Su S (2013) Network Pharmacology: A New Approach for Chinese Herbal Medicine Research. Evidence-Based Complementary and Alternative Medicine, pp 1–9
50. Zhang L, Previn R, Lu L, Liao R-F, Jin Y, Wang R-K (2018) Crocin, a natural product attenuates lipopolysaccharide-induced anxiety and depressive-like behaviors through suppressing NF-kB and NLRP3 signaling pathway. Brain Res Bull 142:352–359
51. Zhou DX, Zhao Y, Baker JA, Gu Q, Hamre KM, Yue J, Jones BC, Cook MN, Lu L (2017) The effect of alcohol on the differential expression of cluster of differentiation 14 gene, associated pathways, and genetic network. PLoS ONE 12:0178689
52. Zhou Y, Zhou B, Pache L, Chang M, Khodabakhshi A, Tanaseichuk O, Benner C, Chanda S (2019) Within any publication that makes use of analyses inspired by Metascape. Nat Commun 10(1):1523
53. Zuo C, Cao H, Song Y, Gu Z, Huang Y, Yang Y, Miao J, Zhu L, Chen J, Jiang Y, Wang F (2022) Nrf2: An all-rounder in depression. Redox Biol 58:102522

Figures

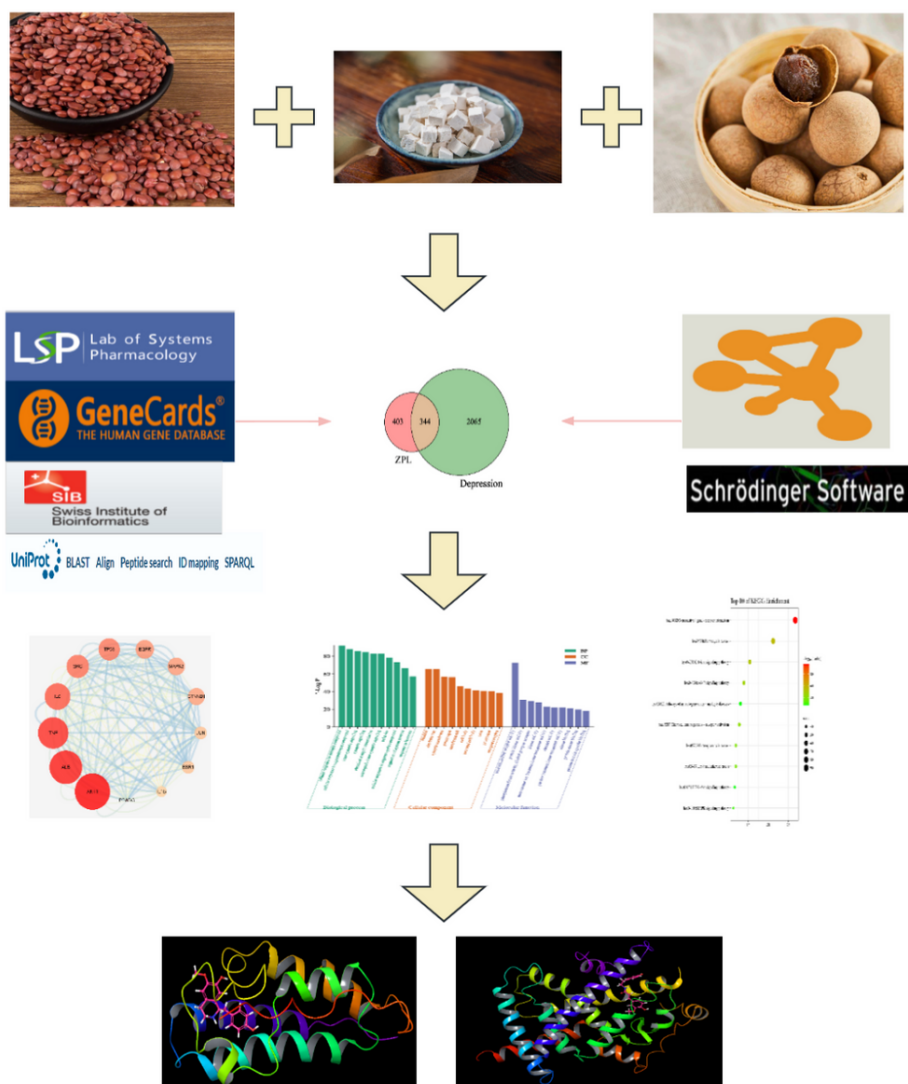


Figure 1

Flow chart to study the mechanism of action of ZPL on depression.

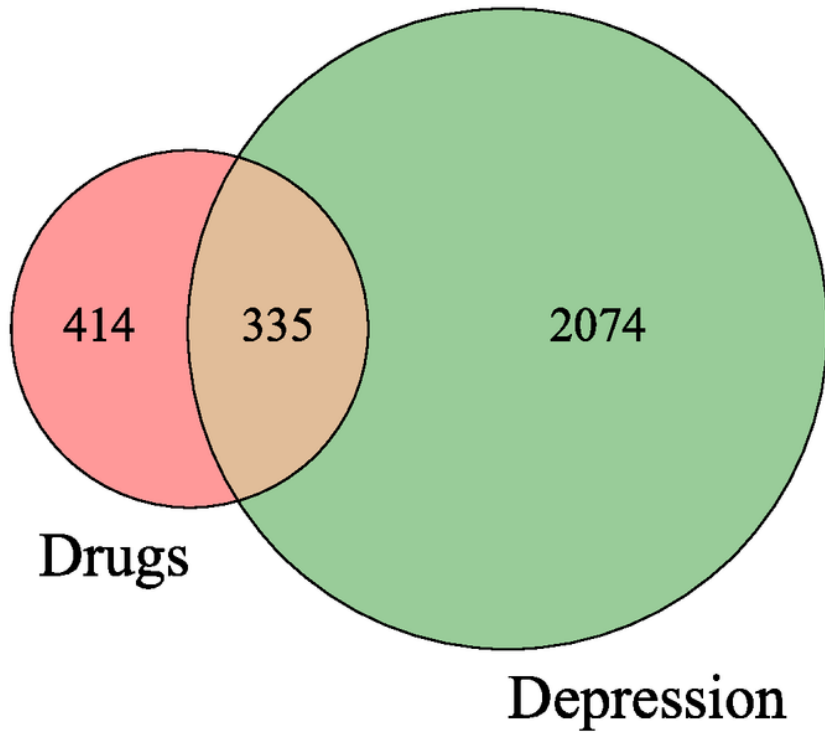


Figure 2
 Venn diagram of related targets of ZPL and depression.

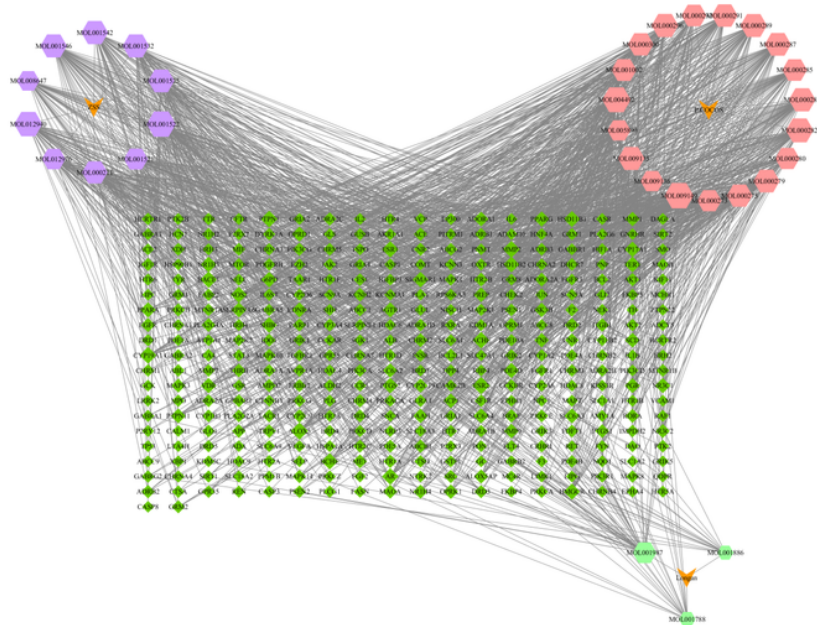


Figure 3

Drug-component-gene-disease network diagram.

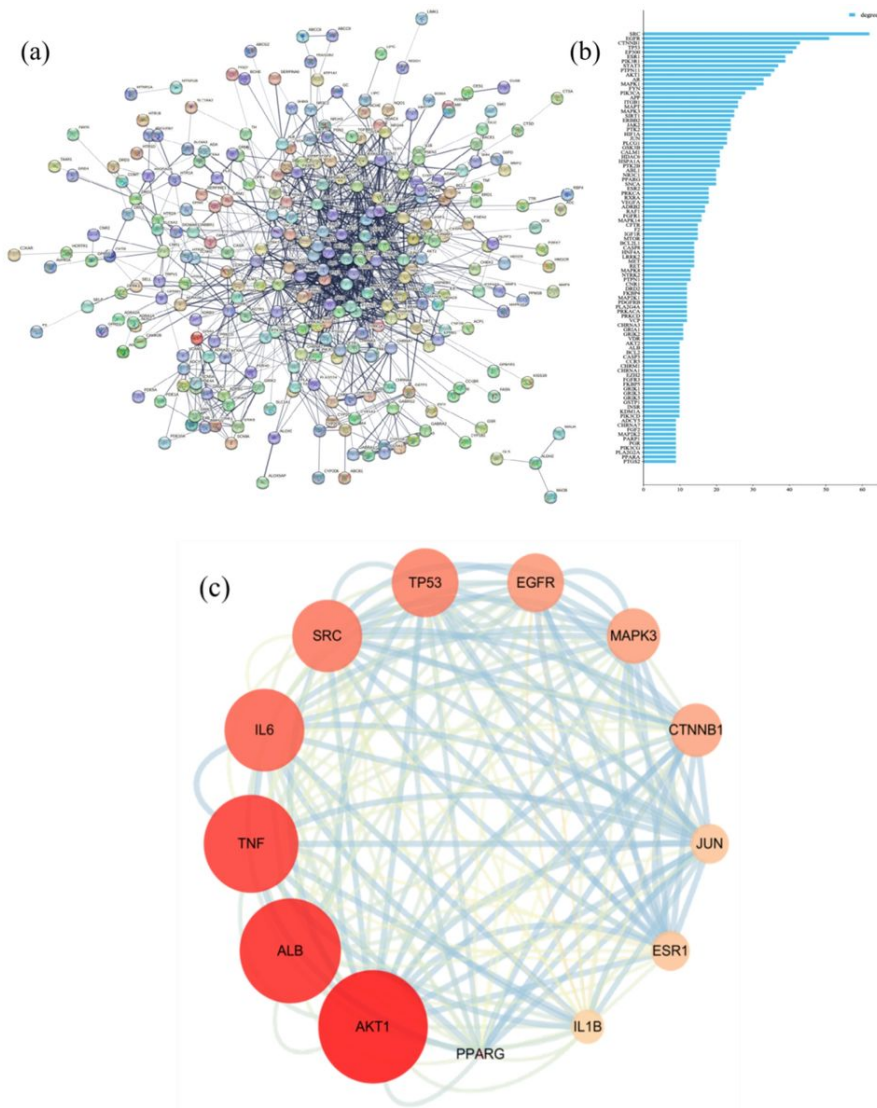


Figure 4

(a) The PPI network diagram from STRING database. (b) Core gene degree barplot diagram. (c) PPI network arranged according to degree value (The node size and the shade color represent the node degree of the target protein).

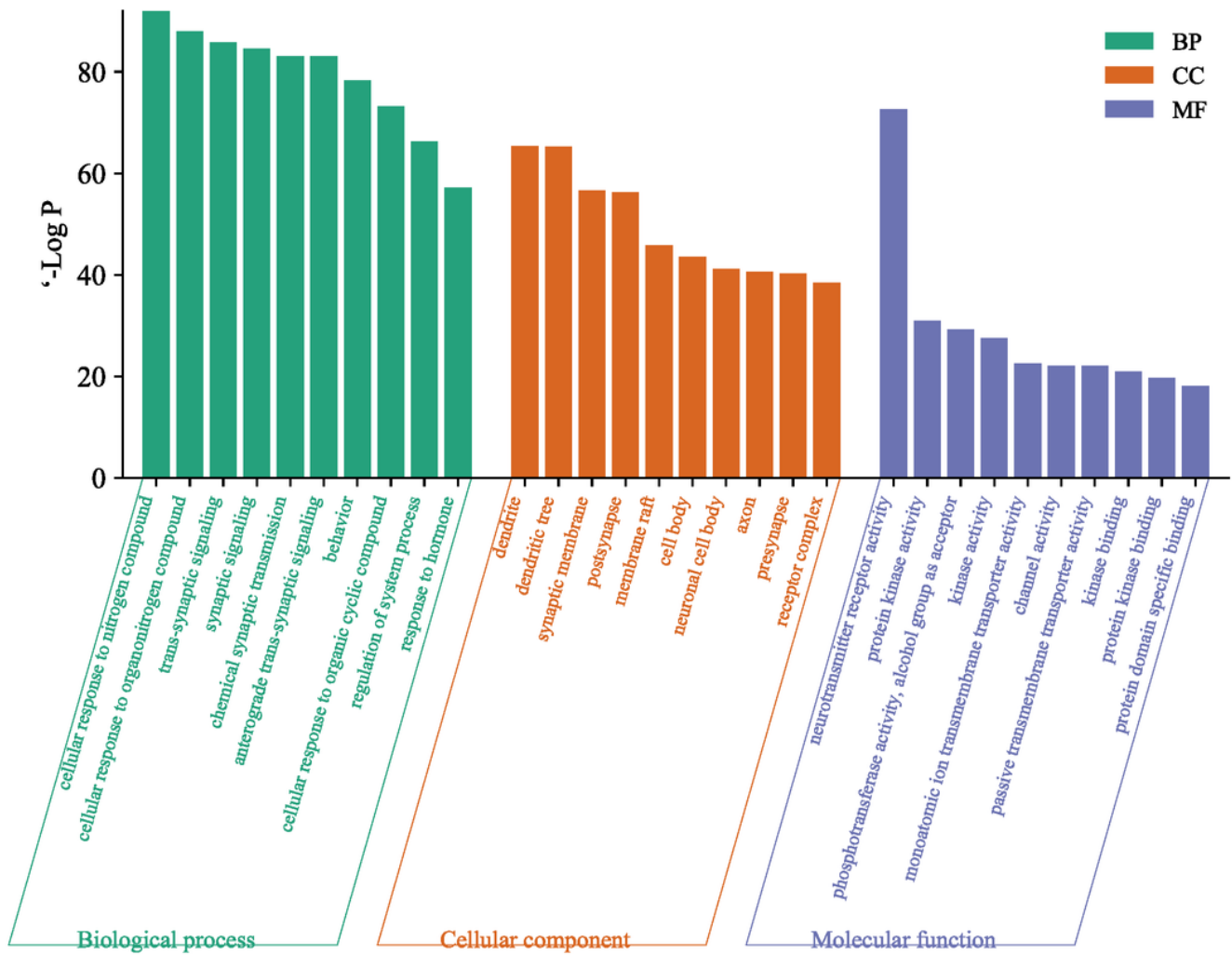


Figure 5

GO BP, GO CC, GO MF pathways p value analysis.

Top 10 of KEGG Enrichment

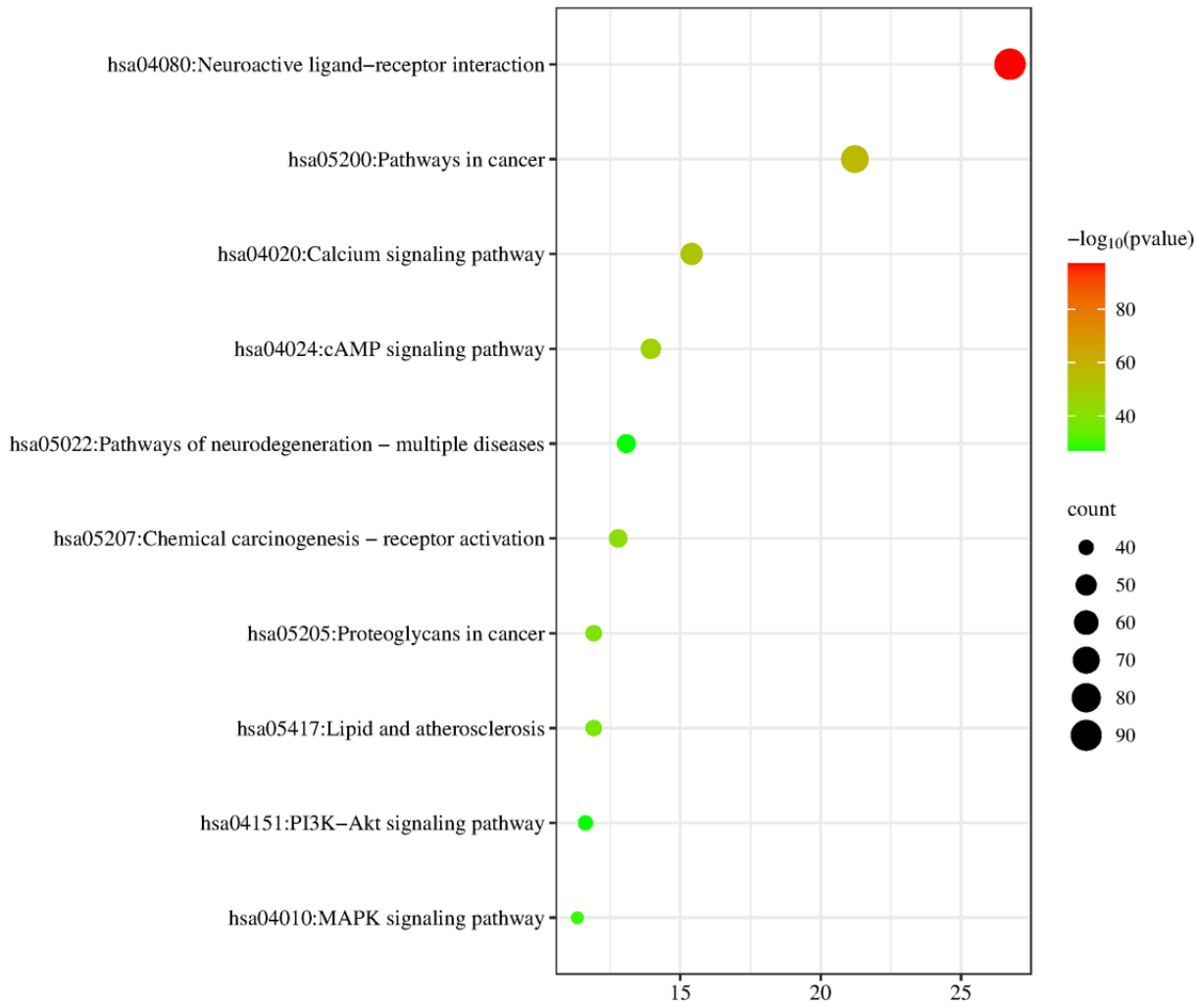


Figure 6

KEGG pathways enrichment analysis.

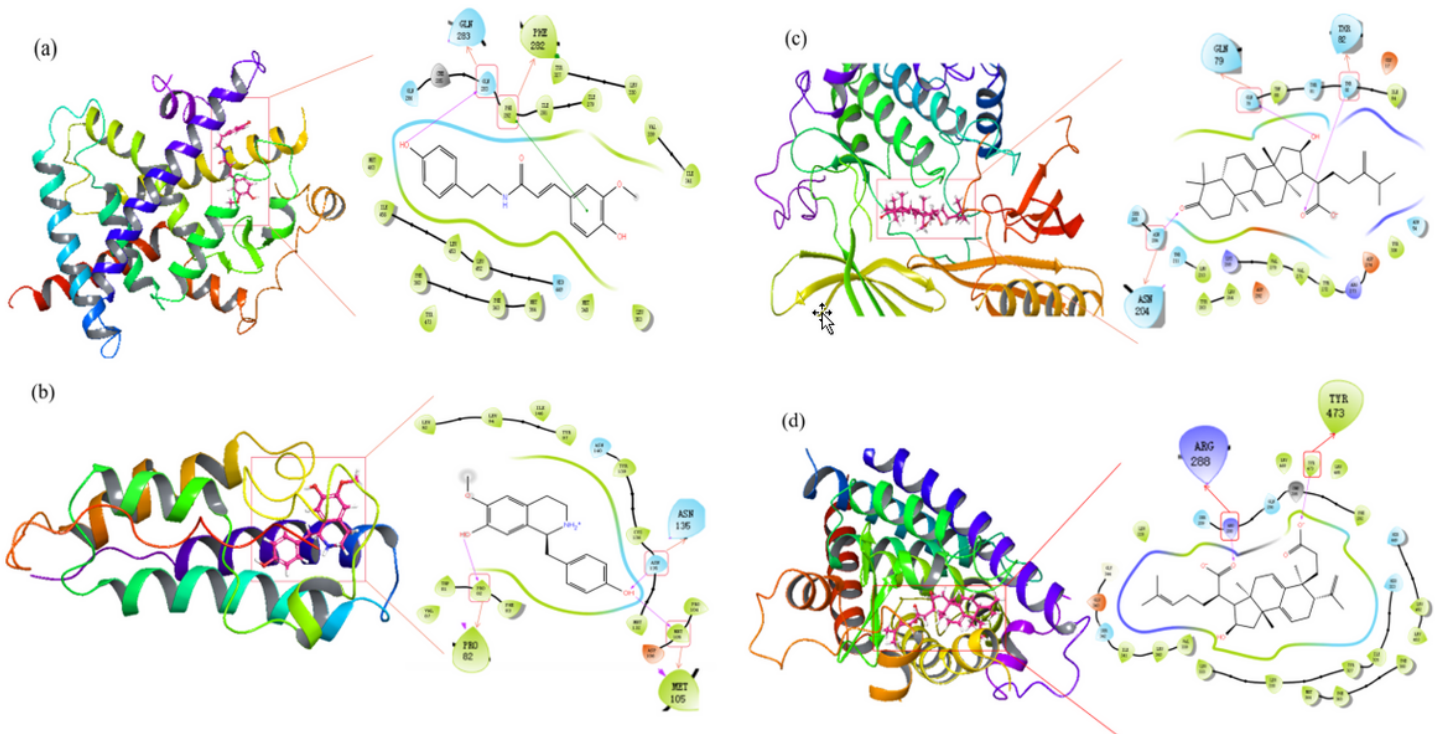


Figure 7

The molecular docking mode of active compounds with 13 target genes. (a) Action mode of n-trans-feruloyltyramine (MOL008647) with PPARG (PDB: 6K0T); (b) Action mode of (S)-Coclaurine (MOL001522) with ESR1 (PDB: 7JKY); (c) Action mode of enoic acid (MOL000285) with AKT1 (PDB: 7NH5); (d) Action mode of Poricoic acid B (MOL000291) with PPARG (PDB: 6K0T).