Network pharmacology-based active compounds and pharmacological mechanisms of ginseng for depression in post-COVID-19

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Research

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Abstract

Introduction: The novel coronavirus disease 2019 (COVID-19) is in the midst of worldwide panic. Sudden onset of an immediate life-threatening illness, quarantine and unemployment caused by epidemic are all contributors to depression. Ginseng has been reported to be an effective and safe clinical treatment on both immune-regulation and anti-depression. However, the mechanism of its anti-depression effect has not been fully characterized. In order to provide theoretical guidance for further clinical application in post-pandemic, we investigated active compounds and pharmacological mechanisms of ginseng to exert anti-depressant activity using network pharmacology, and discussed the active ingredients with immune-regulation and anti-depression.

Methods: Information on compounds in ginseng was obtained from public databases, and genes related to depression were gathered using the GeneCards database. Networks of ginseng-associated targets and depression-related genes were constructed through STRING database. Potential targets and pathway enrichment analysis related to the therapeutic efficacy of ginseng for depression were identified using Cytoscape and Database for Annotation, Visualization and Integrated Discovery (DAVID).

Results: Network pharmacological analysis of ginseng in treatment of depression identified 16 active ingredients, 47 potential targets, 32 GO terms, and 8 target gene-regulated major pathways. Among them, kaempferol, beta-sitosterol, stigmasterol, fumarine and frutinone A are bioactive compounds and key chemicals. Core genes in PPI network were AKT1, CASP3, NOS3, TNF, and PPARG. Enrichment results revealed that ginseng could regulate multiple aspects of depression through neuroactive ligand-receptor interaction, HIF-1 signaling pathway, and Serotonergic synapse. More importantly, we found that frutinone A and kaempferol are key ingredients in ginseng with dual activities of immune-regulation and anti-depression.

Conclusions: We discovered that the therapeutic activities of ginseng for depression mainly involve neurotransmitters, neurotrophic factors, neurogenesis, HPA axis and inflammatory response. Pharmacological network analysis can help to explain the potential effects of ginseng for treating depression, indicating that ginseng is a preferable herb clinically for immune-regulation and anti-depression in post-pandemic.

1. Background

The novel coronavirus disease 2019 (COVID-19), caused by the severe acute respiratory syndrome coronavirus-2, is in the midst of worldwide panic and global health concern since December 2019 [1]. The most common symptoms of COVID-19 illness are fever, cough, and dyspnea [2]. In the most severe cases, people with poor immunity may develop pneumonia and multiple organ failure, eventually leading to death. However, no specific drug or vaccine has yet been developed and the epidemic will not be brought under positive control in the short term. To our knowledge, the onset of a sudden and immediately life-threatening illness could lead to posttraumatic stress disorder [3]. Quarantine is necessary to manage the
outbreak, and the experience of being quarantined can, in some cases, lead to long-term adverse mental health consequences [4]. Furthermore, many other economic problems caused by epidemic, such as financial difficulties and loss of employment, undoubtedly increased public anxiety and depression. Based on these, it is of great significance to prevent the outbreak of depression after a global epidemic while improving the public immunity.

Panax ginseng Meyer, as a precious tonic traditional Chinese medicine (TCM), usually grows in cooler areas like Northeast China, Korea peninsula and Russia. It is known as the king of invigorating Qi and the Greek term “Panax”, which means “cure of all diseases”, implied its important position in the medical field. TCM theory emphasizes "Healthy Qi is stored inside, evil can not invade". Healthy Qi is equivalent to what we now call immunity. Ginseng can regulates each type of immune cells, therefore maintaining homeostasis of the immune system and enhancing resistance to illness or microbial attacks [5]. Quan et al. also found ginseng and salviae herbs play a role as immune activators and modulate immune responses during influenza virus infection [6]. As recorded in the traditional Chinese work, Jingyue Quanshu (Jing-yue’s Complete Works), ginseng has been used in the classic prescription Qifu Yin to treat neurasthenia and other nervous system diseases. Ginsenosides, as the main pharmacologically active components of ginseng, have been found to exhibit as novel antidepressant agents [7]. It can be seen that ginseng has a good effect on both immune-regulation and anti-depression. Hao et al. integrated metabolomics and network pharmacology to study the immune-regulation mechanism of ginseng. Twenty compounds of ginseng associated with metabolomic changes were selected by the network pharmacology analysis, including ginsenoside Re, ginsenoside Rg1, frutinone A, and kaempferol [8]. However, its anti-depression therapeutic mechanisms have not yet been clearly elucidated.

Network pharmacology has been introduced in recent years for exploring the molecular mechanisms of TCM. The key ideas of emerging network pharmacology and network biology shares much with the holistic philosophy of TCM, updating the research paradigm from the current “one target, one drug” mode to a new “network target, multi-components” mode [9]. Through bridging the emerging network science and ancient TCM, we obtain novel methodologies and opportunities for discovering bioactive ingredients and biomarkers, potentially revealing mechanisms of action. Even though the anti-depression effects of ginseng have been reported, network pharmacology-based prediction of the bioactive components and target pathways has not been performed.

In this study, we investigated active compounds and pharmacological mechanisms of ginseng to exert anti-depressant activity using network pharmacology. The flowchart of the experimental procedures of our study is shown in Fig. 1. We also discussed the active ingredients with immune-regulation and anti-depression, in order to provide theoretical guidance for further clinical application in post-pandemic.

2. Methods

2.1 Database construction and ADME screening of ginseng ingredients
All of the known ingredients of ginseng were manually collected from related literature and phytocchemical databases [10]: Traditional Chinese Medicine Systems Pharmacology Database (TCMSP, http://tcmspw.com/tcmsp.php). An in silico integrative model—ADME (absorption, distribution, metabolism, and excretion) was used to select the ingredients with favorable pharmacokinetics properties. The parameter used in this study included oral bioavailability (OB) and drug-likeness (DL). OB refers to the rate and degree of drug absorption into human circulation after oral administration and DL refers to the similarity of a compound to a known drug. They are two of the most commonly used pharmacokinetic properties in drug screening. Ingredients conforming to the requirements of both OB \( \geq 30\% \) and DL \( \geq 0.18 \) were regarded as active ingredients in ginseng for further analysis.

2.2 Target genes related to the identified compounds

In the TCMSP database, the potential targets of each active ingredient were found. The Uniprot data library (https://www.uniprot.org/) was used to convert the selected target into a gene name of Homo sapiens (Human) to prepare the target genes related to the identified compounds. Component-target data of ginseng were obtained after deleting the duplicates.

2.3 Collection of therapeutic targets of ginseng for depression

Information on depression-associated target genes was collected from the human gene database (GeneCards, http://www.genecards.org/) using “depression” as the keywords, and only “Homo sapiens” proteins linked to depression were selected. With relevance score > 7.32 as the threshold, the obtained targets were screened. In order to obtain the potential anti-depressant targets of ginseng, the targets of each active ingredient were intersected with depression-related targets through an online mapping platform (http://bioinfo. Genotoul.fr/jvenn/example.html).

2.4 Protein-protein interaction data

The potential anti-depressant targets of ginseng were imported into the String database (http://stringdb.org/) for the analysis of protein-protein interaction (PPI). A confidence score above 0.4 indicates an interaction relationship. So the confidence was set as confidence > 0.4, while other parameters remained defaults.

2.5 Construction of the pharmacological networks

All visualized network models were established via Cytoscape 3.8.0 (http://www.cytoscape.org/), an open software package project for visualizing, integrating, modeling and analyzing the interaction networks [11]. In the network, nodes represent compounds or target genes, while edges represent the interactions between them. The “Network Analyze” in Cytoscape 3.8.0 is used to analyze the topology properties of the Network. Three networks were constructed: (1) Compound-compound target network. The potential targets of each active ingredient in ginseng were used to generate it; (2) Compound-target-disease network. The potential anti-depressant targets of ginseng were used to generate it; (3) PPI network of depression-related targets in ginseng.
2.6 Gene ontology and pathway enrichment analysis

The potential anti-depressant targets of ginseng was input into the Database for Annotation, Visualization and Integrated Discovery (DAVID, https://david.ncifcrf.gov/tools.jsp) for Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analysis. Gene species and background were limited to human. The false discovery rate (FDR) < 0.05 was used as the filter to obtain the top GO terms of biological process (BP), cell component (CC) and molecular function (MF). The enrichment analysis of KEGG metabolic pathway was screened under the conditions of P < 0.01 and FDR < 0.05. The results were visualized using Graphpad Prism 8.0.2 (https://www.graphpad.com/).

3. Results

3.1 Candidate compound screening for ginseng

Retrieved from TCMSP, there were 190 components related to ginseng in total. Under the screening thresholds of OB ≥30% and DL ≥0.18, 22 active ingredients were selected for further analysis (Table 1). Among them, there are five active ingredients with higher OB: Celabenzine (101.88%), Aposiopolamine (66.65%), Frutinone A (65.9%), Inermin (65.83%) and Girinimbin (61.22%). These components play important roles in the pharmacological activities of ginseng.

3.2 Potential target prediction for ginseng

Among the 22 active components, 5 of them did not find corresponding targets, while the remaining 17 components obtained 109 potential targets after removing duplicates. All active compounds, their targets and the interactions between them are presented in the compound-compound target network (Fig. 2), which is composed of 126 nodes (17 components and 109 targets) and 246 edges. The pink nodes represent drug targets while the blue nodes represent compounds, and the edges represent the interactions between them. Degree, a topological parameter describing the importance of a node, stands for the number of edges connecting to the node. We used it to further determine the importance of each active component. Through analysis, we found that RS5 exhibited the largest number of potential targets connections (Kaempferol, degree = 59), followed by RS3 (Beta-sitosterol, degree = 36), RS2 (Stigmasterol, degree = 31), RS17 (Fumarine, degree = 24) and RS4 (Inermin, degree = 16). The same active ingredient can act on different targets, and different active ingredients can also act on the same target, which fully reflects the multi-component, multi-target action characteristics of ginseng.

3.3 Collection of therapeutic targets of ginseng for depression

A total of 11478 disease targets were obtained from GeneCards with “depression” as the keyword. Among them, 975 depression-related genes meet the requirement of relevance score > 7.32 and ginseng have 47 potential targets for depression (Fig. 3a). Compound-target-disease network (Fig. 3b) with 63 nodes and 102 edges linked 16 compounds and 47 target genes related to depression. Among the 16 candidate
compounds, RS5 exhibited the largest number of potential anti-depression targets connections (Kaempferol, degree = 24), followed by RS3 (Beta-sitosterol, degree = 18), RS2 (Stigmasterol, degree = 14), RS17 (Fumarine, degree = 9) and RS10 (Frutinone A, degree = 8). For the 47 potential anti-depression targets, the network showed PTGS2 had the largest number of compound-target interactions, followed by SCN5A, GABRA1, CHRNA7 and SLC6A4, while the remaining 42 targets showed interactions with up to three compounds.

3.4 Protein-protein interaction analysis

After removing a free target, the PPI network (Fig. 4) contains 46 nodes and 259 edges, with average node degree of 11 and average local clustering coefficient of 0.533. Target size and color are used to reflect the degree, while edge thickness and color are used to reflect the combine score. The important targets were painted red and located centrally in the network. AKT1 (degree = 25), CASP3 (degree = 20), NOS3 (degree = 19), TNF (degree = 19), PPARG (degree = 18), SLC6A4 (degree = 18), ACHE (degree = 18), IL1B (degree = 17), PTGS2 (degree = 17) and MAOA (degree = 16) were the top ten genes regarding their degree. AKT1 also has the highest closeness centrality (0.67), indicating the faster the signal is transferred to other nodes. Due to the highest betweenness centrality (0.14), AKT1 is considered a bottleneck node in monopolistic position between modules in the network and more suitable as a therapeutic target.

3.5 GO biological process and KEGG pathway enrichment analysis

Through DAVID database, we obtained 305 GO terms (226 BP terms, 31 CC terms and 48 MF terms) and 68 pathways in total. With FDR < 0. 05 as the screening condition, 32 GO terms (22 BP terms, 6 CC terms and 4 MF terms) were selected (Fig. 5). GO analysis revealed that target genes were majorly associated with the biological process of response to drug (GO:0042493), positive regulation of cell proliferation (GO:0008284), positive regulation of nitric oxide biosynthetic process (GO:0045429), response to hypoxia (GO:0001666), positive regulation of ERK1 and ERK2 cascade (GO:0070374), cellular response to organic cyclic compound (GO:0071407), response to nicotine (GO:0035094), regulation of insulin secretion (GO:0050796), cellular response to hypoxia (GO:0071456), response to ethanol (GO:0045471) and etc. The 6 cell component terms were involved in plasma membrane (GO:0005886), integral component of plasma membrane (GO:0005887), membrane raft (GO:0045121), neuron projection (GO:0043005), postsynaptic membrane (GO:0045211) and Caveola (GO:0005901). Among them, plasma membrane and integral component of plasma membrane accounted for the largest proportion, with 29 and 15 targets respectively. Depending on the outcomes of GO enrichment, the enriched molecular function ontologies were dominated by protein homodimerization activity (GO:0042803), enzyme binding (GO:0019899), heme binding (GO:0020037), extracellular ligand-gated ion channel activity (GO:0005230). Under the conditions of P < 0.01 and FDR < 0.05 (Table 2), we found that pathways in neuroactive ligand-receptor interaction (hsa04080), African trypanosomiasis (hsa05143), HIF-1 signaling pathway (hsa04066), Leishmaniasis (hsa05140), Toxoplasmosis (hsa05145), Serotonergic synapse (hsa04726), Tuberculosis (hsa05152) and Malaria (hsa05144) are the interaction pathways that ginseng exerts combined antidepressant effects (Fig. 6).
4. Discussion

Depression is a complex disorder with multiple etiologies including genetic, epigenetic, and environmental factors. Some possible pathophysiological mechanisms of depression include altered neurotransmission, hypothalamic-pituitary-adrenal (HPA) axis abnormalities involved in chronic stress, inflammation, reduced neuroplasticity, and network dysfunction. All of the proposed mechanisms are not separate but integrally related and interact bidirectionally [12]. Currently, many antidepressants work on a single target. Selective serotonin reuptake inhibitors (SSRIs) have been the first line of antidepressants for the past 25 years, although they have only moderate efficacy and may take weeks to produce measurable benefits. TCM is characterized by multi-target, multi-pathway and low side effect. Multi-component therapy produces "synergies", in which the combined effects are greater than the sum of the individual effects. To our knowledge, the anti-depression mechanisms of active ingredients from TCM can be summed up as following: increasing synaptic availability of monoamines [13], alleviation of the HPA axis dysfunctions [14], regulation of the cAMP signaling pathway [15] and caspases [16], and amelioration of the dysregulation of immune and inflammation [17]. Ginseng has been shown to be a precious herbal medicine with dual activities of immune-regulation and anti-depression. In this study, we investigated active compounds and pharmacological mechanisms of ginseng to exert anti-depressant activity using network pharmacology.

The screened active components can be classified as flavonoids, saponins, sterols, organic acids and etc. It is not difficult to find that some nodes are more concentrated in the network than others. That is, the compound-compound target space was biased towards certain compounds and targets [18]. As the most common flavonoid, kaempferol has attracted much attention due to its anti-cancer, anti-inflammatory, antioxidant, antibacterial and antiviral effects. Gao et al. have confirmed that antidepressive effects of kaempferol are mediated by reduction of oxidative stress, proinflammatory cytokines and up-regulation of AKT/β-catenin cascade [19]. Stigmasterol has been found to exert neuro-protective effect through reduction of oxidative stress and inactivation of autophagy via AMPK/mTOR and JNK pathways [20]. It has been reported that β-sitosterol has protective effects on various brain-related diseases independent of their lipid-lowering effects. β-sitosterol can also suppress the development of cerebral aneurysm by inhibiting inflammatory reactions including TNF-α [21]. This indicated that our prediction of the active ingredient in ginseng that plays an antidepressant role is reasonable. However, frutinone A are mostly considered as an active ingredient of the broad spectrum antimicrobial herbal extract up to now and anti-depression may be its potential pharmacological activity that has not yet been discovered. The compound-target-disease network showed intimate communications between several components and multiple targets, and facilitated a better understanding of the possible curative mechanism of ginseng in the treatment of depression.

In the PPI network, AKT1 has the highest degree, closeness centrality and betweenness centrality. It is in the monopolistic position between modules in the network and more suitable as a therapeutic target. AKT1, a downstream enzyme that has been implicated in the pathogenesis of serotonin-related disorders, facilitates growth factor-mediated cell survival and block apoptosis through the regulation of its
downstream effector (GSK-3) in depressants [22-23]. Thus, the regulation of AKT1 activity by ginseng (kaempferol) inhibit and alleviate depression by a variety of ways. Moreover, SLC6A4 is responsible for the serotonergic neurotransmission, which is an important substance that regulates neural activity. The serotonin-linked polymorphic region (5-HTTLPR) is a degenerate repetition of the gene encoding the 5-hydroxytryptamine transporter (SLC6A4). The S/S genotype in this region is associated with decreased serotonin expression and increased susceptibility to depression [12]. TNF and IL-1B may be involved in major depression not just by activating certain cascades of inflammation but by many other mechanisms [24]. It is not difficult to see that ginseng's treatment of depression involves neurotransmitters (especially serotonin), inflammation, neurotrophic factors and many other aspects. Furthermore, inflammatory cytokines have been shown to reduce monoamine levels in depressed patients by increasing the metabolism of tryptophan, an important precursor to serotonin [12]. It also reconfirmed the factors that influence depression are not separate but integrally related and interact bidirectionally. These results were consistent with our enrichment analysis, and have been previously reported to be related to depression.

GO analysis revealed that target genes were majorly associated with the biological process of response to drug, indicated the key requirement of avoiding drug dependence in depression drug development and clinical treatment [25]. Through positive regulation of cell proliferation, ginseng affect neurogenesis, which is thought to contribute to anti-stress recovery and underlie the clinical efficacy of antidepressants [26]. Extracellular signal-regulated kinase 1/2 (ERK1/2) signaling, which belongs to the large family of mitogen-activated protein kinase signaling cascades, showed low activity in the frontal cortex (Brodmann 8, 9, 10) and hippocampus in depressed patients [27]. It has been shown to have significant effects on the regulation of long-term enhancement, long-term depression, and neuronal survival through neurotrophic/growth factors [28-29]. In recent years, more and more studies have proved that astrocytes not only have a supporting effect, but also regulate physiological processes by secreting neurotrophic factors. This suggests that ginseng can play a direct and indirect therapeutic role by acting on astrocytes or nerve cells. Glucocorticoid is the main cytokine released by macrophages, and ginseng's effect on lipopolysaccharide-mediated signaling pathway is important to regulate inflammation and HPA axis dysfunction. The main cell component terms were involved in plasma membrane and integral component of plasma membrane, while the enriched molecular function ontologies were dominated by protein homodimerization activity and enzyme binding. In addition, KEGG pathway analysis were primarily pertaining to neuroactive ligand-receptor interaction, serotonergic synapse and HIF-1 signaling pathway, which followed the previous reports that these pathways participate in crucial functions in the progression and development of depression. It is worth mentioning that there were several pathways which are concerned with diseases, such as African trypanosomiasis, Leishmaniasis, Toxoplasmosis, Tuberculosis and Malaria, prompting that ginseng may have potential therapeutic effects on these diseases. Twelve targets enriched onto neuroactive ligand-receptor interaction pathway were mostly neurotransmitter receptors. Monoamine neurotransmitters (serotonin, norepinephrine and dopamine) have a potential role in the pathogenesis of depression. The monoamine hypothesis explained how antidepressants work [30]. HIF-1 signaling pathway is associated with AKT, NF-κB signaling pathways and the transcription of VEGF, which interferes with inflammatory injury, oxidative stress, apoptosis and
tumor growth. It is FG-4592 that improves depressive-like behaviors through HIF-1-mediated neurogenesis and synapse plasticity in rats [31]. All of the proposed biological pathophysiological mechanisms of depression are reciprocally connected with each other. For example, inflammation leads to HPA hyperactivity, which in turn plays a permissive role in the inflammatory process, forming a vicious circle. Through enrichment analysis, we discovered that the therapeutic activities of ginseng for depression involve neurotransmitters, neurotrophic factors, neurogenesis, HPA axis and inflammatory response. Taken together, these results indicate that ginseng can regulate whole-body systems through a complex genes-interaction network, resulting in a certain effect in depression.

As mentioned above, ginsenoside Re, ginsenoside Rg1, frutinone A and kaempferol were the key ingredients in ginseng for immune-regulation, regulating the choline metabolism through acetylcholinesterase or cholinesterase [32]. Through analysis, we found frutinone A and kaempferol are also the major ingredients in ginseng to exert anti-depressant activity. Therefore, these two ingredients are the key ingredients in ginseng with dual activities of immune-regulation and anti-depression. These findings could provide theoretical guidance in the investigation of ginseng for further clinical application in post-COVID-19. However, experiment is still needed to validate the interactions between drugs and proteins based on theoretical predictions.

5. Conclusions

Past experience shows that mental health problems often occur in the public following a major epidemic situation. In this study, we investigated the active compounds and pharmacological mechanisms of ginseng to exert anti-depressant activity using network pharmacology, and discussed the active ingredients with immune-regulation and anti-depression. Kaempferol, beta-sitosterol, stigmasterol, fumarine and frutinone A are major ingredients in ginseng to exert anti-depressant activity. We discovered that the therapeutic activities of ginseng for depression mainly involve neurotransmitters, neurotrophic factors, neurogenesis, HPA axis and inflammatory response. A total of 47 potential targets were identified, which were significantly enriched in the neuroactive ligand-receptor interaction, serotonergic synapse and HIF-1 signaling pathway, indicating that the therapeutic effects of ginseng were produced by synergistic complex interactions among its chemical compounds and related targets. More importantly, frutinone A and kaempferol are key ingredients in ginseng with dual activities of immune-regulation and anti-depression.

These findings could be guidelines in the investigation of ginseng to exert anti-depressant activity, in order to provide theoretical guidance for further clinical application in post-COVID-19.

Abbreviations

COVID-19, coronavirus disease 2019; TCM, traditional Chinese medicine; OB, oral bioavailability; DL, drug-likeness; TCMSP, Traditional Chinese Medicine Systems Pharmacology Database; ADME, absorption, distribution, metabolism, and excretion; DAVID, Database for Annotation, Visualization and Integrated
Discovery; PPI, protein-protein interaction; HPA axis, hypothalamic-pituitary-adrenal axis; GO, Gene Ontology; KEGG, Kyoto Encyclopedia of Genes and Genomes; FDR, false discovery rate; BP, biological process; CC, cell component; MF, molecular function.

**Declarations**

**Author contributions**

NW wrote the paper and drew the figures; XLW sorted the data and created the tables; MJH and WXZ modified the tables and revised the paper; MQD and XQC revised the paper; YQZ and CCH directed the research and revised the paper.

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Not applicable.

**Competing interests**

The authors declare no competing interests.

**Availability of data and materials**

The data and materials generated or analyzed during this study are available from the corresponding author on reasonable request.

**Consent for publication**

The manuscript is approved by all authors for publication.

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Tables

Table 1 Active compounds and ADME parameters of ginseng (OB ≥30%, DL ≥0.18).
| ID  | MOL ID      | Molecule Name      | OB(%) | DL  |
|-----|-------------|--------------------|-------|-----|
| RS1 | MOL002879   | Diop               | 43.59 | 0.39|
| RS2 | MOL000449   | Stigmasterol       | 43.83 | 0.76|
| RS3 | MOL000358   | Beta-sitosterol    | 36.91 | 0.75|
| RS4 | MOL003648   | Inermin            | 65.83 | 0.54|
| RS5 | MOL000422   | Kaempferol         | 41.88 | 0.24|
| RS6 | MOL005308   | Aposiopolamine     | 66.65 | 0.22|
| RS7 | MOL005317   | Deoxyharringtonine | 39.27 | 0.81|
| RS8 | MOL005318   | Dianthramine       | 40.45 | 0.20|
| RS9 | MOL005320   | Arachidonate       | 45.57 | 0.20|
| RS10| MOL005321   | Frutinone A        | 65.9  | 0.34|
| RS11| MOL005344   | Ginsenoside Rh2    | 36.32 | 0.56|
| RS12| MOL005348   | Ginsenoside Rh4    | 31.11 | 0.78|
| RS13| MOL005356   | Girinimbin         | 61.22 | 0.31|
| RS14| MOL005376   | Panaxadiol         | 33.09 | 0.79|
| RS15| MOL005384   | Suchilactone       | 57.52 | 0.56|
| RS16| MOL005399   | Alexandrin         | 36.91 | 0.75|
| RS17| MOL000787   | Fumarine           | 59.26 | 0.83|
| RS18| MOL004492   | Chrysanthemaxanthin| 38.72 | 0.58|
| RS19| MOL005314   | Celabenzine        | 101.88| 0.49|
| RS20| MOL005357   | Gomisin B          | 31.99 | 0.83|
### Table 2 Functions of potential target genes based on KEGG pathway analysis.

| Pathway ID | Term                                  | Target genes                                    | Count |
|------------|---------------------------------------|-------------------------------------------------|-------|
| hsa04080   | Neuroactive ligand-receptor interaction | OPRM1, DRD1, GABRA2, GABRA1, CHRMA2, GABRA5, DRA2A, ADRA1A, CHRMA7, NR3C1, OPRD1, HTR2A | 12    |
| hsa05143   | African trypanosomiasis                | PRKCA, VCAM1, ICAM1, TNF, IFNG, IL1B             | 6     |
| hsa04066   | HIF-1 signaling pathway               | PRKCA, AKT1, HMOX1, BCL2, IFNG, NOS3, NOS2, INS | 8     |
| hsa05140   | Leishmaniasis                         | TNF, PTGS2, IFNG, IL1B, NOS2, STAT1, TGFB1      | 7     |
| hsa05145   | Toxoplasmosis                         | AKT1, CASP3, TNF, BCL2, IFNG, NOS2, STAT1, TGFB1| 8     |
| hsa04726   | Serotonergic synapse                  | PRKCA, CASP3, PTGS2, MAOA, SLC6A4, MAOB, HTR3A, HTR2A | 8     |
| hsa05152   | Tuberculosis                          | AKT1, CASP3, TNF, BCL2, IFNG, IL1B, NOS2, STAT1, TGFB1 | 9     |
| hsa05144   | Malaria                               | VCAM1, ICAM1, TNF, IFNG, IL1B, TGFB1            | 6     |

**Figures**
Figure 1

The flowchart of the network pharmacology-based strategy for deciphering the mechanisms of ginseng on depression.
Figure 2

Compound-compound target network. The size of the node is proportional to the value of the degree.
Figure 3

(a) Venn diagram. (b) Compound-target-disease network with 63 nodes and 102 edges linking 16 compounds and 47 target genes related to depression.
Figure 4

PPI network of depression-related targets in ginseng.
Figure 5

GO functional annotation analysis of 47 target genes were sorted according to FDR < 0.05.
KEGG pathways analysis of 47 target genes were sorted according to $P < 0.01$ and FDR $< 0.05$. 

**Figure 6**

KEGG pathways analysis of 47 target genes were sorted according to $P < 0.01$ and FDR $< 0.05$. 

**Fig. 6**