Research Square

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Novel Compatibility of Huanglianjiedu Decoction in Behavioral and Psychological Symptoms of Dementia in Alzheimer's Disease[1]

Su-Ya Ma
Beijing University of Chinese Medicine
Xu Wang
Beijing University of Chinese Medicine
Xiang-Yi Du
Beijing University of Chinese Medicine
Ming-Wan Su
Beijing University of Chinese Medicine
Xuan Yao
Beijing University of Chinese Medicine
Jing Shi (shijing872020@163.com)
Department of Neurology, Dongzhimen Hospital, Beijing University of Chinese Medicine, Beijing, 100007, China

Research

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Abstract

Background: Alzheimer's disease (AD) is characterized by progressive cognitive decline. Behavioral and psychological symptoms in dementia (BPSD) are another critical characterization of AD besides cognitive deficit. However, the pharmacological treatment of BPSD remains challenging. HuanglianJiedu decoction (HLJDD), which consists of four herbs, is applied to treat Alzheimer's disease in traditional Chinese medicine, especially AD with BPSD. While the frequently used compatibility of HLJDD, whose principal ingredient is Coptidis Rhizoma (Huang-lian, CR), may not be suitable for treating BPSD. Elucidating the mechanism underlying each herb is critical to HLJDD's pertinent compatibility.

Methods: We utilized network pharmacology to analyze the herbs' targets and biological processes for treating BPSD in HLJDD and employed molecular docking to explore the binding activity between herbs' main active ingredients and neurotransmitter receptors.

Results: Results showed that Scutellariae Radix (Huang-qin, SR) and Phellodendri Chinrnsis Cortex (Huang-bai, PCC) have better anti-BPSD effects than CR and Gardeniae Fructus (Zhi-zi, GF). SR has a better anti-neuroinflammation function, can better regulate blood vessels. PCC has a higher binding affinity with Dopamine D2 receptor (DRD2) and 5-hydroxytryptamine receptor 2A (HTR2A). CR and GF may be better for neuronal signaling.

Conclusion: For treating BPSD, SR and PCC are the principal ingredients while CR and GF are the ancillary herbs.

Background

In traditional Chinese medicine (TCM), herbs combined into a formula to reinforce the overall effects or eliminate the adverse effects [1], which is called the compatibility of TCM. Although formulas generally have conventional compatibility, it is essential to consider novel compatibility with different diseases. In our research, we proposed an innovative method for the compatibility of herbs based on molecular mechanisms. We investigate HLJDD’s novel compatibility in BPSD.

BPSD appears in more than 90% of AD patients, including anxiety, agitation, aggression, irritation, depression, apathy, disinhibition, delusions, or hallucinations [2]. Molecules that involve in BPSD pathogenesis have been researched by mounting studies. Neuroinflammation, a response that involves neurons and microglia, has been reported to characterize many neurodegenerative diseases and neuropsychiatric conditions, resulting in the elevated production of pro-inflammatory cytokines, like IL-6, TNF-α, IL-8, IL-4, *etc*. Microglia activation is the first sign of neuroinflammation. Activated microglia can release various oxidants such as reactive oxygen species and activate several genes and proteins, such as inducible nitric oxide synthase [3]. Neurotransmitters and their receptors are considered to play a potential role in BPSD [4]. Genetic risk factors offer a powerful approach for the elucidation of mechanisms underlying BPSD. APOE epsilon4, the most recognized genetic risk factor for late-onset AD, increasing the risk of BPSD [5]. However, the efficacy of antipsychotics for the treatment of BPSD is scanty. Antipsychotic drugs that are often used to treat BPSD have extrapyramidal severe side effects (EPS). Memantine, the NMDA receptor antagonist, a cure for treating moderate-to-severe AD, can reduce the antipsychotic-induced EPS [6] but had a controversial effect in treating BPSD. Acetylcholinesterase inhibitors, the current primary medication for AD, is controversial when treating BPSD [7]. Many researchers turn their attention to natural products as an alternative or complementary method to BPSD for their clinical efficacy with minimal side effects.

BPSD has different TCM syndrome differentiation. “Toxin damaging brain collaterals,” an essential differentiation of BPSD, is put forward by academician Yongyan Wang in the study process of dementia. For this syndrome differentiation, the brain collaterals are injured by the toxins of fire. For the treatment, purging fire for removing toxins is commonly used. HLJDD is a classic prescription for heat-clearing away and detoxifying, which is composed of SR (Huang-qin), CR (Huang-lian), PCC (Huang-bai), and Gardeniae Fructus (Zhi-zi). Studies show that HLJDD is frequently applied for Alzheimer’s disease [8], especially with BPSD [9]. While the TCM physicians seldom treat BPSD only with HLJDD. Making out the principal herbs is essential to BPSD, for the intrinsic compatibility of HLJDD maybe not suitable. To better understand HLJDD’s compatibility in treating BPSD, we utilized network pharmacology and molecular docking to provide useful drugs for BPSD therapeutics by emphasizing their molecules’ activities.

Material And Methods

Herbs' main active ingredients and their matched potential target proteins

A natural plant contains various chemical compounds. Our study used the TCM systems pharmacology database and analysis platform (TCMSP, https://tcmspw.com/index.php) to detect herbs' main active ingredients. And filtered compounds by terms of oral bioavailability (OB) ≥ 30%, drug-likeness (DL) ≥ 0.18, drug half-life (HL) ≥ 4 h. Then predicted the active ingredients' potential molecular targets by the search tool for interactions of chemicals database (STITCH, http://stitch.embl.de/) with the species limited to "Homo sapiens."

Potential targets prediction for behavioral and psychological symptoms of dementia in Alzheimer's disease

We identified BPSD-associated protein targets by the GeneCards database (http://www.genecards.org/) [10], with a higher rank score, higher correlation with BPSD. The searched keywords were "behavioral and psychological symptoms of dementia in Alzheimer's disease." We chose the top 50% of predicted targets as the potential targets for BPSD.

Network construction and analysis

We constructed the "compounds-targets-BPSD" networks by Cytoscape 3.7.2 software [11]. The .csv format files whose data combined compounds targets with BPSD targets were imported into Cytoscape. The node size was based on the target proteins score values provided by the GeneCards database. The

overlapping targets between active compounds and BPSD were the herbs' putative targets related to BPSD and set with a red rectangle node. Memantine was applied to choose the potential neurotransmitter receptors in this process.

Annotation enrichment analysis of target proteins

We performed Gene Ontology (GO) functional enrichment analysis using the ClueGO plugin [12], with the species limited to "Homo sapiens" and p-value < 0.05.

Molecular docking for active compounds with DRD2 and HTR2A

Molecular docking studies were conducted using AutoDock software [13] to evaluate the active compounds' binding affinity with neurotransmitter receptors. The neurotransmitter receptors DRD2 and HTR2A were selected as the docking receptor. The memantine's mechanism on BPSD determined to choose them. We downloaded the proteins' crystal structure from the RCSB Protein Data Bank (http://www.rcsb.org/). We chose 6CM4 and 6A93, whose ligand is Risperdal as the crystal structure. Then memantine, Risperdal, and the active compounds were docked with DRD2 and HTR2A. The binding pocket of DRD2 and HTR2A was set up regarding 6CM4 and 6A93.

Results

HLJDD's main active compounds and potential target proteins

After searching the TCMSP database, We sifted out 80 compounds in HLJDD. SR, CR, PCC, and GF have 32, 10, 27, and 11 ingredients, respectively. Then import those compounds into the STITCH database to achieve the potential target proteins (Table 1).

"Compounds-targets-BPSD" network and GO biological process analysis

In this network, the red rectangular nodes represent herbs' key protein targets of BPSD were the most in SR, followed by PCC, 24, and 15, respectively. For further research of the herbs' key protein targets, we applied GO biological process analysis. Fig. 1 b, c, d, and Tab.2 show the biological processes of the key target proteins of SR, include positive regulation of smooth muscle cell proliferation, lipopolysaccharide-mediated signaling pathway, regulation of neuroinflammatory response, *etc*. Fig. 2 b, c, d, and Tab.3 reveal the biological processes of the key target proteins of CR. Its biological processes are cellular response to cadmium ion, response to nicotine, and negative regulation of macroautophagy. Fig. 3 b, c, d, and Tab.4 illustrate PCC's key target proteins' biological processes. Include response to nicotine, glial cell apoptotic process, plasma lipoprotein particle, *etc*. Fig. 4 b, c, d, and Tab. 5 show the biological processes of the key target proteins of GF, which are mainly associated with regulation of amyloid-beta formation, regulation of membrane protein ectodomain proteolysis and regulation of nitric oxide biosynthetic process.

"Compounds-targets-BPSD" network of memantine

Patients with moderate to severe AD exhibit relatively severe cognitive and psychological symptoms. N-Methyl-D-aspartic acid (NMDA) is one of the main treatments. Memantine is the most prevalent choice of NMDA [14]. In our research, we took memantine as a reference drug. We did the "compounds-targets-BPSD" network of memantine to search its potential mechanism on BPSD, especially for the neurotransmitter receptor. The network result showed that DRD2 and HTR2A were the neurotransmitter receptors that worked on BPSD. We chose DRD2 and HTR2A to do molecular docking for their crystal structure to be better docked and studied with Risperdal ligand in the PDB database.

Results of molecular docking

Serotonin and its receptors, particularly the HTR2A, are considered to play a potential role in cognitive behaviors and psychiatric conditions such as depression, schizophrenia, and AD [15]. A multitarget-directed ligand, acting on HTR2A and DRD2, exerting an anti-aggressive and antipsychotic activity, posing a promising strategy for the treatment of BPSD [16]. In our study, the active ingredients were selected as the docked compounds with DRD2 and HTR2A. The 3D structures of the active ingredients were downloaded in .mol2 format from the TCMSP database. They were later converted into .pdb format with the Open Babel GUI software [17]. We used the local search parameters and rigid filename of the macromolecule model when docking. We set the software's default values as our docking parameters. We set grid box size as follows: DRD2: x-dimension: 62, y-dimension: 84, z-dimension: 126, spacing: 0.375, X center: 17.469, y center: 7.307, z center: 2.7. HTR2A: x-dimension: 126, y-dimension: 62, z-dimension: 84, spacing: 0.497, X center: 30.895, y center: 0.518, z center: 63.697. These parameters were minor adjusted during docking.

In our result, beta-sitosterol had the lowest docking energy with DRD2 (-9.58 Kcal/mol) and HTR2A (-8.2 Kcal/mol) than other docked compounds, include memantine and Risperdal. Stigmasterol, chelerythrine, and campesterol also have lower docking energy than memantine and Risperdal. PCC contains most of these compounds. (Tab.6 and Fig. 5).

Discussion

TCM plays an essential role in medical diagnosis and treatments. Based on the TCM theory. Chinese formulas contain a mixture of herbs, which combined the following compatibility principle "monarch, minister, assistant, and guide," meaning herbs play a primary, secondary, auxiliary, or harmonic roles, respectively [18]. The primary herbs are substances that provide the main therapeutic thrust. The second primary herbs enhance or assist the therapeutic actions of the first. The rest serve one of the following functions: treat accompanying symptoms, moderate the harshness or toxicity of the primary ones, guide the medicine to the proper organs, or exert a harmonizing effect [19]. In HLJDD, CR acts as the monarch role. SR plays the minister role. PCC and GF were the assistant and guide role. However, this compatibility is not suitable for treating BPSD. Our research studied drugs from the molecular perspective, which provides a novel method for the compatibility of formulas. Our results showed that SR and PCC are the principal herbs, CR and GF are the assistant herbs.

BPSD is a critical neuropsychiatric feature in Alzheimer's disease [4]. In Alzheimer's disease, abnormal accumulation of amyloid-β released from amyloid precursor protein and neuroinflammation are the partially pathologic hallmarks. Accumulation of amyloid-β also causes indirect injury to neurons by inducing neuroinflammation [20]. Microglia, the resident innate immune cells in the brain, is pivotal for the immune response observed in AD, acting as sentinel and protective cells, but may become inappropriately reactive in AD to drive neuropathology [21]. Lipopolysaccharide (LPS) is a gram-negative bacterial endotoxin released from the cell wall component that contributes to inflammation in the body. LPS is involved in the regulation of the expression of potent inflammatory factors [22]. Studies showed that with age, microglia exhibit enhanced sensitivity to inflammatory stimuli, similar to that observed in brains with ongoing neurodegeneration [23]. An increasing number of data has linked schizophrenia with neuroinflammatory conditions and microglia, which have been related to the pathogenesis of schizophrenia. Evidence suggests that neuroinflammatory changes observed in schizophrenia involve abnormal astrocyte functions [24]. In our research, Scutellaria baicalensis has anti-inflammatory roles in the biological process of treating BPSD, including regulating neuroinflammatory response, mediating lipopolysaccharide-mediated signaling pathway, cellular response to interleukin-6, and regulation of interferon - α production. GF can regulate amyloid-beta formation in treating BPSD.

Smooth muscle cell proliferation, especially vascular smooth muscle cells, is essential during cell growth or injury [25]. Blood vessels composed of vascular smooth muscle cells play a crucial role in normal brain function for its integrity structure and function and supplying adequate blood. Cerebral blood flow shortfalls and blood-brain barrier dysfunction are early findings in neurodegenerative disorders. Cerebral blood flow reductions, impaired cerebrovascular reactivity, and impaired hemodynamic responses are increasingly recognized in AD's early stages [26]. Platelet, a critical factor for blood flow, is an anucleate cell in blood, whose principal function is to stop bleeding by forming aggregates for hemostatic reactions. Platelet aggregates are also involved in pathological thrombosis and play an essential role in inflammation [27]. Nitric oxide (NO) is a small free radical molecule with an endothelium-derived relaxing factor. Its adequate production levels in the vascular endothelium are critical for regulating blood flow and vasodilation. Besides, NO plays a vital neuronal signaling role [28]. Cadmium, a metal that resembles zinc and calcium, is also crucial for neuronal signaling. Cadmium exposure is linked to neurodegenerative diseases like Alzheimer's disease. It can alter neurotransmitters' release, cause oxidative stress, damage mitochondrion, and induce apoptosis [29]. In our study, SR can positively regulate smooth muscle cell proliferation, positively regulate the endothelial cell proliferation, and positively regulate the blood vessel endothelial cell migration. CR can mediate cellular response to cadmium ion. PCC can regulate platelet activation. GF can regulate the nitric oxide biosynthetic process.

Macroautophagy is an evolutionarily conserved dynamic pathway that functions primarily in a degradative manner. A wide range of diseases is associated with dysregulation of macroautophagy. Macroautophagy has a critical role in cellular homeostasis. Either insufficient or excessive macroautophagy can seriously compromise cell physiology, and thus, it needs to be regulated appropriately [30]. CR can negatively regulate macroautophagy. PCC can regulate the glial cell apoptotic process.

Research showed that Nicotine might be involved in the pathophysiology of psychosis. Smoking has a relationship with depression. In animal models, Nicotine shows anxiolytic properties. Depression people are more likely to smoke and more likely to develop severe depressive episodes upon smoking cessation. Nicotine has also been observed to produce similar cognitive improvements in AD patients [31]. However, the relationship between smoking and AD is still debatable [32]. In our herbs, CR, PCC has the function to respond to Nicotine.

It has been demonstrated that serotoninergic, dopaminergic, and cholinergic systems are mainly involved in the pathogenesis of BPSD, and the role of HTR2A and DRD2 as therapeutic targets appear to be evident [16]. Our research utilized molecular docking to find potential active ingredients with good binding activities to DRD2 and HTR2A. The best-docked compound was beta-sitosterol. The free binding energy of beta-sitosterol with DRD2 and HTR2A was −9.58 kcal/mol, -8.2 kcal/mol, respectively. Stigmasterol, chelerythrine, and campesterol also have good binding activities. Beta-sitosterol, Stigmasterol, chelerythrine, and campesterol are the active ingredients of PCC. It is the only herb containing these five ingredients.

The TCM theory verifies our results. Triple energizers mean upper, middle, and lower energizer in TCM theory. They are the birth and channel to run for Qi, blood, thin, thick fluids, and essence. Moreover, they also contact five Zang-organs and six Fu-organs. SR affects the upper energizer, which is the brain and heart. PCC works on the lower energizer that is the kidney and liver. CR influences the middle energizer that consists of the spleen and stomach. Kidney essence deficiency is a primary syndrome differentiation of AD in TCM theory. "Liver fire" is the largest contributor to BPSD due to the imbalance between yin and yang of liver function [33, 34]. PCC acts on the lower energizer can purge the liver fire. "Su Wen" puts forward that "the mind is the monarch's official, and the gods come out of it." In the compendium of Materia Medica, Shizhen Li of the Ming Dynasty proposed that "the brain is the house of primordial God." SR works on the heart and brain, belongs to the upper energizer, is beneficial for the heart and brain. In conclusion, SR and PCC are HLJDD's primary herbs based on the TCM theory.

Conclusions

The therapeutic value of natural products in BPSD has increased in reputation due to their clinical impact and insignificant side effects. Recently, different types of compounds were reviewed for their biological activities. In this review, we summarize the natural products of HLJDD for the molecules' targets and biological processes involved in the treatments of BPSD. Furthermore, put forward novel compatibility of HLJDD to BPSD. Our results showed that SR has more molecule targets and biological processes involved in BPSD, PCC contains more good-docked compounds: poriferast-5-en-3beta-ol (beta-sitosterol), Stigmasterol, chelerythrine, and campesterol, which have lower affinity energy with DRD2 and HTR2A. SR and PCC are the primary drugs in treating BPSD. SR plays an anti-inflammatory role; PCC can regulate the apoptotic process and respond to nicotine. All of them can regulate blood vessels. CR and GF play the assistant role in BPSD. They have a better position on neuronal signaling.

Abbreviations

AD
Alzheimer's disease; BPSD:Behavioral and psychological symptoms in dementia; HLJDD:HuanglianJiedu decoction; CR:Coptidis Rhizoma (Huang-lian); SR:Scutellariae Radix (Huang-qin); PCC:Phellodendri Chinrnsis Cortex (Huang-bai); GF:Gardeniae Fructus (Zhi-zi); DRD2:Dopamine D2 receptor; HTR2A:5-hydroxytryptamine receptor 2A; TCM:traditional Chinese medicine; EPS:extrapyramidal severe side effects; TCMSP:TCM systems pharmacology database and analysis platform; OB:oral bioavailability; DL:drug-likeness; HL:half-life; STITCH:search tool for interactions of chemicals database; GO:Gene Ontology; D-CS:BPSD correlation score; NMDA:N-Methyl-D-aspartic acid; LPS:Lipopolysaccharide; NO:nitric oxide.

Declarations

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Not applicable.

Authors' contributions

SM, XW, and XD conceived and designed the study. SM, XW, and MS constructed the pharmacology networks. SM, XD, and XY performed molecular docking analysis. SM wrote the manuscript. XW and XD edited pictures. JS revised the manuscript. All authors were responsible for reviewing data. All authors read and approved the final manuscript.

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Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing financial interest.

Author details

[1] Department of Neurology, Dongzhimen Hospital, Beijing University of Chinese Medicine, Beijing, 100007, China.

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Tables

Table1 HLJDD's active compounds and potential molecular target proteins

Herbs	Active ingredients	Potential target proteins
SR	acacetin	IL13，CYP1A2，VEGFA，JUN，STAT1，NR1I2，SELE，CYP1A1，IL5，CYP1B1
SR	wogonin	MMP9，MYC，HMGB1，MCL1，PTGS2，CDK9，GATA1，KCNK10，PLSCR1，CCL2
SR	baicalein	MAPK1，ALOX15，CDK4，MMP2，PLAU，CYP1A2，AKT1，CYP3A4，ALOX12，MMP9
SR	oroxylin a	MAPK3，BDNF，HK2，SIRT3，CASP8，PPIF，IL6，SOD2，PARP1，NOS1
SR	Panicolin，skullcapflavone I，	CASP3
SR	beta-sitosterol	DHCR24，CASP3，ABCG5，SREBF2，ICAM1，ABCG8，CYP7A1，SREBF1，APOE，ABCB11
SR	Stigmasterol	TNF，IL8，ABCA1，ABCG8，NR1H2，SREBF2，ABCG5，NR1H3，IL10，SLCO1B1
SR	coptisine	PAWR，XPO1，TNFSF11，ESF1
CR	quercetin	MCL1，ATP5B，CYP1A1，HIBCH，HCK，STK17B，SLC2A2，CYP1B1，CYP2C8，PIM1
CR	berberine	CCND1，CASP3，DPP4，TP53，AKT1，MAPK1，HMOX1，LDLR，PCSK9，ATP5G2
CR	coptisine	PAWR，TNFSF11，ESF1，XPO1
PCC	berberine	PCSK9，ATP5G2，HMOX1，MAPK1，TP53，CASP3，AKT1，DPP4，LDLR，CCND1
PCC	coptisine	PAWR，XPO1，ESF1，TNFSF11
PCC	rutaecarpine	CYP1A1，TNF，CYP1A2
PCC	Chelerythrine	GAPDH，PRKCE，PLA2G1B，F2，CFTR，CHI3L2，SELP，PLA2G2A，CORT，ABCB11
PCC	Stigmasterol	IL10，TNF，SREBF2，IL8，SLCO1B1，ABCG8，ABCG5，NR1H2，NR1H3，ABCA1
PCC	beta-sitosterol	ABCG5，SREBF2，DHCR24，CYP7A1，ABCB11，CASP3，ICAM1，SREBF1，ABCG8，APOE
PCC	Fumarine，protopine，	HRH1，F2，KIAA0101
PCC	quercetin	CYP1B1，STK17B，PIM1，SLC2A2，CYP1A1，HCK，ATP5B，HIBCH，CYP2C8，MCL1
PCC	poriferast-5-en-3beta-ol，beta-sitosterol，	CASP9，NR1H2，PARP1，CASP3，ABCG8，ABCG5，ICAM1
PCC	campesterol	HSD3B2，ABCG8，ABCG5，CYP7A1，DHCR24，CSN1S1，NR1H2
GF	Sudan III	CELA1，CELA3B
GF	quercetin	CYP1B1，SLC2A2，STK17B，CYP2C8，HCK，CYP1A1，MCL1，PIM1，HIBCH，ATP5B，ATP5F1B
GF	beta-sitosterol	SREBF1，CYP7A1，ABCB11，CASP3，ABCG5，APOE，ABCG8，SREBF2，DHCR24，ICAM1
GF	kaempferol	UGT1A9，UGT1A8，UGT3A1，UGT1A7，CYP1B1，CDK1，AHR，UGT1A3，NR1I2，RPS6KA3
GF	Stigmasterol	ABCG8，IL8，TNF，NR1H2，NR1H3，SREBF2，SLCO1B1，ABCG5，ABCA1，IL10

Table 2 The biological function of SR involved in BPSD

Function	Groups	Group Genes
positive regulation of smooth muscle cell proliferation	Group15	AKT1|APOE|BDNF|CASP3|CCL2|HMGB1|ICAM1|IL10|IL13|IL6|JUN|MAPK1|MAPK3|MMP9|MYC|PTGS2|SOD2|STAT1|TNF|
lipopolysaccharide-mediated signaling pathway	Group14	AKT1|CCL2|ICAM1|IL10|IL13|IL6|JUN|MAPK1|MAPK3|MMP9|MYC|PTGS2|SOD2|TNF|TNFSF11
regulation of neuroinflammatory response	Group13	AKT1|CASP3|HMGB1|IL10|IL6|MAPK1|MMP9|PTGS2|SOD2|TNF|VEGFA
regulation of nitric oxide biosynthetic process	Group12	AKT1|HMGB1|ICAM1|IL10|IL13|MAPK1|MAPK3|PTGS2|TNF|VEGFA
positive regulation of endothelial cell proliferation	Group11	AKT1|CASP3|HMGB1|ICAM1|IL10|IL6|JUN|MMP9|PTGS2|VEGFA
regulation of smooth muscle cell proliferation	Group10	AKT1|IL10|IL13|IL6|JUN|MMP9|MYC|PTGS2|SOD2|STAT1|TNF
positive regulation of nucleotide biosynthetic process	Group09	MYC|NOS1|PTGS2
positive chemotaxis	Group08	AKT1|CASP3|HMGB1|IL6|PTGS2|TNF|VEGFA
negative regulation of epithelial cell differentiation	Group07	ICAM1|IL13|MMP9|MYC|STAT1|VEGFA
regulation of endothelial cell proliferation	Group06	AKT1|APOE|CCL2|HMGB1|IL10|JUN|PTGS2|STAT1|TNF|VEGFA
negative regulation of cysteine-type endopeptidase activity involved in apoptotic process	Group05	AKT1|HMGB1|IL6|MMP9|PTGS2|TNF|VEGFA
positive regulation of blood vessel endothelial cell migration	Group04	AKT1|CASP3|HMGB1|IL6|PTGS2|VEGFA
cellular response to interleukin-6	Group03	ICAM1|IL6|STAT1
vasodilation	Group02	APOE|NOS1|SOD2
regulation of DNA-templated transcription, initiation	Group01	CCL2|HMGB1|JUN
regulation of interferon-alpha production	Group00	HMGB1|IL10|STAT1

Table 3 The biological function of CR on BPSD

Function	Groups	Group Genes
cellular response to cadmium ion	Group2	AKT1|HMOX1|MAPK1
response to nicotine	Group1	CASP3|HMOX1|MAPK1
negative regulation of macroautophagy	Group0	AKT1|HMOX1|TP53

Table 4 The biological function of PCC on BPSD

Function	Groups	Group Genes
response to nicotine	Group4	AKT1|APOE|CASP3|CASP9|HMOX1|ICAM1|IL10|MAPK1|SELP|TNF|TP53
glial cell apoptotic process	Group3	AKT1|APOE|CASP3|CASP9|HMOX1|TNF|TP53
plasma lipoprotein particle clearance	Group2	APOE|HMOX1|LDLR
regulation of platelet activation	Group1	APOE|F2|SELP
positive regulation of anion transport	Group0	APOE|CFTR|TNFSF11

Table 5 The biological function of GF on BPSD

Function	Groups	Group Genes
regulation of amyloid-beta formation	Group2	APOE|CASP3|TNF
regulation of membrane protein ectodomain proteolysis	Group1	APOE|IL10|TNF
regulation of nitric oxide biosynthetic process	Group0	ICAM1|IL10|TNF

Table 6 The docking results of the active ingredients with DRD2 and HTR2A

Name	DRD2 (PDB ID:6CM4) binding energy (Kcal/mol)	HTR2A (PDB ID:6A93) binding energy (Kcal/mol)
memantine	-6.72	-6.02
Risperdal	-7.71	-7.9
beta-sitosterol	-9.58	-8.2
stigmasterol	-9.17	-8.9
chelerythrine	-8.1	-8.12
campesterol	-7.37	-8.67
berberine	-7.87	-7.61
Oroxylin a	-7.38	-6.42
acacetin	-7.31	-5.8
Sudan III	-6.87	-6.95
baicalein	-6.7	-6.0
kaempferol	-6.33	-5.49
wogonin	-5.68	-5.95
quercetin	-5.82	-5.04
panicllin	-5.53	-5.74
coptisine	-	-
rutaecarpine	-	-
fumarine	-	-

Note: "-" indicates that the result has not been calculated.

Figures

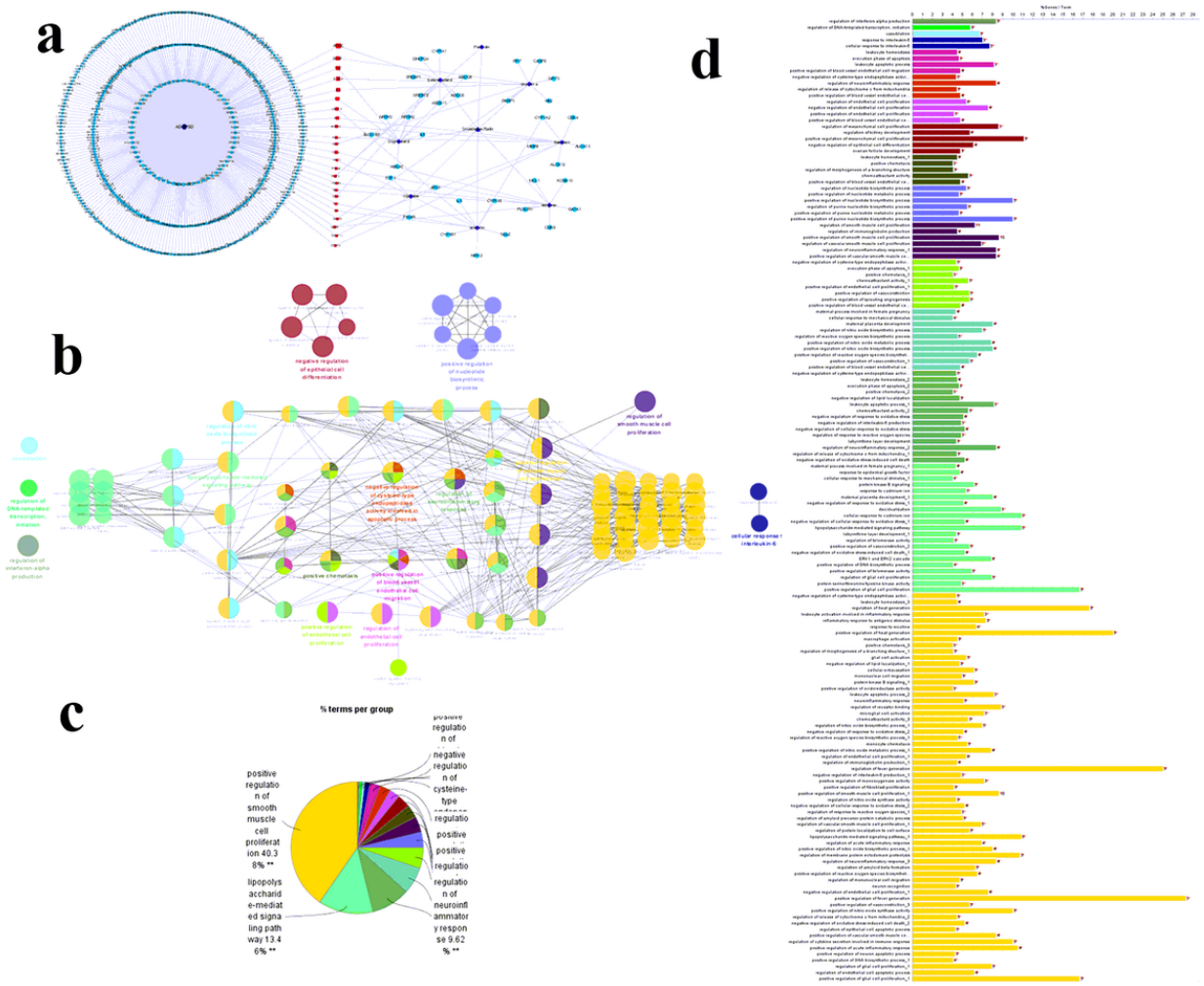

Figure 1

Schematic representation of SR's potential mechanism in treating BPSD. a The "compounds-targets-BPSD" network of SR. This systematic approach successfully revealed 24 key protein targets related to BPSD: APOE ((BPSD correlation score, D-CS): 156.68), TNF (D-CS: 114.77), SOD2 (D-CS: 114.23), IL 6 (D-CS: 105.12), BDNF (D-CS: 104.54), IL 10 (D-CS: 86.21), VEGFA (D-CS: 72.66), AKT1 (D-CS: 66.45), CCL2 (D-CS: 66.37), MAPK1 (D-CS: 52.1), PLAU (D-CS: 41.02), CASP3 (D-CS: 37.17), ICAM1 (D-CS: 37.08), PTGS2 (D-CS: 35.32), NOS1 (D-CS: 32.33), MMP9 (D-CS: 30.39), MYC (D-CS: 30.15), IL 13 (D-CS: 29.05), MAPK 3 (D-CS: 26.17), HMGB 1 (D-CS: 23.14), JUN (D-CS: 22.83), STAT1 (D-CS: 20.14), TNFSF 11 (D-CS: 18.98), CYP3A4 (D-CS: 17.31). b, c, d GO biological processes analysis of SR. SR's key protein targets involved 156 biological processes, like positive regulation of smooth muscle cell proliferation, lipopolysaccharide-mediated signaling pathway, and neuroinflammatory response.

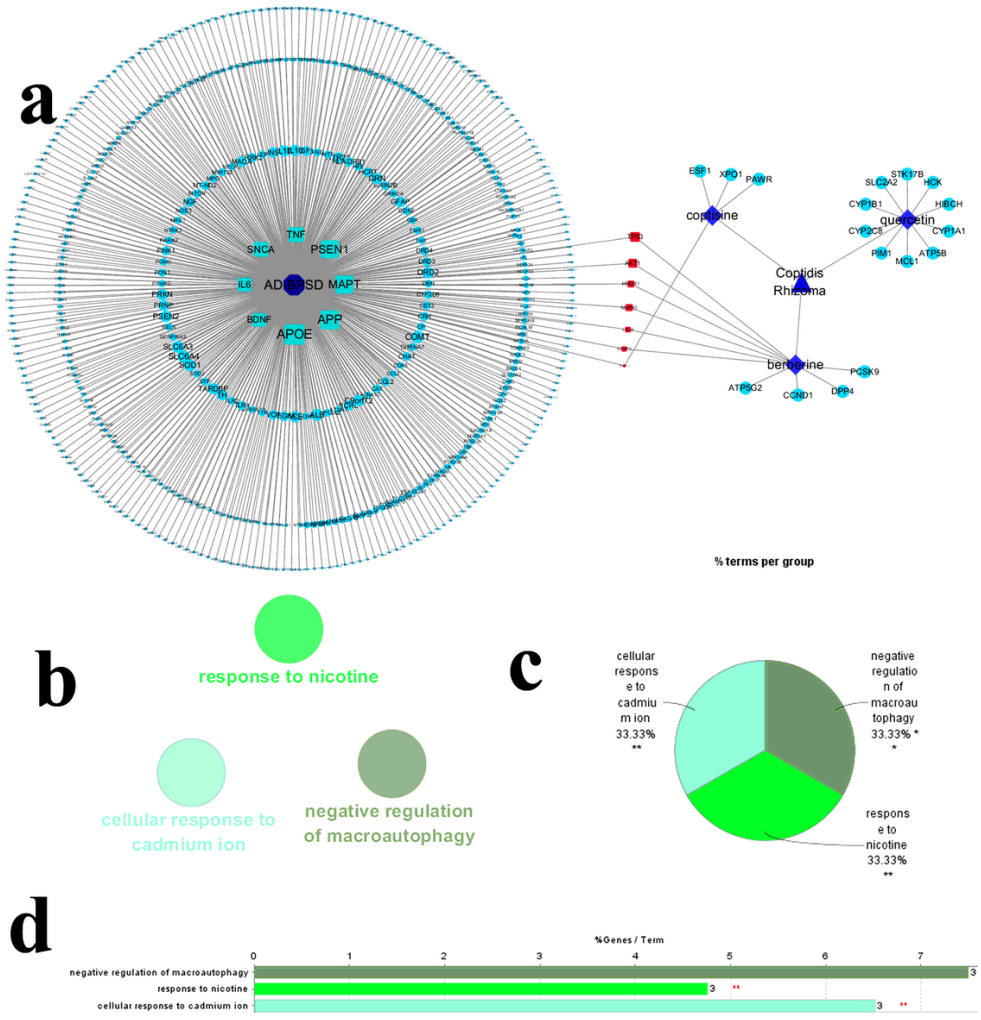

Figure 2

Schematic representation of CR's potential mechanism in treating BPSD. a The "compounds-targets-BPSD" network of CR. It revealed that seven key protein targets were related to BPSD: TP53 (D-CS: 72.85), AKT 1 (D-CS: 66.45), HMOX 1 (D-CS: 50.08), LDLR (D-CS: 40.09), MAPK 1 (D-CS: 52.1), CASP 3 (D-CS: 37.17). TNFSF 11 (D-CS: 18.98). b, c, d GO biological processes analysis of CR. The key protein targets of CR involved three biological processes they are negative regulation of macroautophagy, response to nicotine, and cellular response to cadmium ion.

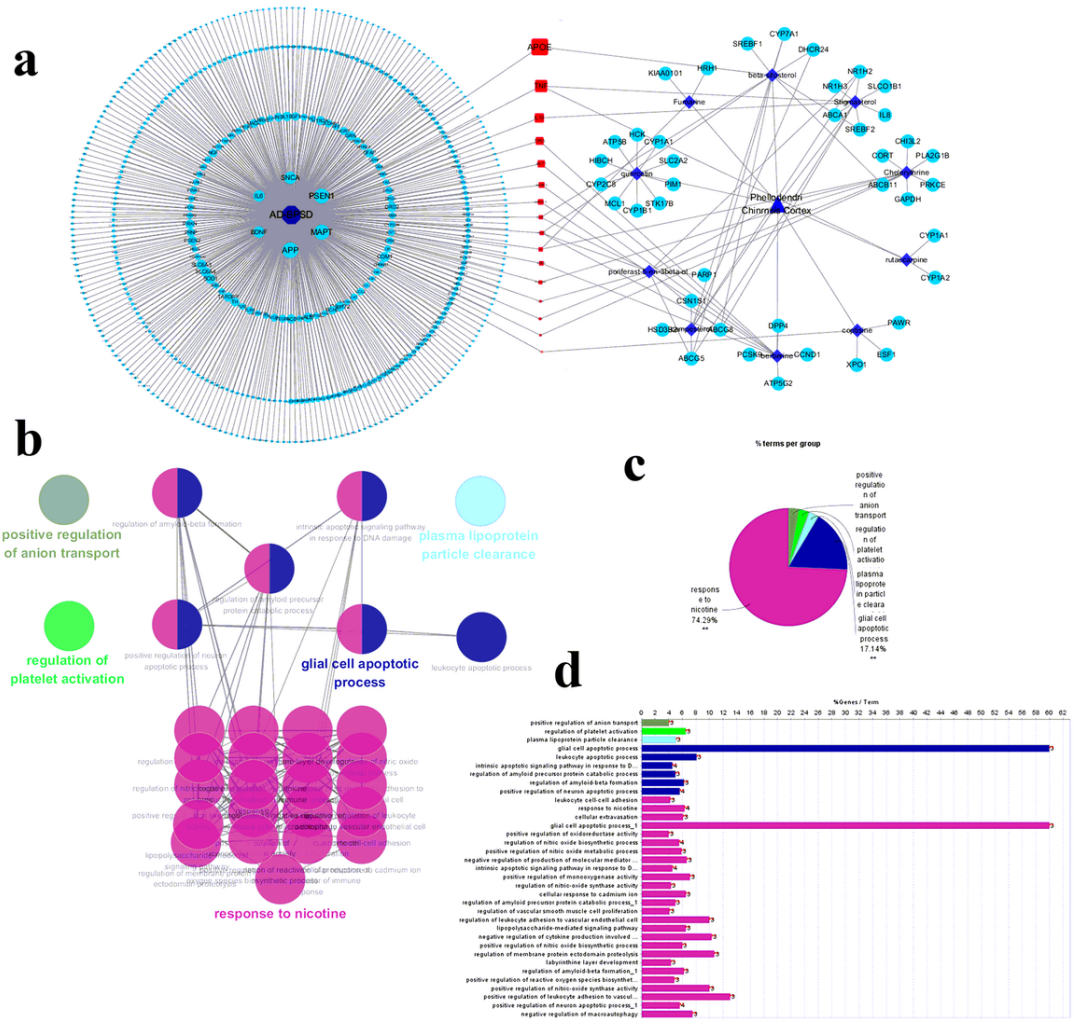

Figure 3

Schematic representation of PCC's potential mechanism of BPSD. a The "compounds-targets-BPSD" network of PCC. It revealed that 15 key protein targets were involved in BPSD: APOE (D-CS: 156.68), TNF (D-CS: 114.77), IL 10 (D-CS: 86.21), TP 53 (D-CS: 72.85), AKT 1 (D-CS: 66.45), MAPK 1 (D-CS: 52.1), HMOX 1 (D-CS: 50.08), LDLR (D-CS: 40.09), F 2 (D-CS: 38.83), CASP 3 (D-CS: 37.17), ICAM 1 (D-CS: 37.08), PLA2G2A (D-CS: 33.98), CFTR (D-CS: 21.04), CASP 9 (D-CS: 20.76), TNFSF 11 (D-CS: 18.98), SELP (D-CS: 17.65). b, c, d GO biological process analysis of PCC. PCC's key protein targets involved 35 biological processes like a response to nicotine, glial cell apoptotic process, and positive regulation of anion transport.

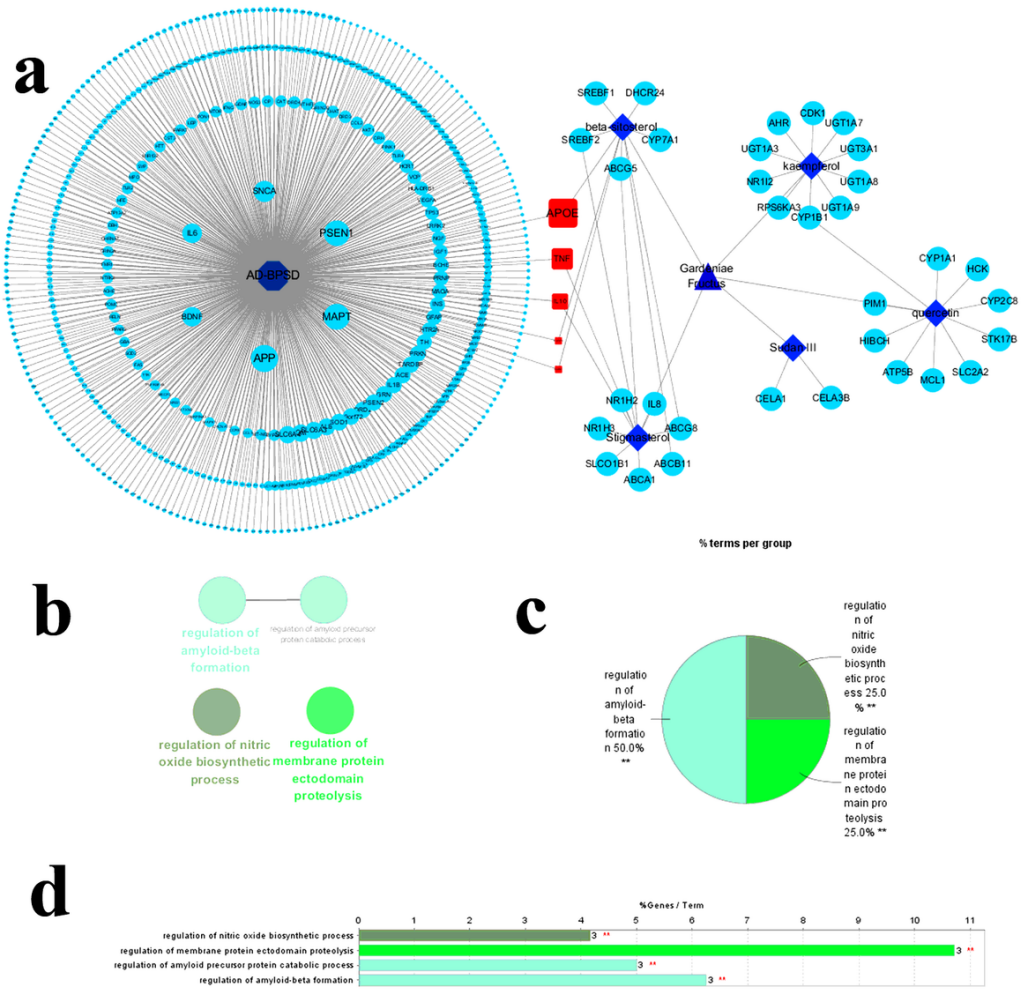

Figure 4

Schematic representation of GF's potential mechanism involved in BPSD. a The "compounds-targets-BPSD" network of GF. It revealed that five key protein targets were sifted out. They are APOE (D-CS: 156.68), TNF (D-CS: 114.77), IL 10 (D-CS: 86.21), CASP 3 (D-CS: 37.17), ICAM 1 (D-CS: 37.08). b, c, d GO biological processes analysis of GF. GF's key protein targets involved in 4 biological processes, they are the regulation of amyloid precursor protein catabolic process, regulation of membrane protein ectodomain proteolysis, regulation of nitric oxide biosynthetic process, and regulation of amyloid-beta formation.

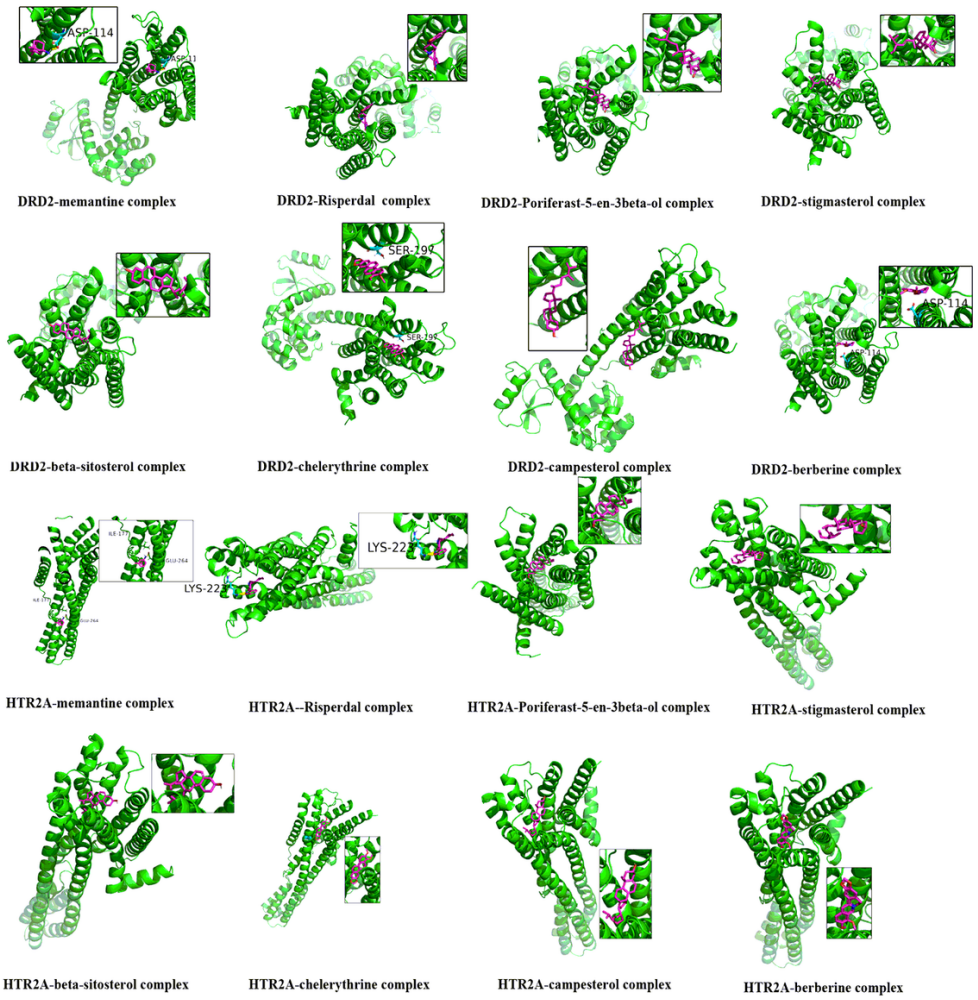

Figure 5

The Schematic 3D representation of the molecular docking model with DRD2 and HTR2A.