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Using network pharmacology analysis and molecular docking of quercetin compound for treatment of Alzheimer

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ABSTRACT

Alzheimer's disease is currently increasing in risk with age. This study investigates the inhibitory activity of Quercetin (QC) compounds in slowing the progression of Alzheimer's disease (AD). The research employs network pharmacology and molecular docking methods. Furthermore, it conducts screening of important herbal medicines from traditional Chinese medicine and integrates them with the GeneCards database and AD-related targets. Overlapping herbal medicines and targets have been identified as significant candidates. A total of 10 target genes have been selected for QC in AD treatment. The JUN gene shows the highest binding affinity. Gene Ontology (GO) analysis was performed to identify AD-related biological processes and neural cell components. Additionally, 10 candidate targets with homologous genes participating in signaling pathways have been identified. QC binding molecules exhibit high binding affinity for 10 target proteins, elucidating candidate targets for QC in alleviating AD. The study explores protein-protein interactions and associated signaling pathways, confirming QC's inhibition of AD. This provides a basis for AD therapy monitoring.

Introduction

Recent years have seen a surge in interest in developing new active drugs, particularly small-molecular-weight pharmaceuticals that interact with biological targets. This approach offers a streamlined drug design but requires extensive research and expensive resources [1]. Only a small fraction of novel compounds receive regulatory approval. The process involves numerous preclinical and clinical experiments, evaluating the safety and effectiveness of new treatments [2]. Preclinical testing involves discovering potential drugs for improved models. Screening strategies involve identifying chemicals against biochemical or cellular targets, selecting suitable compounds, and examining physicochemical and pharmacological parameters. These compounds are assessed for lead compound potential, but screening data may have limitations [3]. Positive results can lead to further tests, while negative results may become less important. Computational methods can help overcome these limitations and improve the screening process. The fingerprinting approach is a highly effective tool for detecting new drugs and retrieving chemicals bound to protein targets [4].

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The risk of developing AD in older people is increasing. This is an age-dependent neuropathy, due to a neurodegenerative disorder that currently has no cure. This is also a phenomenon of the gradual decline in the activity of the nerve cells. The nerve cells die gradually leading to cognitive impairment [5]. Alzheimer's disease, initially identified by a German psychiatrist and neurologist, is a type of dementia that has been acknowledged as the primary clinical manifestation in individuals, resulting in alterations in daily routines and a dependence on their caregivers [6]. The common symptoms of dementia are caused by the progression of AD in the elderly. Nowadays, AD is prevalent in underdeveloped countries, accounting for almost 50% of people with AD worldwide. There are many different paths to slow down the risk of AD progression and may also relieve symptoms. Alzheimer disease is caused by a peptide containing 36-43 amino acids due to the distribution of amyloid-beta (Aβ). This may be the main cause of neurodegeneration. Amyloid-beta is created by the chain division of the amynoid precursor protein. (APP) [7]. Caspases-3 is essential for biological functions like apoptosis and pyroptosis, producing cytokines and reducing nerve death. It plays a central role in cell differentiation, limiting the rate of APP breakdown, leading to toxic neuronoid proteins and neurodegeneration [8]. Forecast strong Caspase-3 inhibitors as potential treatment options for Alzheimer disease management. Caspase-3 is present in humans and is essential for the development of nerve cells. Therefore, inhibiting Caspase-3 represents a beneficial target to slow down or prevent AD. Over time, various QSAR models have been constructed to develop Caspase-3 inhibitors [9]. Heart failure and Alzheimer's disease are prevalent in older adults, with heart failure patients at risk of developing AD. Risk factors include age, vitamin D deficiency, kidney disease, and diabetes. Quercetin can detect potential AD prevention [10,11].

Numerous studies have explored the link between cardiovascular disease and Alzheimer's disease, with clinical trials and animal studies proving the efficacy of incorporating Jin Hong Tang into the diet for treating Alzheimer's disease. However, the scientific paper only provides preliminary reports. This study aims to study the molecular mechanisms behind the treatment of AD using network pharmacology, gene analysis, and molecule connectivity.

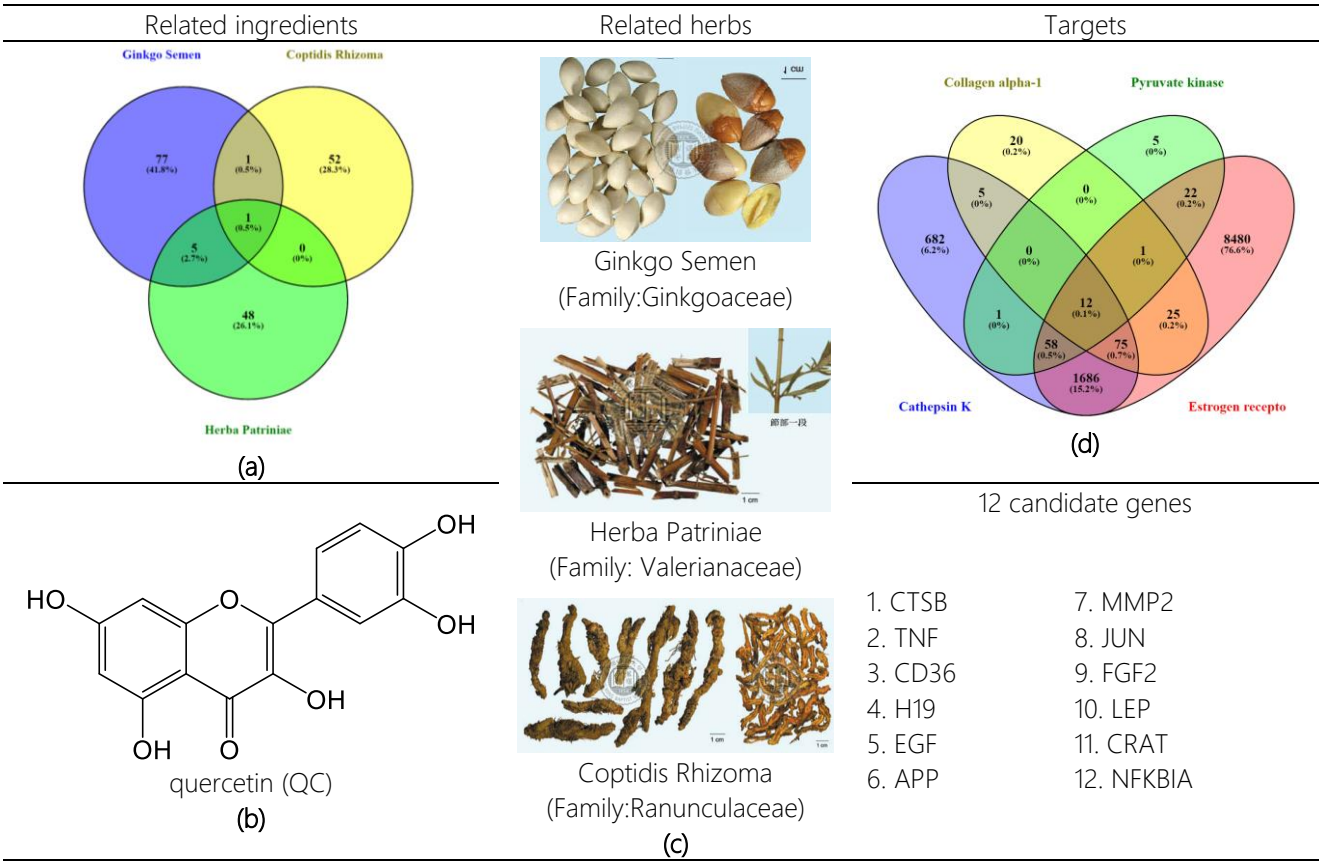

Fig 1: (a) Venn diagram of related ingredients to AD; (b) Quercetin in an intersection of three herbs; (c) Three related herbs: Ginkgo Semen, Herba Patriniae, Coptidis Rhizoma; (d) The related targets: Cathepsin K, Collagen alpha-1, Pyruvate kinase and Estrogen receptor, and 12 candidate genes in an intersection.

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Materials and Methods

The Targets Associated with QC and AD

Figure 1a describes the chemical structure of quercetin (QC) compounds obtained from PubChem [12]. We searched the genetic databases (NCBI), GeneCards, and DisGeNet using the search terms "Alzheimer" and "Homo sapiens" to identify the genes associated with AD [13]. GeneCards are used to find the QC target protein. As shown in Figure 1b, we then use the Venny chart to identify potential QC targets for AD reduction. Cathepsin K: 2519 genes, Collagen alpha-1: 138 genes; Pyruvate kinase: 99 genes. Estrogen receptor: 10359 genes. To analyze the protein interaction network of candidate targets, the species was set to Homo sapiens and the confidence score was set to greater than 0.7.

Enrichment Analysis of the Candidate Targets

The aforementioned candidate targets underwent gene ontology (GO) functional enrichment analysis and concomitant genomic and metabolic pathway (KEGG) enrichment analysis [14]. The species preference was set to Homo sapiens, and cellular components, molecular functions, biological processes, and signaling pathways associated with the targets were analyzed. The enrichment diagrams were created using the bioinformatics website.

Docking calculation

Autodock can be used to evaluate the binding efficiency between QC and candidate target proteins. The RCSB Protein Data Bank (http://www.pdb.org) was used to determine the X-ray crystal structures of the predicted targets. ChemDraw also allows for the reconstruction of QC 3D structure. Then, edit candidate target proteins located at the intersection of QC and AD using MOE Autodock [15]. The study applied structural modifications to candidate proteins, evaluated the connection between QC and targets using MOE Autodock, and used RMSD docking models for accurate results. Protein-ligand interactions were visualized using MOE.

Results and discussion

Identify the Candidate Targets

As illustrated in Fig. 1b, after removing redundant information, we obtained AD-related therapeutic targets including cathepsin K: 2519 genes, collagen alpha-1: 138 genes, pyruvate kinase: 99 genes, estrogen receptor: 10359 genes, of which 12 genes were overlapped between 4 targets. Human (Homo sapiens) candidate genes are located within the junction region including CTSB, TNF, CD36, H19, EGF, APP, MMP2, JUN, FGF2, LEP, CRAT, and NFKBIA intersection. We selected one common candidate JUN gene with largest interacting partners [13]. These genes are potential molecular targets that mediate the anti-AD effects of QC.

Proteins-Protein Interaction

A protein-protein interaction network was made using a candidate JUN gene of four candidate protein targets cathepsin K, collagen alpha-1, pyruvate kinase, and estrogen receptor. The candidate genes CTSB, TNF, CD36, H19, EGF, APP, MMP2, JUN, FGF2, LEP, CRAT, and NFKBIA [13]. The 12 target genes are described in detail in Fig. 1. The interaction of the 12 candidate protein genes CTSB, TNF, CD36, H19, EGF, APP, MMP2, JUN, FGF2, LEP, CRAT, and NFKBIA with other protein genes is depicted in Figure 2a. There are a number of target protein loci with the greatest number of protein interactions. We discovered that the JUN protein gene with the most interactions may be the best candidate for Alzheimer therapy. Figures 2b and 2c suggest a JUN target intersecting with four target groups as a potential anti-AD effect of QC in the protein-protein interaction network. The interactions between JUN protein subunits have 26 nodes, 166 edges, and an average node degree of 12.8, with nodes with the highest degree of connectivity to other gene symbols representing the most promising drug targets. These targets are also associated with other proteins at various confidence probabilities (p). Figure 2d shows the distribution of similarity proteins, with 12 candidate genes being selected as potential proteins. Gene JUN is crucial for the covalent propagation of immunoglobulins and immune complexes, enhancing the solubility of immune aggregates, and may be responsible for efficient binding to form amide bonds with immune aggregates.

Enrichment Analysis of the Candidate Targets

GO analysis

GO analysis to identify biological processes and pathways with a significance level of 0.05. The histogram columns represent the number of enriched genes, while the lesser P-values indicate a higher level of confidence in the enrichment results. They are functionally molecularly enhanced in gene ontology (GO) by binding to the transcriptional core regulator, DNA-binding transcription factors, RNA polymerase II, etc., and upregulatory small molecule metabolism. The

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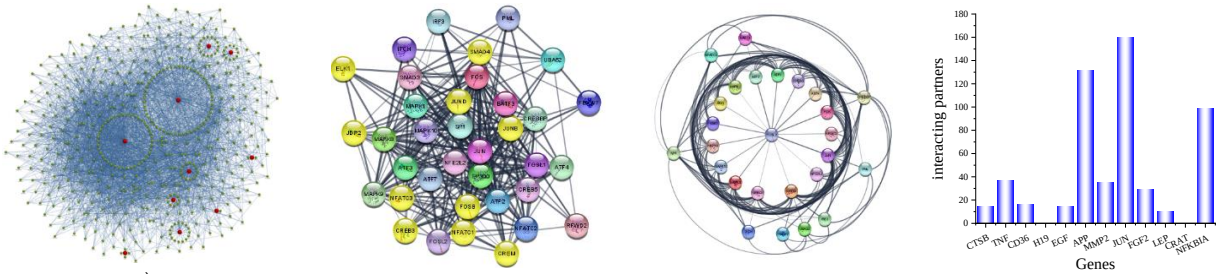
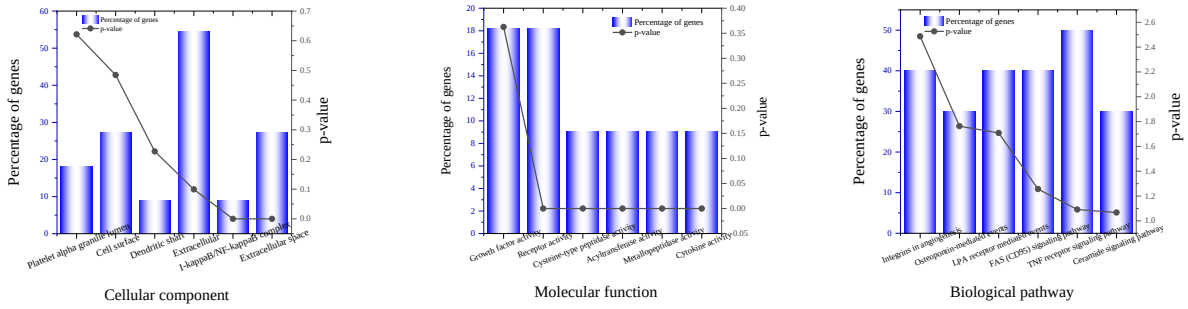
GO analysis graph in Fig. 3 clarifies the relationship between each target, cellular component, biological process, molecular function, and biological pathway in ascending P-value order. The study focuses on the cellular components, biological processes, molecular functions, and biological pathways of 12 genes, including CTSB, TNF, CD36, H19, EGF, APP, MMP2, JUN, FGF2, LEP, CRAT, and NFKBIA [13,14].



a) b) c) d)

Fig 2: Analysis of network pharmacology presents in a) importance of 12 candidate targets in protein-protein interaction (PPI) network; b) and c) protein-protein interaction network: 26 nodes (proteins) and 166 edges (protein interactions); d) the interacting partners.



a) b) c)

Fig 3: GO analysis presents in a) cellular component, b) molecular function and c) biological pathway of the 11 target proteins at the p confidence level.

The study reveals that JUN's high binding center is located at the intersection of four targets, with the anti-AD effect being the primary binding center. Three key procedures are positive transcription regulation from the RNA polymerase II promoter, DNA template regulation, and negative transcription regulation. Molecular function refers to responses to factors like polysaccharide, glucocorticoids, antibiotics, hydrogen peroxide, blood pressure regulation, and aging cells linked to Alzheimer's disease. Six cellular components, including Platelet alpha granule lumen, cell surface, dendritic shaft, extracellular, I-kappaB/NF-kappaB complex, and extracellular space, play a role in the anti-AD effect of QC. The GO analysis reveals that positive transcriptional regulation from the RNA polymerase II promoter has the most biological targets, while protein binding has the most molecular function targets, with two transcriptional variants. These protein genes can be expressed as a single chain precursor.

KEGG pathway analysis

The study used a KEGG pathway enrichment analysis to identify potential pathways involved in the anti-AD effects of QC. It found that 11 interferon targets are primarily enriched in signaling pathways, suggesting interaction with multiple sequenced pathways [14]. The KEGG analysis pathway diagram in Fig. 4 shows the significant role of these pathways in Alzheimer treatment, with the graph indicating the relationship between target enrichment and leading pathways. [13,14]. The study reveals that the highlighted genes in fluid shear stress, atherosclerosis, and hypoxia-inducible pathways are part of the PPI network of common targets, suggesting they may mediate QC's anti-AD effects. The JUN gene also interacts with these pathways, as depicted in Fig 4.

Alzheimer's disease is a severe condition characterized by gene inactivation, disrupting signal transduction pathways under stress and normal conditions. Symptoms include cognitive impairment, tremors, speech difficulties, delayed reflexes, coordination

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deficits, and nerve tremors. Early neurological disorders are also observed in AD patients. The study also highlights the involvement of ATM mRNA in various cellular processes beyond DNA repair [13]. Furthermore, The JUN gene plays a crucial role in nerve cell receptor regulation, which triggers cytokine production, cell survival, development, and differentiation, as in Fig 4. CD28, CD45, and CD4 regulate TCR signals, enhancing IL-2 production and preventing cell dysfunction. TCR activation also prompts bone cell rearrangement via Rac and PAK [14]. Negative regulation of TCR nerve signaling is essential to prevent excessive response activation, with substances like SIT and CTLA4 acting as negative regulatory substances. The body uses natural mechanisms to regulate T-cell nerve pathways post-activation to prevent uncontrolled reactions.

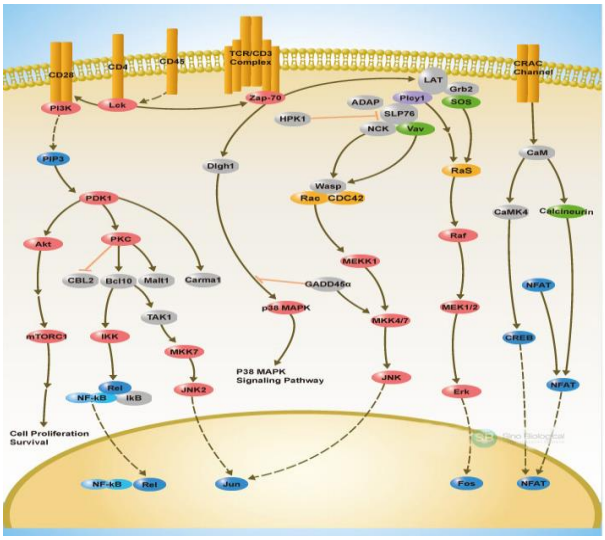


Fig. 4: Biological pathways presented for JUN Gene

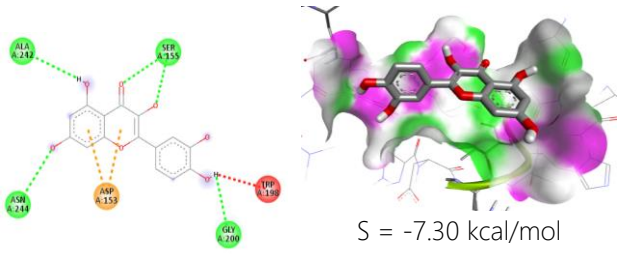
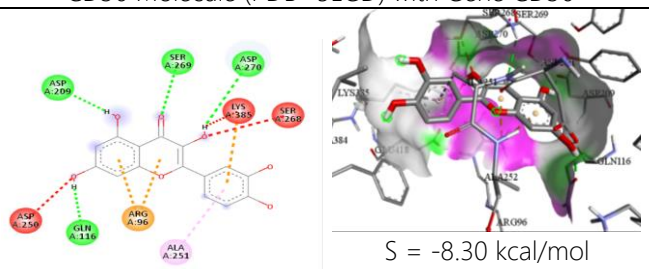
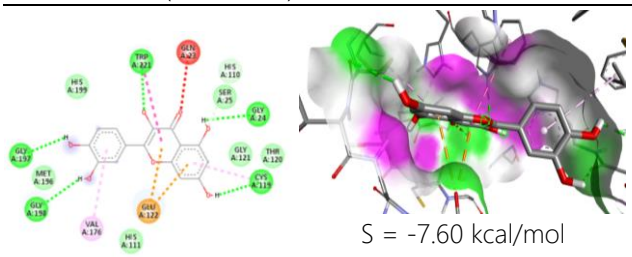
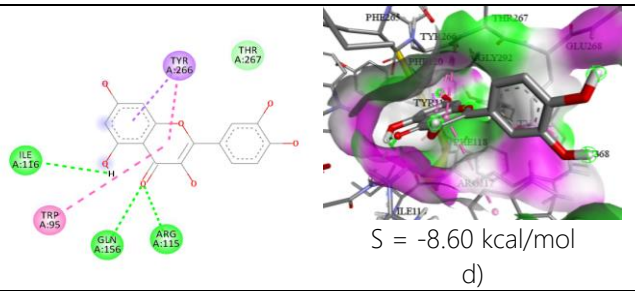
Interaction 2D,3D and Score (S)/ kcal/mol	Interaction 2D, 3D and Score (S)/ kcal/mol
Cathepsin B (PDB-1CPJ) with Gene CTSB	CD36 Molecule (PDB- 5LGD) with Gene CD36
 S = -7.30 kcal/mol a)	 S = -8.30 kcal/mol b)
Jun Proto-Oncogene, AP-1 Transcription Factor Subunit (PDB-3U85) with Gene JUN	Leptin (PDB-6E2P) with Gene LEP
 S = -7.60 kcal/mol c)	 S = -8.60 kcal/mol d)

Fig. 5: The docking results are illustrated for QC bonds to the most important target proteins:
a) PDB-1CPJ complex; b) PDB- 5LGD complex; c) PDB-3U85; d) PDB-6E2P

Docking calculation

The study analyzed the interaction of QC protein with specific docking targets, including the critical JUN gene target in QC's anti-Alzheimer's effect. The docking targets included CTSB, TNF, CD36, EGF, APP, MMP2, JUN, FGF2, LEP, and NFKBIA genes, with ten having partners; H19 and CRAT lacked partners. These targets evaluated QC binding, with Fig.5 showing QC's predominant bonding via hydrogen bridges and hydrophobic interactions [11]. Docking data revealed QC mainly binds through pi-H interaction, hydrophobicity, and hydrogen bonding. Lower affinity energy indicates strong binding, with most proteins having binding energies below -5 kcal/mol, suggesting significant bindings, and genes with the lowest binding energy suggesting the highest binding capacity [15]. Overall, the docking process accurately predicted QC's interaction with target protein binding pockets. Additionally, QC's effects on Alzheimer's disease were confirmed. The binding outcomes of the QC-H group resembled those of other target protein interactions,

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and QC therapy may vary in Alzheimer's treatment, with possible variations based on treatment duration.
Using computational docking techniques for QC interactions with the target proteins Cathepsin B (PDB-1CPJ) with the gene CTSB, CD36 Molecule (PDB-5LGD) with the gene CD36, (PDB-3U85) with the gene JUN, and Leptin (PDB-6E2P) with the gene LEP further demonstrated that these are core important genes in the PPI network, as seen in Fig. 2. The results of the PPI interaction screening using the network pharmacology obtained from this study align with the research on the mechanism of quercetin therapeutic targets for Alzheimer's disease, as indicated by the work of Guoxiu Zu *et al*. [16]. The binding energies (kcal/mol) between QC and the proteins were all less than -7.0 kcal/mol, as confirmed by molecular docking simulations. However, the JUN gene appears to be the most important, with multiple links between QC and the target protein. Four core genes were validated in the docking simulation (Fig. 5). The binding energies of QC with the proteins ranged from -8.60 kcal/mol to -7.30 kcal/mol. The average binding energy was -7.95 kcal/mol. Biological pathways presented for the JUN gene were also identified, as shown in Fig. 4.

Discussion

QC, a natural or extracted derivative, possesses anti-inflammatory, anticancer, and antidiabetic properties. Late modulation of neurogenesis in Alzheimer's disease (AD) [11]. New extraction methods may ensure sustainable QC supply, opening up promising potential for developing AD drugs. This study explores network pharmacology, analyzing interactions between proteins and chemical components to discover standalone medications. Using network pharmacology techniques, key targets for AD treatment using QC derivatives have been identified. Target networks have revealed eleven potential candidates for QC use in AD treatment, with eleven proteins identified as crucial molecular targets for QC's anti-AD effects. Among them, the JUN protein gene target has been prominently selected for molecular docking, complemented by KEGG analysis. Gene Ontology analysis has highlighted QC's involvement in AD-related biological processes such as aging and nerve influence. Recent research emphasizes the link between AD and age-related increase, reinforcing the importance of targeting these pathways in AD treatment. Cytokines contribute to Alzheimer's disease (AD), causing loss of control over habits. Aging accelerates AD, and Quality Control (QC) can regulate protein targets and modulate pathological processes. Molecular studies show QC interacts with predicted protein targets related to AD, with all four selected proteins being targets in Alzheimer's disease [12,13]. Additionally, the results indicate that AD interaction and hydrogen bonding are the predominant forms of interaction, suggesting the molecular mechanism underlying QC in AD. Future research may investigate specific techniques to yield more precise results.

Conclusion

The study focuses on screening herbal materials and analyzing major anti-Alzheimer's disease targets using network pharmacology. The research findings reveal that QC may alleviate AD through multiple targets and pathways, suggesting potential for AD treatment and providing a mechanism of action for monitoring AD therapy. The study provides a comprehensive understanding of its therapeutic effects. This study serves as a reference for validation assays of QC based on these protein candidate targets.

References

1. Lalitkumar K. Vora, Amol D. Gholap, Keshava Jetha, Raghu Raj Singh Thakur, Hetvi K. Solanki, and Vivek P. Chavda, Pharmaceutics. 15(7) (2023) 1916. https://doi.org/10.3390/pharmaceutics15071916
2. Joseph G. Lombardino and John A. Lowe, Nature Reviews Drug Discovery, 3 (2024) 853–862. https://doi.org/10.1038/nrd1523
3. J.P. Hughes, S. Rees, S.B. Kalindjian, and K.L. Philpott, Br J Pharmacol. 162(6) (2011) 1239–1249. https://doi.org/10.1111/j.1476-5381.2010.01127.x
4. A.I. Visan, and I. Negut, Life 14 (2024) 233. https://doi.org/10.3390/life14020233
5. Y. Hou, X. Dan, M. Babbar, Y. Wei, S.G. Hasselbalch, D.L. Croteau, V.A. Bohr, Nat. Rev. Neurol, 15(10) (2019) 565–581. https://doi.org/10.1038/s41582-019-0244-7
6. H. Hippius, G. Neundörfer, Dialogues Clin Neurosci.; 5(1) (2003) 101–108. https://doi.org/10.31887/DCNS.2003.5.1/hhippius
7. P. Carrillo-Mora, R. Luna, and L. Colín-Barenque, Oxid. Med. Cell Longev 2014 (2014) 795375. https://doi.org/10.1155/2014/795375
8. N. Papadopoulos, N. Suelves, F. Perrin, D.M. Vadukul, C. Vrancx, S.N. Constantinescu, and P. Kienlen-Campard, Biomedicines, 10(11) (2022) 2753. https://doi.org/10.3390/biomedicines10112753
9. J.H. Su, M. Zhao, A.J. Anderson, A. Srinivasan, C.W. Cotman, Brain Res. 898(2) (2001) 350–7. https://doi.org/10.1016/s0006-8993(01)02018-2

10. Y. Arega and Y. Shao, Front Genet 13 (2022) 1015674. https://doi.org/10.3389/fgene.2022.1015674
11. H. Khan, H. Ullah, M. Aschner, W.S. Cheang, and E.K. Akkol, Biomolecules 10(1) (2020) 59. https://doi.org/10.3390/biom10010059
12. The National Center for Biotechnology Information advances science and health by providing access to biomedical and genomic information: https://www.ncbi.nlm.nih.gov/
13. https://www.disgenet.org/dbinfo
14. L. Chen, C. Chu, J. Lu, X. Kong, T. Huang, and Y.-D. Cai, PLoS One 10(5) (2015) e0126492. https://doi.org/10.1371/journal.pone.0126492
15. https://www.chemcomp.com/Products.htm
16. G. Zu, K. Sun, L. Li, X. Zu, T. Han, and H. Huang, Scientific Reports, 11(1) (2021) 22959. https://doi.org/10.1038/s41598-021-02248-5