MINISTRY OF EDUCATION AND SCIENCE OF UKRAINE
KYIV NATIONAL UNIVERSITY OF TECHNOLOGIES AND DESIGN
Faculty of Chemical and Biopharmaceutical Technologies
Department of Biotechnology, Leather and Fur

QUALIFICATION THESIS

on the topic **Study on the effective ingredients and mechanism of action of *Gardenia jasminoides* in the treatment of hyperuricemia**

First (Bachelor's) level of higher education

Specialty 162 "Biotechnology and Bioengineering"

Educational and professional program "Biotechnology"

Completed: student of group BEBT-20
Liyuan XU

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Educational and professional program Biotechnology

APPROVE
Head of Department of Biotechnology, Leather and Fur, Professor,
Doctor of Technical Science
Olena MOKROUSOVA
_____
«___»_____2024

ASSIGNMENTS
FOR THE QUALIFICATION THESIS
Liyuan XU

1. Thesis topic **Study on the effective ingredients and mechanism of action of *Gardenia jasminoides* in the treatment of hyperuricemia**
Scientific supervisor Olga Iungin, Ph.D., Assoc. Prof.

approved by the order of KNUTD "____"_____2024, №____

2. Initial data for work: assignments for qualification thesis, scientific literature on the topic of qualification thesis, materials of Pre-graduation practice

3. Content of the thesis (list of questions to be developed): literature review; object, purpose, and methods of the study; experimental part; conclusions

4. Date of issuance of the assignments_____

EXECUTION SCHEDULE

№	The name of the stages of the qualification thesis	Terms of performance of stage	Note on performance
1	Introduction	From 01 April 2024 to 11 April 2024	
2	Chapter 1. Literature review	From 06 April 2024 to 20 April 2024	
3	Chapter 2. Object, purpose, and methods of the study	From 21 April 2024 to 30 April 2024	
4	Chapter 3. Experimental part	From 01 May 2024 to 10 May 2024	
5	Conclusions	From 07 May 2024 to 12 May 2024	
6	Draw up a bachelor's thesis (final version)	From 20 May 2024 to 25 May 2024	
7	Submission of qualification work to the supervisor for feedback	From 25 May 2024 to 27 May 2024	
8	Submission of bachelor's thesis to the department for review (14 days before the defense)	27 May 2024	
9	Checking the bachelor's thesis for signs of plagiarism (10 days before the defense)	31 May 2024	
10	Submission of bachelor's thesis for approval by the head of the department (from 7 days before the defense)	03 June 2024	

I am familiar with the task:

Student _____ Liyuan XU

Scientific supervisor _____ Olga IUNGIN

SUMMARY

Liyuan Xu. Study on the effective ingredients and mechanism of action of Gardenia jasminoides in the treatment of hyperuricemia – Manuscript.

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Based on network pharmacology methods and molecular docking techniques, the mechanism of action of Gardenia jasminoides in treating hyperuricemia (HUA) was analyzed. Using the Traditional Chinese Medicine Systems Pharmacology Database and Analysis Platform (TCMSP), a total of 15 active ingredients of Gardenia jasminoides were predicted. The target genes were predicted and obtained through TCMSP and Uniprot databases, resulting in 369 target genes. Using OMIM, GeneCards, and DrugBank databases to search for HUA disease targets, 906 HUA-related pathogenic genes were obtained by merging deduplicated data, and a Venn diagram was drawn to find the relationship between the target of Gardenia jasminoides active ingredients and HUA-related genes, which is reflected by 48 common genes. Protein-Protein Interaction Networks (PPI) for these 48 shared genes were drawn using the String database platform, visual optimization and core gene mining were performed, and core genes such as TP53 and CASP3 were obtained. Relevant pathways of HUA were obtained through the DAVID database, and Cytoscape 3.10.1 software was used to draw the drug-component-target-pathway-disease network diagram to more intuitively show that quercetin, kaempferol, and β-sitosterol in Gardenia jasminoides are the three most effective components for treating hyperuricemia. GO enrichment and KEGG enrichment analyses were conducted using the Metascape platform and microbiota platform, showing 2889 GO enrichment analysis pathways in biological process (GO-BP), 152 in cellular component (GO-CC), and 236 molecular function (GO-MF)-related processes. KEGG enrichment analysis genes were enriched and significant in lipid and atherosclerosis pathways, diabetes complications pathway, AGE-RAGE pathway, etc. Molecular docking ligands and receptors were retrieved from TCMSP and RCSB

PDB databases, and molecular docking was performed on the CB-DOCK2 online molecular docking platform. The results showed that quercetin, kaempferol, and β-sitosterol had good spatial binding ability with TP53 and CASP3, further verifying the binding degree of effective ingredients to core targets.

Keywords: Hyperuricemia; Gardenia; Network pharmacology; Molecular docking technology; Component analysis

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INTRODUCTION

Hyperuricemia (HUA) is a common metabolic disease. In recent years, the incidence rate has increased year by year, which has become an important problem threatening human health. There are about 177 million hyperuricemia patients in China, of which nearly 60% are young people aged 18 to 35. The overall incidence rate of gout is about 1.1%, and the number of patients is about 14.66 million. Hyperuricemia not only causes gout, but also is related to metabolic syndrome, hypertension, diabetes, obesity and other diseases. At the same time, hyperuricemia patients also have an increased risk of kidney disease.

Traditional Chinese medicine treatment has also achieved certain therapeutic effects in the treatment of hyperuricemia. Traditional Chinese medicine believes that hyperuricemia belongs to the categories of "bi syndrome" and "gout", and its treatment mainly focuses on clearing heat and detoxifying, promoting dampness and acid excretion, and promoting blood circulation and removing blood stasis.

On the basis of the same origin of medicine and food, traditional Chinese medicine has many advantages in treating hyperuricemia, such as natural ingredients, low side effects, long-lasting efficacy, and low price.

However, the lack of systematic support in traditional Chinese medicine theory hinders its dissemination and promotion.

Exploring the substances contained in traditional Chinese medicine has always been an important part of aligning traditional Chinese medicine with international standards.

In order to explore the mechanism of Gardenia jasminoides in treating hyperuricemia, we need to know the components and functions of Gardenia jasminoides, the mechanism of action of its components, and how to treat hyperuricemia, among many other issues

In order to systematically explore the drug mechanism of Gardenia jasminoides, this work combines network pharmacology and molecular docking methods to conduct research on this.

CHAPTER 1
LITERATURE REVIEW

Hyperuricemia (HUA) is a metabolic disorder characterized by increased purine metabolism, increased blood uric acid levels, and deposition of monosodium urate crystals in joints. HUA patients may experience a series of joint diseases such as gouty arthritis, interstitial nephritis, and gouty stones. Research has shown that the number of HUA patients is increasing year by year and tends to be younger [1-3]. At the same time, the "National Health Commission: Guidelines for Adult Hyperuricemia and Gout Diet" points out that the incidence of hyperuricemia among adult residents in China is 14%, and the incidence of gout is 0.86% to 2.20%. Moreover, the incidence rate of male is higher than that of female, urban population is higher than that of rural population and coastal living population is higher than that of inland population. HUA is not only closely related to people's dietary habits, but also to other metabolic diseases [4]. Studies have found that obesity, overweight, and central obesity are risk factors for hyperuricemia, which is consistent with previous studies that have suggested that hyperuricemia is associated with obesity and metabolic disorders [5 6]. At present, the number of patients with hyperuricemia in China is about 177 million, accounting for about 13.3% of the total population. Among them, the proportion of patients aged 18 to 25, 26 to 35, and 36 to 45 is 22%, 38%, and 26%, respectively, posing a serious threat to public health. Based on this background, how to prevent and treat hyperuricemia has become one of the focuses of global attention.

Gout is one of the main manifestations of hyperuricemia, but the cause of gout is not yet clear. 1% of cases have been identified as congenital enzyme defects, mostly of two types:

(1) polygenic genetic defects that lead to impaired uric acid secretion in renal tubules, resulting in reduced uric acid excretion and further leading to the occurrence of hyperuricemia;

(2) Enzyme and metabolic defects, which are dominant inheritance of the X chromosome, such as increased activity of 1-pyrophosphate 5-ribopyrophosphate (PRPP) synthase and deficiency of hypoxanthine guanine phosphoribosyltransferase (HGPRT), both of which can increase purine synthesis and lead to increased uric acid production.

Traditionally, people control and correct uric acid levels by adopting a low calorie diet, avoiding high purine foods, and maintaining ideal weight. However, this method is not only difficult to maintain but also difficult to achieve good results in the short term. In terms of Western medicine treatment, it includes probenecid, sulfamethoxazole, benzbromarone, etc., which rely on drugs to inhibit the reabsorption process of uric acid in the proximal renal tubules, leading to uric acid excretion [7]. However, there are many shortcomings in the treatment of Western medicine, the main reason being its obvious side effects. Treatment with allopurine often results in symptoms such as rash, abdominal pain, and diarrhea [8], which cannot be used as a long-term treatment method.

At present, there is no specific treatment for gout and hyperuricemia. Although Western medicine can temporarily alleviate symptoms, the condition is prone to recurrence after discontinuation of medication, with many adverse reactions. The use of traditional Chinese medicine with fewer adverse reactions, or the combination of Chinese and Western medicine, has achieved certain effects in treating gout [9]. So the treatment plan of traditional Chinese medicine with the same source of medicine and food can not only meet people's actual nutritional needs in daily life, but also help to treat and prevent related diseases [10 11]. The concept of "medicine and food have the same origin" originates from the "Huainanzi Xiuwu Training". In ancient traditional Chinese medicine, there was a theory of "four qualities" and "five flavors" in the understanding of traditional Chinese medicine. When applied to food, it was believed that each type of food also had "four qualities" and "five flavors". The concept of "medicine and food are of the same origin" refers to "medicine and food are of the same origin, and medicine and food are of the same root". Through searching many traditional Chinese pharmaceutical classics such as the Compendium of Materia

Medica, Shennong Materia Medica Classic, and Huangdi Neijing, it was found that most of the Chinese medicinal materials included in them that help with physical and mental nourishment can be consumed normally, effectively verifying the traditional Chinese medicine concept of "medicine and food are of the same origin". Traditional Chinese medicine materials with the same origin of medicine and food have the characteristics of less toxic side effects and excellent therapeutic effects [12]. In addition, with the continuous development of China's social economy, the quality of people's daily life has improved. Health products such as medicinal and food homology have received widespread attention from all sectors of society. At the same time, medicinal and food homology is not only limited to the medical field for disease treatment, but also has broad development prospects in the field of food commodities. It can be said that the concept of medicinal and food homology is gradually becoming popular in social life.

Network pharmacology is a discipline that explores the process of disease occurrence and development from the perspectives of systems biology and biological network balance, and combines a holistic perspective of network balance in organisms to understand the interaction between drugs and the body, thereby guiding the development of new drug combinations [13]. Molecular Docking is a computer simulation technique used to predict the binding mode and affinity between a molecule (usually a drug molecule or ligand) and another molecule (usually a biomolecule such as a protein or nucleic acid). It plays a crucial role in drug design and molecular biology research [14]. In the 1990s, network pharmacology developed, and domestic scholars conducted a series of pioneering explorations in network pharmacology, breaking through the limitations of the "single target" reductionism research model [15]. Until March 2021, Li Shao et al. formulated the international standard for network pharmacology, "Guidelines for Evaluation Methods of Network Pharmacology" [16]. Traditional Chinese medicine usually contains multiple chemical components, and traditional pharmacological methods are difficult to fully reveal its complex mechanisms of action. Network pharmacology can systematically analyze the multi component and multi target action patterns of traditional Chinese medicine

by constructing a network model of component target disease, and reveal its overall pharmacological effects. Traditional Chinese medicine usually contains multiple active ingredients and has the characteristic of multi-target action. Network pharmacology can systematically analyze the overall network of action of these drug components, while molecular docking can simulate the interaction between each component and multiple targets separately, providing detailed binding patterns and energy information, helping to understand the multi-component and multi-target mechanisms of traditional Chinese medicine. In the process of new drug development, network pharmacology can help identify new drug targets and candidate drug molecules, and molecular docking is used to optimize the structure of these candidate molecules, improve their targeting and efficacy. Network pharmacology, through its systematic and comprehensive methods, provides new perspectives and tools for the research and development of traditional Chinese medicine, promotes the modernization and internationalization of traditional Chinese medicine[17], and improves the scientific research level and application value of traditional Chinese medicine.

Therefore, this study analyzed the active ingredients and targets of Gardenia jasminoides in the treatment of HUA through network pharmacology and molecular docking, providing scientific basis for the medicinal development of Gardenia jasminoides and more ideas for the prevention and treatment of HUA. Gardenia contains multiple chemical components, and the active ingredients in Gardenia can inhibit the activity of uric acid producing enzymes, thereby reducing the production of uric acid. By using network pharmacology methods, the active ingredients with therapeutic effects on HUA were screened out, and GO enrichment and KEGG enrichment analysis were conducted. Finally, molecular docking was used to perform spatial binding calculations between the main active ingredients screened out and the core target of action, and the mechanism of action of Gardenia jasminoides against HUA was ultimately determined.

CHAPTER 2
OBJECT, PURPOSE AND METHODS OF THE STUDY

Gardenia contains various chemical components, mainly including iridoids, flavonoids, organic acids, volatile oils, etc. Among them, iridoid compounds are one of the main active ingredients of Gardenia jasminoides, including geniposide, geniposide, and hydroisogeniposide. Flavonoids are also one of the important active ingredients of Gardenia jasminoides, including genitalin, quercetin, and kaempferol. Organic acid compounds mainly include chlorogenic acid, caffeic acid, para hydroxybenzoic acid, etc. Volatile oil compounds mainly include terpenes, alcohols, aldehydes, etc.

Using gardenia as a keyword in the Traditional Chinese Medicine System Pharmacology Analysis Platform Database (TCMSP), https://tcms-e.com）[18] To search and screen the effective ingredients and target names of Gardenia jasminoides, and obtain various indicators such as oral bioavailability, distribution, drug like properties, metabolism, and excretion of various effective ingredients of Gardenia jasminoides. The screening criteria are based on the ADME principle, and the screening content includes oral bioavailability (OB) and drug similarity (DL). The screening criteria are set as follows: oral bioavailability (OB) ≥ 30%, drug similarity (DL) ≥ 0.18, and the active ingredients of the drug are obtained. At the same time, the corresponding protein target, i.e. the protein ID, are found in the TCMSP data. Organize and statistically analyze the selected target proteins using Excel. Using the protein database UniProt（ https://www.uniprot.org/ ）Using "Homo Sapiens" as the analysis species, the gene composition of the active ingredient target protein, i.e. the target gene name (gene ID), was sequentially searched and found. During this period, Excel needs to be used for a large amount of database organization to prevent duplication and omissions.

Using the English keyword "Hyperuricemia" for hyperuricemia in the GeneCards database [19 20] （ https://genecards.weizmann.ac.il/v3/ ）Human Online Mendelian Genetic Platform (OMIM) [21 22] （ https://omim.org/ ）Conduct a search to obtain pathogenic genes related to HUA. Due to differences between databases, the combination of the two databases can obtain the most comprehensive disease gene data. The results were merged and then reprocessed using Excel to obtain effective disease targets. Through the Draw Venn Diagram website [23] （ http://bioinformatics.psb.ugent.be/webtools/Venn/ ）The method of drawing a Venn diagram links the corresponding targets of the active ingredients of Gardenia jasminoides with the pathogenic genes of HUA, which can achieve the acquisition of common target genes for diseases and drugs.

Subsequently, a protein-protein interaction (PPI) network was constructed for the common target genes of diseases and drugs, which was published on the STRING website [24] （ https://string-db.org/ ）Input the disease drug common target genes obtained in the previous step, and use "Homo sapiens" as the species condition to obtain the interaction relationship between the common target genes. Subsequently, import Cytoscape 3.10.1 software [25] for visualization operations and optimize the graphics of the PPI network diagram.

Digging the disease related pathways of HUA from the David database and selecting the top 20 pathways for subsequent analysis on the "Micro Bioinformatics" website （ http://www.bioinformatics.com.cn/ ）The GO and Pathway analysis module will perform GO enrichment analysis [26] and KEGG enrichment analysis [27] on the intersection genes of Gardenia jasminoides and hyperuricemia, obtaining GO enrichment information related to molecular function, biological process, and cellular component, as well as KEGG enrichment information of genes in which human pathways. And visualize it, optimize the GO enrichment scatter plot and KEGG

scatter plot, intuitively see which pathway or aspect the gene is enriched in the most number of genes, and how significant it is. This achieves the goal of gene enrichment, obtaining key disease pathways, and laying the foundation for subsequent research.

Organize and summarize the effective ingredients and corresponding targets of Gardenia jasminoides for treating hyperuricemia using Excel, and import them together with the disease-related pathways of HUA into Cytoscape software to draw the Gardenia jasminoides component target pathway HUA network diagram. This makes the entire network pharmacology analysis more intuitive and concise, and thus determines which substance and target in Gardenia jasminoides are related to HUA through which pathways.

After conducting research on the gene level of active ingredients in drugs, a comprehensive evaluation of drug space and energy is also needed. Therefore, further research on molecular docking is needed. Quercetin, kaempferol, and β - stigmasterol obtained from previous experiments were selected as docking ligands, and the two core genes with the highest Degree values for core gene mining were used as receptors for docking [28]. Firstly, it is necessary to obtain the 3D structure of drug active ingredients in the TCMSP database as small molecule ligand molecules for molecular docking. Subsequently, TP53 and CASP3 core protein targets were retrieved in Unipart to obtain corresponding 3D structures. Small molecule ligands were optimized in Chem3D [29]. Discovery Studio was used to remove water molecules, separate the original ligands, add hydrogen atoms to the protein receptors, and molecular docking validation was performed in CB dock2 [30]. Observe the degree of matching and binding to determine the degree of action of the effective ingredients.

CHAPTER 3
EXPERIMENTAL PART

3.1 Obtaining effective ingredients and targets of drugs

Through searching the TCMSP database, a total of 98 active ingredients in Gardenia jasminoides were obtained. After screening with OB ≥ 30% and DL ≥ 0.18, it was found that there were 15 active ingredients in the drug, including quercetin, beta sitosterol, kaempferol, and cilantro. The basic information of drugs is shown in Table 3.1.

Table 3.1 – **Effective Components of Mulberry Leaves**

Name of Traditional Chinese Medicine	ID	Molecular name	OB	DL
Zhizi	MOL001406	crocetin	35.3	0.26
Zhizi	MOL001663	(4aS,6aR,6aS,6bR,8aR,10R,12aR,14bS）-10-hydroxy-2,2,6a,6b,9,9,12a-heptamethyl-1,3,4,5,6,6a,7,8,8a,10,11,12,13,14b-tetradecahydropicene-4a-carboxylic acid	32.03	0.76
Zhizi	MOL001941	Ammidin	34.55	0.22
Zhizi	MOL004561	Sudan III	84.07	0.59
Zhizi	MOL000098	quercetin	46.43	0.28
Zhizi	MOL0003	beta-sitosterol	36.91	0.75

Name of Traditional Chinese Medicine	ID	Molecular name	OB	DL
	58			
Zhizi	MOL000422	kaempferol	41.88	0.24
Zhizi	MOL000449	Stigmasterol	43.83	0.76
Zhizi	MOL001494	Mandenol	42	0.19
Zhizi	MOL001506	Supraene	33.55	0.42
Zhizi	MOL001942	isoimperatorin	45.46	0.23
Zhizi	MOL002883	Ethyl oleate （NF）	32.4	0.19
Zhizi	MOL003095	5-hydroxy-7-methoxy-2-（3,4,5-trimethoxyphenyl）chromone	51.96	0.41
Zhizi	MOL007245	3-Methylkempferol	60.16	0.26
Zhizi	MOL009038	GBGB	45.58	0.83

Subsequently, in the Chemical name search column of TCMSP, these active ingredients were searched to obtain their corresponding target proteins. The target proteins corresponding to these active ingredients were input into the UniProt database to obtain the gene names of the target proteins, namely target genes (hereinafter referred to as targets). In the experiment, the UniProtKB knowledge base

and Excel software were used to merge and deduplicate the compound target genes predicted in the above steps, and a "Gardenia effective ingredient target database" was established. 364 targets and 369 genes related to the active ingredients were obtained.

3.2 Acquisition of Common Target Genes for Drugs and Diseases and Construction of PPI Networks

By obtaining disease genes in GeneCards and OMIM, the obtained results were merged, organized, and deduplicated to obtain 906 effective genes.

In the Draw Venn Diagram, 906 disease pathogenic target genes and 369 drug active ingredient target genes were imported, respectively.

The Venn diagram is shown in Figure 3.1, and a total of 48 intersection genes were identified, XDH、RXRA、BIRC5、PON1、BCL2、ADRA2A、CASP8、PPARG、CRP、CXCL8、SELE、NFE2L2、THBD、TNF、PTGS2、ESR1、ADRB2、MYC、CCNA2、CCL2、STAT1、IL6、CASP3、MMP3、PPARA、IL10、MAPK1、PLAT、IFNG、TOP2A、MAPK8、ICAM1、BCL2L1、RELA、SLC2A4、RB1、SERPINE1、VCAM1、TP53、CASP9、CDKN1A、NCOA2、IL1B、NFKBIA、BAX、CD40LG、SLC6A3、TOP1。

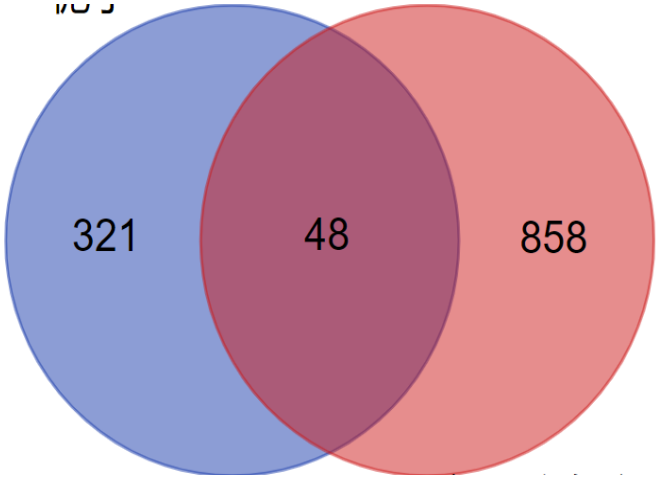

Figure 3.1 – Venn diagram of Gardenia jasminoides - HUA gene

Input the intersection genes mentioned above into the STRING database to obtain the PPI network diagram 3.2.

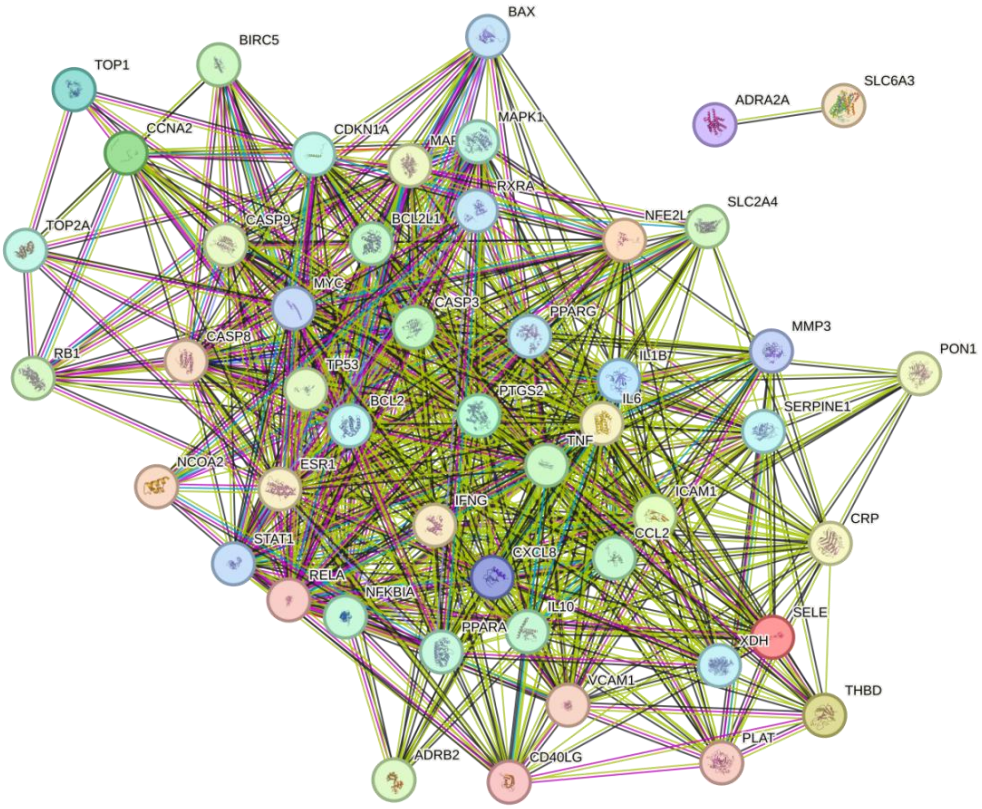

Figure 3.2 – Intersection Gene Protein Interaction PPI Network Diagram

There are only two isolated nodes in the network, and the connectivity between each node is good, which can effectively prove that the effective ingredients of Gardenia jasminoides can regulate multi target collaborative treatment of hyperuricemia, and then save the TSV format of the results. Using Cytoscape 3.10.1 software for visualization processing, core genes were mined using the CytoHubba plugin based on degree values. The top ten genes identified were TP53, CASP3, PTGS2, IL6, TNF, ESR1, IL1B, PPARG, BCL2, and MYC, as shown in Figure 3.3. The degree values correspond to 40, 40, 39, 39, 39, 38, 37, 37, and 37, respectively.

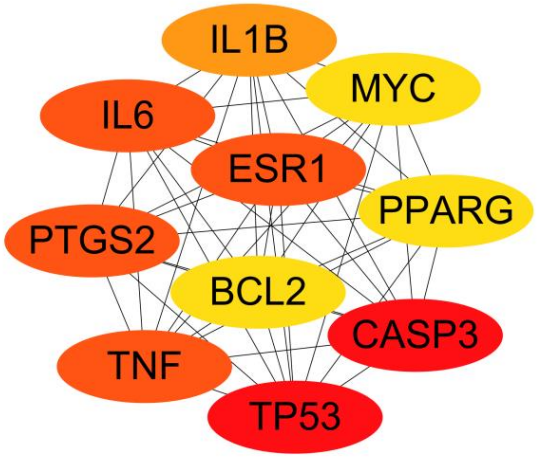

Figure 3.3 – Core gene mining results of cytoHubba Degree value

3.3 Gene enrichment analysis and component target pathway network diagram

The GO enrichment analysis results showed that there were a total of 2889 key targets composed of intersecting genes in the biological process (GO-BP) pathway, including response to lipopolysaccharide, response to molecule of bacterial origin, response to more necrotic factor, regulation of reactive oxygen species metabolic processes, etc. There were a total of 152 cell component expression processes (GO-CC), including membrane raft, membrane microbiome, membrane region, caveolol, etc. There are 236 processes related to molecular function (GO-MF), including DNA binding, transcription factor binding, etc Cytokine receptor binding, RNA polymerase ll specific DNA binding transcription factor binding, and repressing transcription factor binding were all selected, and the top 20 processes with significant rankings were plotted as scatter plots, as shown in Figures 3.4-3.6.

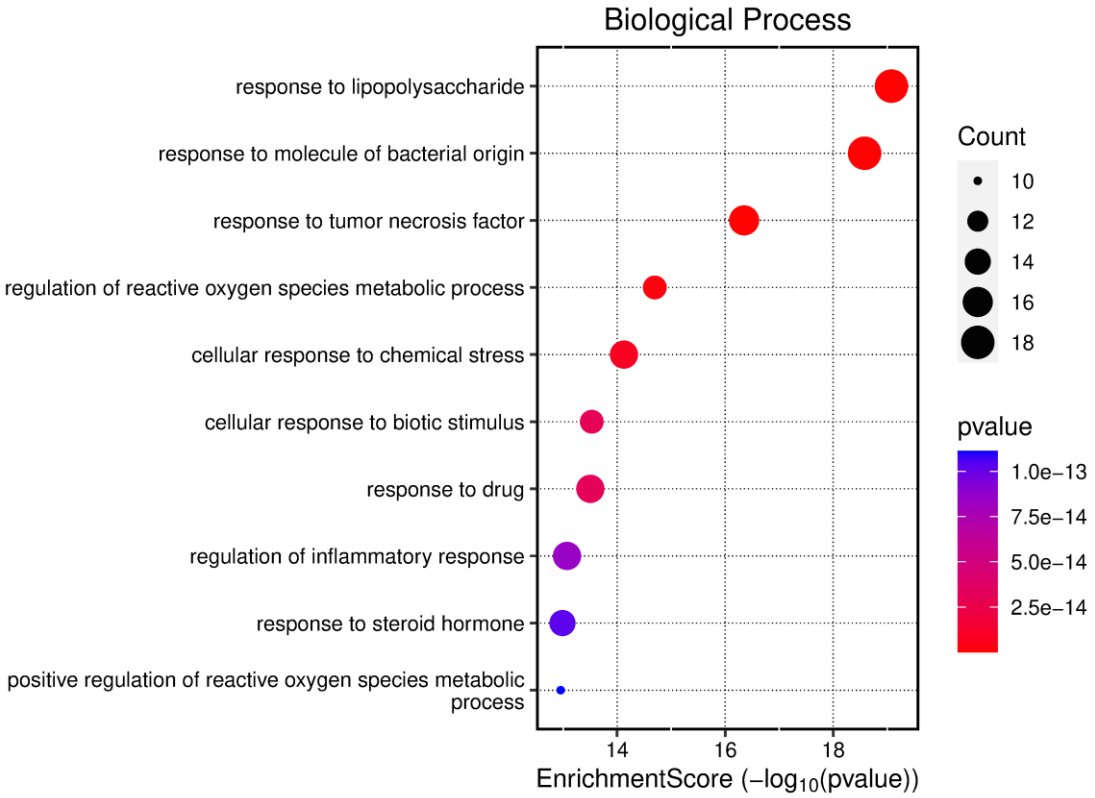

Figure 3.4 – Biological Process

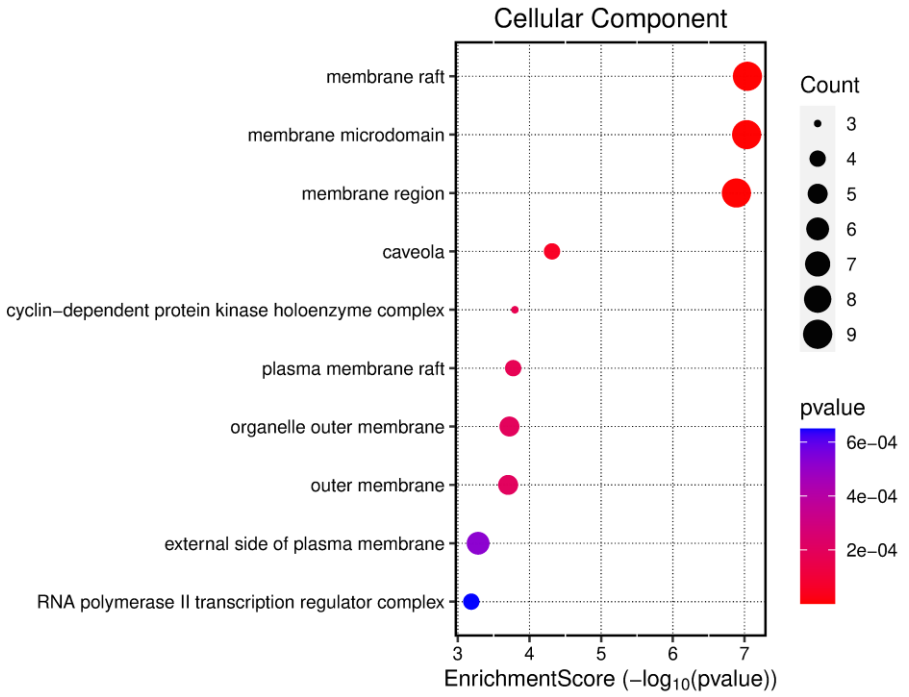

Figure 3.5 – Cellular Component

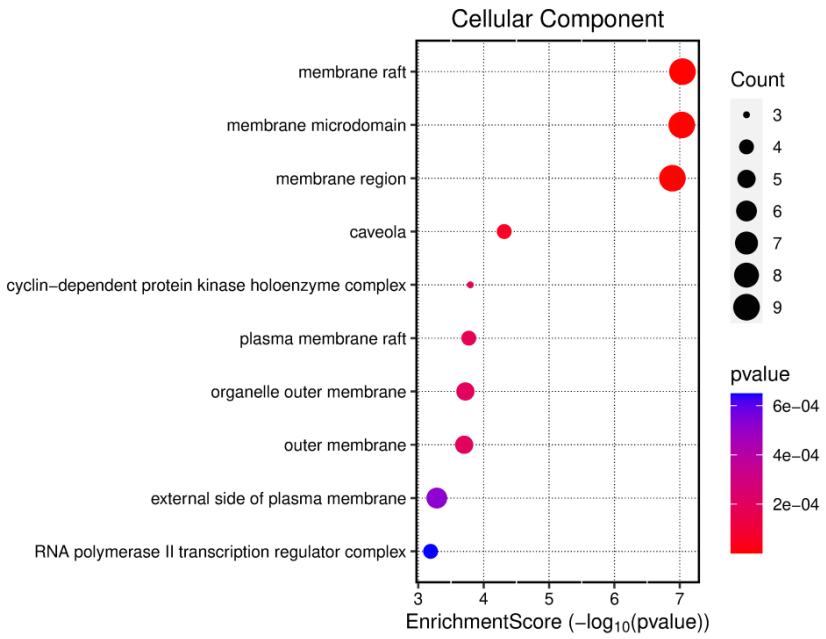

Figure 3.6 – Molecular Function

KEGG enrichment analysis revealed a total of 198 key target related pathways, including those enriched and significant in Lipid and spherosclerosis, AGE-RAGE signaling pathway in radial composites, Hepatitis B, TNF signaling pathway, etc. The KEGG pathway enrichment map was drawn by selecting the top 20 pathways with significance, as shown in Figure 3.7.

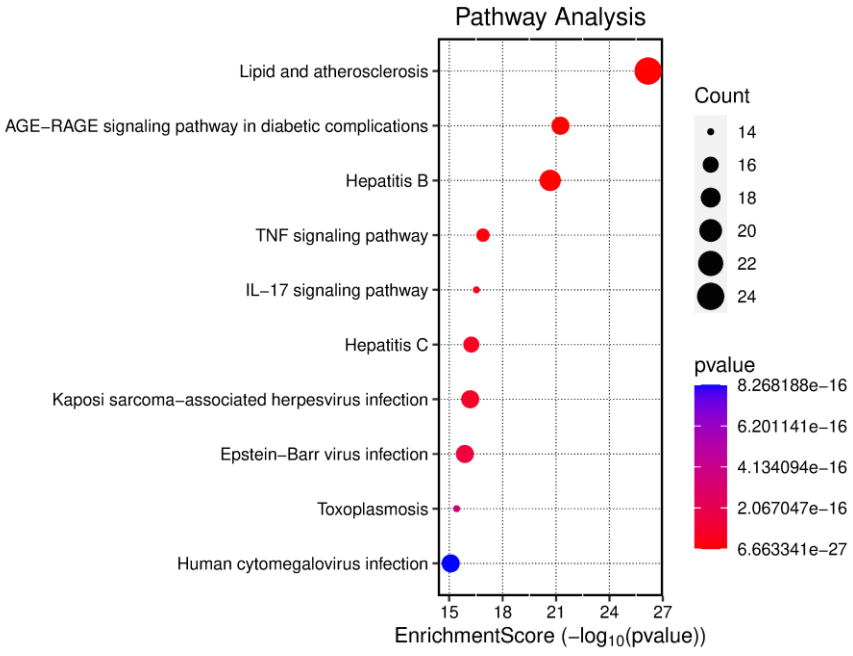

Figure 3.7 – KEGG pathway enrichment analysis

A total of 315 pathways related to HUA disease were excavated from the David database. The top 20 genes and their related genes were selected and merged with the target genes of key effective ingredients in the effective ingredients of Gardenia jasminoides that have therapeutic effects on HUA. They were sorted and named using Excel and imported into Cytoscape software for network drawing, resulting in Figure 3.8. From the graph, it can be visually seen that the active ingredients in Gardenia jasminoides, including MOL000098 quercetin, MOL000422 kaempferol, and MOL000358 beta sitosterol β - stigmasterol, have the highest correlation with HUA and have the best therapeutic potential.

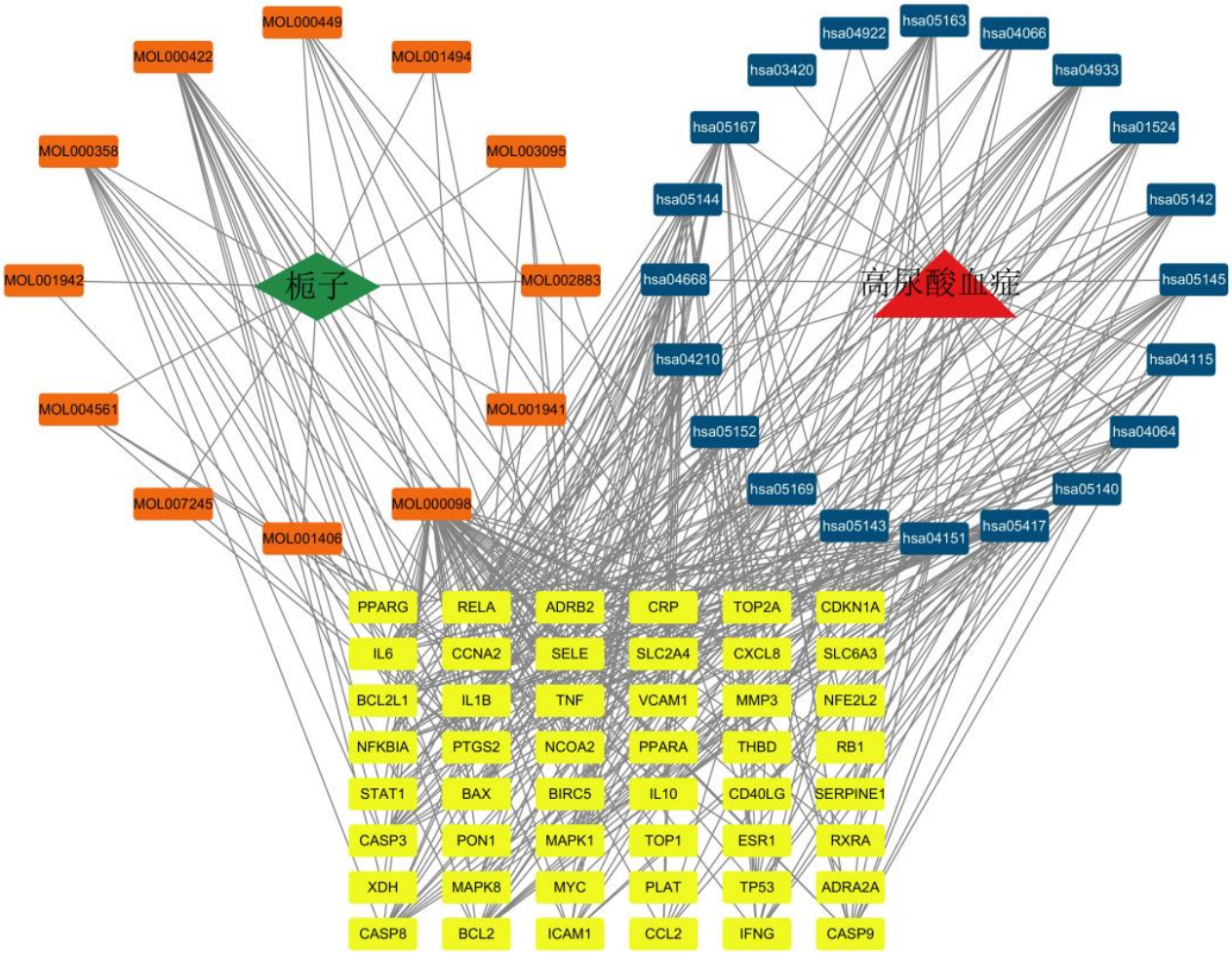

Figure 3.8 – Component Target Pathway HUA Network Diagram

3.4 Molecular docking verification

Using CB-dock2 for molecular docking verification, it was found that quercetin, kaempferol, and β sitosterol have binding energies of less than -5.0 to TP53 and CASP3 receptors. Among them, the fractions of CASP3 quercetin, CASP3 kaempferol, and CASP3 β sitosterol are less than -9.0, as shown in Table 3.3, and they have good docking effects in space, as shown in Figure 3.9. The results of molecular docking provide strong evidence to prove the mechanism of action of effective ingredients.

Table 3.3 – **Molecular docking binding energy table**

	Quercetin	Kaempferol	β - sitosterol
TP53	-7.4	-7.3	-7.8
CASP3	-9.7	-9.2	-9.0

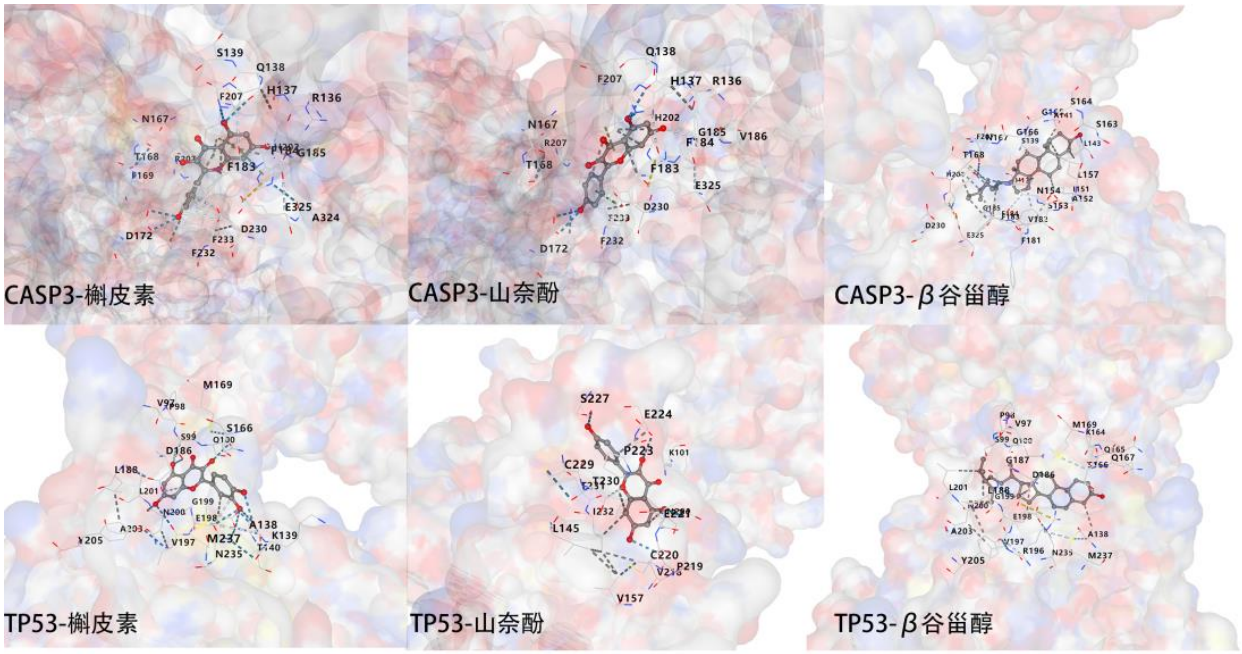

Figure 3.9 – Molecular docking results

3.5 Result analysis

The main symptom of HUA is gouty arthritis, and many patients with hyperuricemia may not have obvious symptoms, especially in the early stages. However, when uric acid levels are too high, it may lead to the occurrence of gout. Gout is an acute inflammatory arthritis caused by the deposition of urete crystals in the joints. It is an immune inflammatory disease mediated by the deposition of urate crystals in synovial tissue, and its inflammation is closely related to inflammatory factors such as tumor necrosis factor alpha [31].

So, HUA patients can alleviate and treat gouty arthritis through immune inflammatory response regulation [32]. According to clinical research results, literature has shown that Gardenia jasminoides can help alleviate the inflammatory cytokine response in patients with gouty arthritis. The active ingredients involved mainly include quercetin, kaempferol, and β - sitosterol, which can enhance the human immune response and inhibit the expression of inflammatory cytokines such as tumor necrosis factor alpha, thus playing a certain role in the treatment of gouty arthritis [33]. KEGG analysis results show that the mechanism of Gardenia jasminoides Ellis in treating gouty arthritis involves lipid and atherosclerosis signaling pathway, AGE-RAGE signaling pathway, IL-17 signaling pathway, TNF signaling pathway, etc. These reactions are closely related to human physiological metabolism. Results The lipid and atherosclerosis pathways showed that hyperuricemia was closely related to lipid metabolism disorder, and hyperuricemia and hyperlipidemia had a very significant interaction.

Hyperuricemia has various effects on human metabolism and cardiovascular health. Hyperuricemia can directly stimulate the liver to synthesize more triglycerides. An increase in uric acid in serum will reduce the activity of lipoprotein lipase (LPL), limit the catabolism of triglycerides, and lead to an increase in the level of triglycerides in the blood. Hyperuricemia can promote the oxidation of low-density lipoprotein (LDL) and lipid peroxidation, thus increasing the risk of atherosclerosis [34]. It seems that hyperuricemia not only affects purine metabolism, but also interferes with lipid metabolism and vascular health,

increasing the risk of cardiovascular disease; Hyperuricemia is closely related to a variety of metabolic diseases, including obesity, diabetes and cardiovascular diseases, involving TNF signaling pathway, IL-17 signaling pathway and AGE-RAGE signaling pathway. Xanthine dehydrogenase (XDH) plays an important role in hyperuricemia [35]. XDH and xanthine oxidase (XOD) are two different forms of the same enzyme class, both expressed by XDH mRNA and can be converted to each other. When the XOD activity in the body abnormally increases, a large amount of uric acid is produced. Therefore, inhibiting XDH is equivalent to inhibiting XOD activity, which can effectively inhibit uric acid production and alleviate symptoms of hyperuricemia [36 37].

CONCLUSIONS

In summary, this qualification thesis breaks through the traditional single target research model through the method of network pharmacology, explores the mechanisms of multiple effective components in Gardenia jasminoides on hyperuricemia at the genetic and metabolic pathways levels, and verifies the feasibility of their effects on the human body through molecular docking technology in space. It is found that quercetin, kaempferol, and β - sitosterol in Gardenia jasminoides are the potential molecular basis for their anti-inflammatory and cardiovascular protective effects. This provides certain reference and experience for the clinical treatment of hyperuricemia in subsequent Gardenia jasminoides.

The shortcomings of this study are that in terms of database maintenance, the data is constantly being updated, and the pathogenic genes of HUA have not been completely identified by people. Moreover, the target proteins of the active ingredients of the drug are still largely unconfirmed, and their ability to act cannot be proven at this stage. Relying solely on the various data in the database is not entirely convincing, and verifying another network data through one network is not rigorous enough. If animal experiments are added in the future, gradually deepening from in vitro research to in vivo research, it may make the experimental data more rigorous and effective.

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