Investigating the mechanism of action of hemofugal and blood-stasis-expelling soup in the treatment of coronary heart disease based on network pharmacology

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Abstract: Objective: To investigate the mechanism of action of Xuefu yuyuxu Tang in the treatment of Coronary Heart Disease (CHD) based on network-based pharmacology. Methods: Traditional Chinese Medicine Systems Pharmacology Databaseand Analysis Platform (TCMSP) was used to retrieve the active ingredients and targets of Chinese medicines in Xufu yuyuxu Tang. Cytoscape3.7.2 was used to draw the network diagram of "TCM-active ingredients-target genes", and Slring database was used to construct the protein-protein interaction (PPI) network diagram, and the PPI network diagram was analyzed with the help of CyoNCA plug-in for network topology analysis, and the key targets were screened with the help of Centiseape plug-in, and the key targets were selected with the help of Centiseape plug-in. The PPI network map was analyzed by CyoNCA plug-in for network topology analysis, and Centiseape plug-in was used to screen the key targets, and MetaScape database was used for GO function analysis and KEGG pathway enrichment analysis. Molecular docking prediction was performed by AutoDockTools-1.5.6. Results: By searching TCMSP, 195 active ingredients were screened, and the pathways related to coronary heart disease were lipid and atherosclerosis, hypoxia-inducible factor-1, relaxin, and nuclear factor-KB signaling pathway, etc. The results of topology analysis showed that the target genes with the top 10 residences of Desree value were, in order, PTGS2MMP2, VEGFA, AKTI, ALB, CASP3, STAT3, II6, MM9SERPINEI. The main active ingredients of this formula, lignocaine, kaempferol, naringenin, bound well to the target proteins protein kinase 1, albumin and insulin. Conclusion: Based on the network pharmacology, this paper analyzed the active ingredients and their mechanism of action in the treatment of coronary artery disease with blood-fu chasing blood stasis soup, and found that blood-fu chasing blood stasis soup treats coronary artery disease through multi-targets, and several ingredients showed good activity, which provided a theoretical basis for the further study of the mechanism of action of the formula.

1 Introduction

It has been found [1] that CHD may lead to different degrees of dysfunction and affect patients' quality of life, and its common risk factors are blood pressure level and smoking. Ancient medical practitioners believed that the pathogenesis of CHD is closely related to exogenous evils, phlegm, qi and blood deficiency, and yang deficiency, and the pathogenic factors are qi stagnation and blood stasis [2] - [3]. Hematophagus and Blood Stasis Soup is a classic formula for activating blood circulation and removing blood stasis, which is composed of a variety of traditional Chinese medicines. It is commonly used clinically for the treatment of cardiovascular and cerebrovascular diseases, with precise efficacy [4] - [5]. In this study, the research method of network pharmacology was used to investigate the molecular mechanism of bloodfuyuyuzhu Tang for the treatment of CHD by combining the database of traditional Chinese medicine components, potential target prediction technology and network analysis platform, aiming to provide a theoretical basis for clinical application.

2 Information and methods

2.1 Screening of the active ingredients of Hematopoietic Stasis-Expelling Tang and obtaining the corresponding targets

TCMSP was used to retrieve the drug composition and active ingredients in Blood Palace and Blood Stasis Expelling Tang (oral bioavailability (OB) ≥30% and drug likeness (DL) ≥0.18 as conditions [6] predicted target genes of the corresponding ingredients), with the keywords "peach kernel", "safflower", "licorice", "chaihu", "dihuang", and ", "saffron", "licorice",

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"chaihu", "dihuang" "hyssop" and "platycodon", and the active ingredients and target genes of traditional Chinese medicines that met the criteria were obtained according to the above screening criteria.

2.2 Screening of disease targets

The target genes associated with the development of coronary heart disease were searched in OMIM (https://omim.org), GeneCards (https ://www.genecards, org/), DisGeNET (https: //www, disgenet. org/), TTD (https://omim.org), and DrugBank (https://omim.org) databases, respectively, with the keyword of "coronary heart disease." //www, disgenet. org/), TTD (http://metascape.orggp), and DrugBank (https://go.drugbank.com) databases were searched for target genes associated with the development of coronary heart disease. After summarizing and de-emphasizing the disease targets from the above databases, the correlation between the target genes and the disease was calculated by the Gis algorithm, and the target genes with the correlation degree of > 30 were screened out, which are the potential targets for the treatment of CHD disease. Through the targets, the association between diseases and active ingredients can be constructed, and then the relationship with each herb can be established.

2.3 "Traditional Chinese medicine-active ingredient-target gene" network diagram and PPI network construction

Based on the relationship between the active ingredients in Blood Palace and Blood Stasis Relieving Tang and the target genes of coronary heart disease, we uploaded the traditional Chinese medicine, active ingredients and the corresponding targets of Blood Palace and Blood Stasis Relieving Tang in Cyloscape 3.7.1 software, constructed a network diagram of traditional Chinese medicine-ingredient-targets, and intersected the targets of the corresponding targets of the active ingredients with the targets of the disease. We imported the STRING11.5 database (hlps://en,string-db.org/), limited the species to "homo sapiens", and the protein-protein interaction (PP) ≥ 0.4, and downloaded the PPI data. The PPI data were used to obtain the key target information and visualized by Cyloscape 3.7.2 software.

2.4 Topology analysis of target gene regulatory networks

The PP network map of disease and compound intersection genes obtained from the STRING11.5 database was imported into Cytoscape, and the network topology analysis was performed using the CytoNCA plug-in in the software to screen the core target genes of the treatment of coronary artery disease with Hematopoeia and Blood Stasis Soup.

2.5 Gene ontology function and genomic pathway enrichment analysis

The intersecting targets were enriched by Gene0ntology (GO) database and Kyoto Encyclopedia of Genes and Genomes (KEGG) database with a set value of P≤0.5, and the species was set as "H. sapiens H. sapiens", the minimum overlap value was 3, P<0.01, and the top 10 results of the three modules of Biological Process (BP), Molecular Funclion (MF) and CellCommposilion (CC) were selected for GO enrichment analysis. For pathway analysis, KEGG was selected, and the results of the first 20 entries were visualized to construct bar graphs and bubble graphs. See figure 2 and figures 3.

2.6 Molecular docking

The 2D structure of the active ingredient was obtained from the database in mol2 format and the 3D structure of the target was obtained from the database in mol2 format, and the molecular docking was carried out by using AuloDockTools-1.5.6 software, and the results of the docking were presented by the Pymol software for visualization.

3 RESULTS

3.1 Screening of active ingredients and target genes of Hematopoietic Blood Stasis Dispelling Tang

By searching TCMSP, the active ingredients and target genes of Hematopoietic Stasis-Expelling Tang were obtained, and a total of 195 active ingredients were obtained after removing the duplicates according to the standard, including 23 peach kernels, 24 saffron, 2 Angelica sinensis, 4 cowslips, 7 Rhizoma Ligustici Chuanxiong, 8 Platycodon grandiflorus, 30 Paeonia lactiflora, 5 Hovenia citri Reticulatae, 19 Chai Hu and 92 Glycyrrhiza glabra, and there were 11 common ingredients, and 3 active ingredients of Sheng Dihuang were added in combination with literature The total number of components was 11. The network diagram was visualized by Cytoscape 3.7.1 software, and the network consisted of 543 nodes and 2504 edges.

3.2 Target gene screening and target gene database construction

With the help of Per software, the target genes of coronary heart disease with correlation degree >30 were combined with the target genes with potential effects of Hematopoeia and Blood Palace bypassing Blood Stasis Soup, and 70 intersecting target genes were obtained, which were constructed as a database of target genes of Hematopoeia and Blood Palace bypassing Blood Stasis Soup for the treatment of coronary heart disease and used in the subsequent analysis.

3.3 Construction of "Traditional Chinese Medicine-Active Ingredient-Target Gene" Compound Network Diagram

Based on the relationship between the interaction of active ingredients in Haifuyuyuyuetu Tang and the target genes of coronary heart disease, a network diagram of the compound was constructed and visualized, see Figure 1.

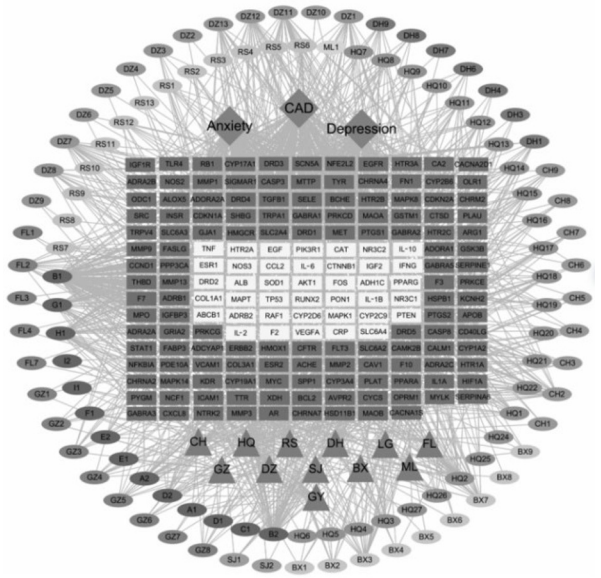

Figure 1. "Traditional Chinese Medicine - Active Ingredient - Target Gene" Compound Network Diagram.

3.4 Construction and screening of PPI networks

The intersecting genes were used to construct a PPI network using STRING11.5, in which "edges" are the associations between target genes and nodes are target genes. There were 65 nodes and 1141 edges, with an average node degree of 31.2 and an average local clustering coefficient of 0.740; the confidence level was set to be greater than 0.9, and the PPI core network was screened. See Figure 2.

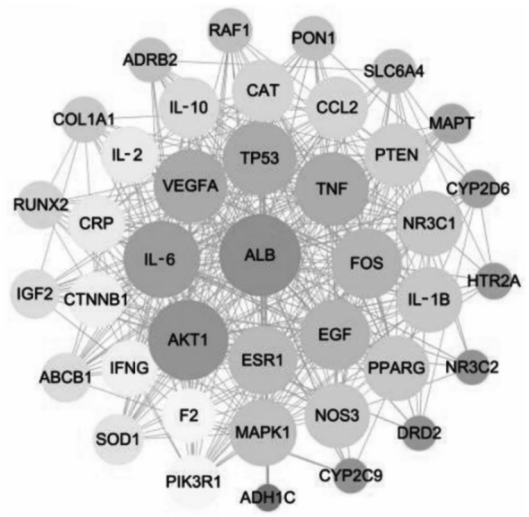

Figure 2. PPI network diagram.

3.5 Topology Analysis

The PPI network map of disease and compound intersection genes obtained from the STRING11.5 database was imported into Cytoscape, and the network topology analysis was performed using the CytoNCA plug-in in the software to screen the core target genes of the treatment of coronary artery disease with Hematopoeia and Blood Stasis Soup.

3.6 Enrichment analysis

The biological processes involved in the treatment of CHD with Bloodfu Yucha Tang are mainly involved in hormonal responses, responses to nutrient levels, and inflammatory responses. In addition, screening the key target genes mapping KEGG pathway for enrichment analysis, we can get that the treatment of CHD with bloodfu yuicha is mainly involved in lipid and atherosclerosis, hypoxia inducible factor-1 (HIF-1) relaxin, nuclear factor kB (nuclear factor kappa-B) signaling pathway and so on. NF-KB) signaling pathway, etc. See Figure 3 and Figure 4.

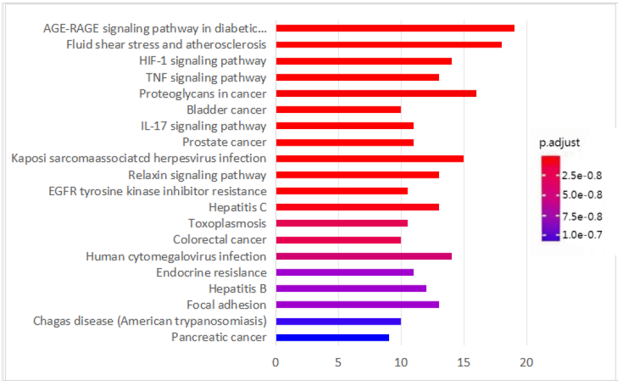

Figure 3. Bar graph of KEGG enrichment analysis.

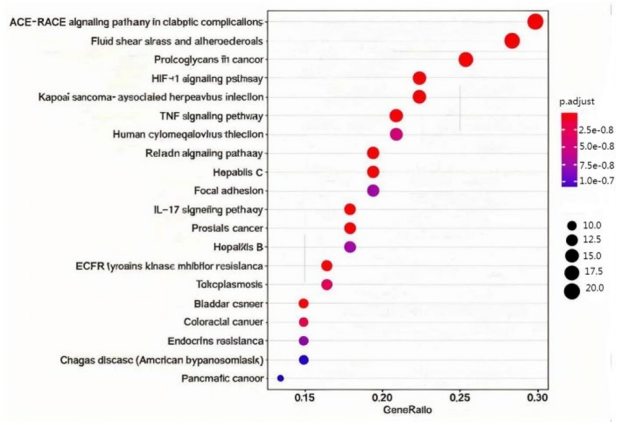

Figure 4. Bubble plot of KEGG enrichment analysis.

3.7 Molecular docking results

The seven mass markers screened from the Blood Palace Chasing Blood Stasis Soup were molecularly docked with five target proteins, and the binding energy <0 indicated that the receptor and the ligand could

spontaneously bind; the smaller the binding energy, the better the binding between the two. The specific results are shown in Table 1 and Figure 5.

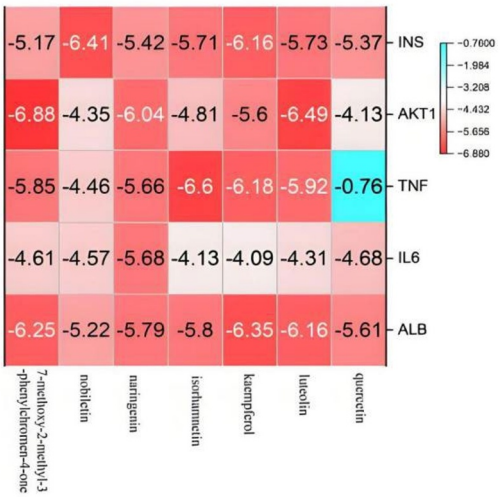

Figure 5. Molecular docking diagram.

Table 1. Active ingredient-core target docking results binding energy.

chemical compound	Free energy of binding (kcal/mol)				
	ALB	IL-6	TNF	AKT I	INS
quercetin	-5.47	-4.70	-0.69	-4.12	-5.31
luteolin	-6.10	-4.34	-5.93	-6.41	-5.77
Kaempferol	-6.38	-4.03	-6.21	-5.60	-6.22
isorhamnetin	-5.82	-4.15	-6.65	-4.88	-5.67
naringenin	-5.69	-5.62	-5.66	-6.10	-5.41
Nobiletin (Chuanpianin)	-5.18	-4.59	-4.42	-4.41	-6.45
7-methoxy-2-methyI-3-phenyIchromen-4-one	-6.23	-4.64	-5.88	-6.90	-5.22

4 Discussion

In this study, we investigated the multiple pathways, components and targets of bloodfuyuyuetu Tang in the treatment of CHD by integrative application of network pharmacology. These components can affect important biological processes such as lipid and atherosclerosis, HIF-1 relaxin, NF-KB and insulin resistance by interfering with the key targets, thus modulating the body's inflammatory response, oxidative stress, angiogenesis and vascular dysfunction, apoptosis and lipid metabolism, and ultimately realizing the therapeutic effect of treating coronary heart disease and stroke. However, due to the incomplete information in the database, further clinical and experimental studies are needed to verify this in the future.

References

1. KANG Huifang, LI Jing, GUO Zhihua, LIU Yi, WANG Mingyun, ZHANG Qiuyan. Discussion on the mechanism of "different diseases treated with the same treatment" of coronary artery disease and ischemic stroke by hemifluvialyushu soup based on network pharmacology and molecular docking technology [J]. Global Chinese Medicine, 2024, 17(4):585-594.
2. WANG Xinhui, WANG Sutong, WANG Yongcheng, LI Xiao. Exploring the mechanism of Chai Hu plus Long Bone Oyster Soup in the treatment of coronary heart disease combined with anxiety and depression based on network pharmacology [J]. China Traditional Chinese Medicine Modern Distance Education, 2025, 23(2):147-150
3. LIN Jing, MOK Junxiu, SONG Yan, ZHOU Xin, LAI Yongsheng. Discussion on the mechanism of action of hemifugal and blood-stasis-expelling soup in the treatment of idiopathic pulmonary fibrosis based on network pharmacology and experimental validation [J]. Proprietary Chinese Medicine, 2024, 46(2):658-665
4. YE Jia-Hao, HU Zhi-Xi, ZHONG Sen-Jie, YANG Meng, YAO Tao, QIU Hong, XIONG Xia-Jun. Discussion on the mechanism of action of hemifuture and chiropractic soup in the treatment of coronary heart disease based on network pharmacology [J]. World TCM, 2021, 16(9):1400-1405
5. HONG Wei, YU Ting. Evaluation of the pharmacological effects of hemofoo chi yu tang combined with the addition and subtraction of danshen drink in the treatment of cardiovascular blood stasis type coronary heart disease[J]. Chinese Science and Technology Journal Database (Citation Edition) Medicine and Health, 2024(10):0124-0127
6. HUANG Min,LIU Shuihua, ZHANG Shiyu, SHI Weiqi, CAI Zongyu, LU Jianqi. Progress in the treatment of coronary artery disease with hemofluorescence and blood circulation[J]. Bright Chinese Medicine, 2024, 39(4):831-832F0003, F0004