A network-based pharmacologic investigation of the mechanism of action of frankincense-myrrh in the treatment of painful diabetic peripheral neuropathy

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Abstract: *To explore the mechanism of mastic and myrrh in the treatment of painful diabetic peripheral neuropathy (PDN) using network pharmacological method. The active ingredients of frankincense and myrrha and their related targets were retrieved from the Chinese Medicine System Pharmacology Database and Analysis Platform (TCMSP), and the gene names were converted using Uniprot protein database. GeneCards, NCBI and OMIM databases were used to search the targets of painful diabetic peripheral neuropathy, and VENNY2.1.0 was used to obtain the intersection targets. Based on the intersection targets, protein interaction analysis, GO cell function and KEGG signaling pathway enrichment analysis were used to analyze the mechanism of mastic and myrrha in the treatment of painful diabetic peripheral neuropathy (PDN). Eight mastic compounds and 11 targets were found. There were 45 bioactive components and 184 targets in myrrh. There were 2265 PDN related genes and 123 common targets of drugs and diseases. Protein network interaction revealed Recombinant Nitric Oxide Synthase 2 (NOS2), factor II (F2) and Recombinant Prostaglandin Endoperoxide Synthase 2 (PTGS2), Dipeptidyl peptidase-4 (DPP4), Recombinant Protease, Serine 1 (PRSS1), estrogen receptor1 (ESR1) and other targets may be the key targets of frankincense and myrrh in the treatment of PDN. GO enrichment analysis included 320 items, involving cellular reactive oxygen species metabolism, response to lipopolysaccharide, epithelial cell proliferation, etc. KEGG enrichment analysis identified 166 signaling pathways, among which the AGE-RAGE signaling pathway in diabetic complications, lipid and atherosclerosis, fluid shear stress and atherosclerosis were the possible pathways of mastic and myrrha in the treatment of PDN.*

Keywords: *frankincense-myrrh; painful diabetic peripheral neuropathy; network pharmacology*

1. Introduction

Painful diabetic peripheral neuropathy (PDN) is one of the common complications of diabetes mellitus, which is mainly characterized by nociceptive hypersensitivity, nociceptive hypersensitivity, spontaneous pain and a certain degree of sensory loss, with severe and persistent pain, which can lead to depression, anxiety, and sleep disorders, seriously affecting the quality of life[1] . Epidemiological surveys have found that about 1/3 of diabetes mellitus and 1/4 of early diabetes mellitus patients develop the disease, with a high prevalence [2] , symmetric peripheral neuropathic pain is the main manifestation of the disease, which can also be manifested as mononeuralgia or brachial plexus or lumbosacral plexus neuralgia[3] , and patients' pain symptoms are not relieved with standardized clinical treatments, such as antidepressant and anticonvulsant medications. A study at[4] confirmed that only about 15% of PDN patients improved on internationally recommended analgesics, while the rest had no change in pain, and even had increased pain in 8% of patients. In the face of the increasing number of PDN patients, how to effectively relieve pain and improve the quality of survival of patients has become the focus of the treatment of this disease.

Frankincense and myrrh, as the classic Chinese medicine combination of activating blood circulation and removing blood stasis, promoting qi and relieving pain, have been used as a pair of medicines as early as in the Ming Dynasty in the "Guidelines for the Treatment of Evidence", and the two medicines have the effect of relieving pain, and they are often used in combination with each other. Clinical studies have found that frankincense and myrrh are not only anti-inflammatory and analgesic, but also regulate neuronal function, and play an indispensable role in the pathogenesis and treatment of neuropathic pain[5] . However, the mechanism of action of frankincense-myrrh pairs in painful diabetic peripheral neuropathy is still unclear, and it is not possible to deeply explore the relationship between drug components-targets of action-disease. As a multidisciplinary technology integrating systems biology, network biology,

molecular pharmacology, molecular dynamics, etc., network pharmacology[6] can help to analyze the interactions between drug components and disease nodes in the network by constructing a multilevel network of "disease-phenotype-gene-drug". Based on this, this paper attempts to systematically explore the mechanism of Boswellia serrata-myrrh pairs and painful diabetic peripheral neuropathy from macro to micro by means of network pharmacology, so as to provide a theoretical basis for the clinical use of Boswellia serrata and myrrh in the treatment of painful diabetic peripheral neuropathy.

2. Materials and Methods

2.1. Query for active ingredient and target information of frankincense-myrrh

Through TCMSP database[7] Compound compositions and targets of Boswellia serrata and Myrrh were searched and compositional screening conditions were OB ≥ 30% and DL ≥ 0.18. All targets were uniprot database (https://www.uniprot.org/)[8] calibrated to remove non-human targets and gene name conversion of target proteins.

2.2. PDN disease target finding

The keywords "painful diabetic peripheral neuropathy" were used in GeneCards[9] database (https://www.genecards.org/), NCBI gene database (https://www.genecards.org/), and OMIM database[10] (https://www.ncbi.nlm.nih.gov/), and OMIM database (https://www.ncbi.nlm.nih.gov/).[11] (https://www.omim.org/), and OMIM database to search for targets related to painful diabetic peripheral neuropathy. The screened drug targets and disease targets were entered into Venny 2.1, a software for creating Wayne diagrams, to obtain the common targets, which were used as predictive targets for drug action in the disease for pathway enrichment analysis.

2.3. Construction and analysis of the PPI interaction network

The drug-disease intersection targets were imported into the STRING11.5 database (http://cn.string-db.org), and the protein species was set to human (Homosapiens) to construct a protein-protein interaction network model. Cytoscape 3.9.1 software was used to construct the frankincense-myrrh-painful diabetic peripheral neuropathy protein-protein interaction visualization network diagram, and the PPI network was imported into Cytoscape 3.8.0.[12] The PPI network was imported into Cystoscape 3.8.0, and the genes with scores greater than the average score were sorted by degree, and the NetworkAnalyzer tool was applied to analyze the topology by selecting the genes with scores greater than the average score as the key targets.

2.4. Component-Disease Target Network Construction

In order to better understand the complex relationship between components, diseases and corresponding targets, the chemical constituents, PDNs and targets of Boswellia serrata and myrrh were included as the basis for the construction of component-disease-target network diagrams by using Cytoscape 3.9.0 software.

2.5. GO enrichment analysis and KEGG pathway enrichment

Enrichment of biological processes (BP), molecular functions (MF) and cellular components (CC) of GO was performed for drug-disease shared targets, and the String database was used to filter the items with corrected P-value <0.05, and the enriched BP, MF and cellular components were plotted as bar charts and bubble plots using R 4.0.3 software. The KEGG pathway enrichment analysis of drug-disease shared targets was performed by referencing the String database, and the items with corrected P < 0.05 were screened, and the histograms and bubble plots were drawn using R 4.0.3 , after installing and referencing the clusterProfiler package.

2.6. Component-Disease-Pathway-Target Network Construction

Component-disease-pathway-target network files were imported into Cytoscape 3.8.0 for pathway network mapping. The multi-component-multi-target action of active ingredients of traditional Chinese medicine in the treatment of painful diabetic peripheral neuropathy was characterized more intuitively.

3. Results

3.1. Results of screening of active ingredients and targets of frankincense-myrrh

Through TCMSP database[7] (https://tcmspw.com/tcmsp.php) to retrieve compound compositions and targets of frankincense and myrrh, and the composition screening conditions were OB ≥ 30% and DL ≥ 0.18. Then all the targets were uniprot database (https://www.uniprot.org/)[8] Then all targets were corrected by uniprot database (), non-human targets were removed and target proteins were genetically converted, and after summarizing and deleting, Boswellia serrata obtained 8 compound components and 11 targets; myrrh obtained 45 compound components and 184 targets.

3.2. Results of PDN disease target screening

The GeneCards, NCBI and OMIM databases were used to find the targets related to painful diabetic peripheral neuropathy. After searching, the GeneCards database yielded 2,217 related targets; the NCBI database yielded 6 related targets; and the OMIM database yielded 158 related targets. After combining the genes from these three databases and deleting them, 2265 genes related to painful diabetic peripheral neuropathy were obtained. The screened 184 frankincense and myrrh targets and 2265 painful diabetic peripheral neuropathy targets were inputted into the Wayne diagram creation software Venny 2.1, and 123 intersecting targets were obtained, and the top 20 were Recombinant Nitric Oxide Synthase 2 (NOS2), and factor II (F2), respectively, Recombinant Prostaglandin Endoperoxide Synthase 2 (PTGS2), Dipeptidyl peptidase-4 (DPP4), Recombinant Protease, Serine 1 (PRSS1), estrogen receptor1 (ESR1), aldose reductase (AR), pepsinogenratio (PGR), Recombinant Human Cyclin-Dependent Kinase Inhibitor 1A (CDKN1A), matrix metalloproteinase-2 (MMP2), matrix metalloproteinase-9 (MMP9), α nuclear factor of kappa light polypeptide gene enhancer in Bcells inhibitor, (NFKBIAB), C-X-C motif chemokine ligand 8 (CXCL8), protein kinase C, beta 1 (PRKCB), insulin like growth factor 2 (IGF2), glutathione S-transferase M1 (GSTM1), Cyclooxygenase 1 (PTGS1), Hreonine-proteinkinase (AKT), Interleukin 6 (IL-6), Interleukin 2 (IL-2), and Human topoisomerase 2a (TOP2a) as shown in Figure 1. as shown in Figure 1.
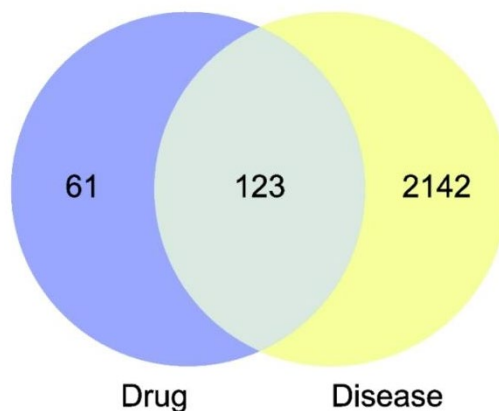

Figure 1: Venn diagram of drug-disease intersection targets

3.3. Drug-disease PPI construction

Entering Drug-Disease Shared Targets into the String Database[13] (https://string-db.org/cgi/input.pl The PPI network was constructed by setting the biological species as "Homo sapiens", and the PPI network was obtained (Figure 2). 123 nodes and 2161 edges were found in the network, and the average degree value was 35.1.) There are 123 nodes and 2161 edges in the network, and the average degree value is 35.1. In order to get a more intuitive protein-protein interaction network, the TSV file of the protein-protein interaction network was imported into Cytoscape 3.8.0 for Degree analysis, and the nodes were constructed according to the node degree value, and the node color and size were adjusted according to the degree value, the larger the node, the more colorful the node. Adjustment, the larger the node, the bluer the color, the larger the degree value, and the thickness of the line, from thick to thin indicates that the edge betweenness is from large to small, which can get a more intuitive association network diagram (Figure 3).
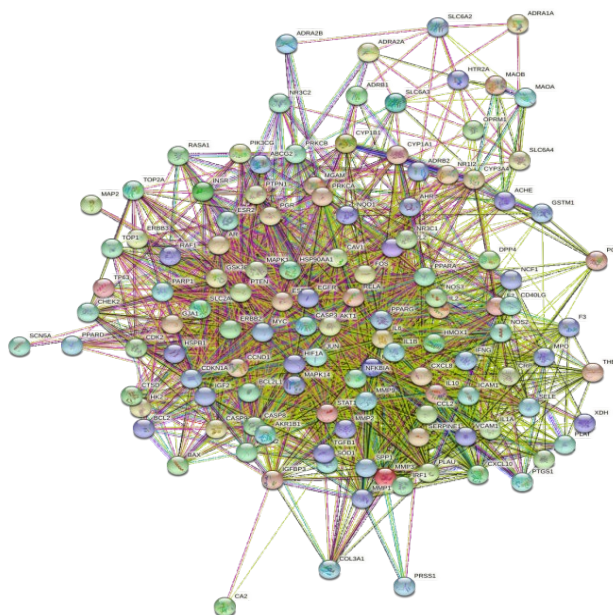
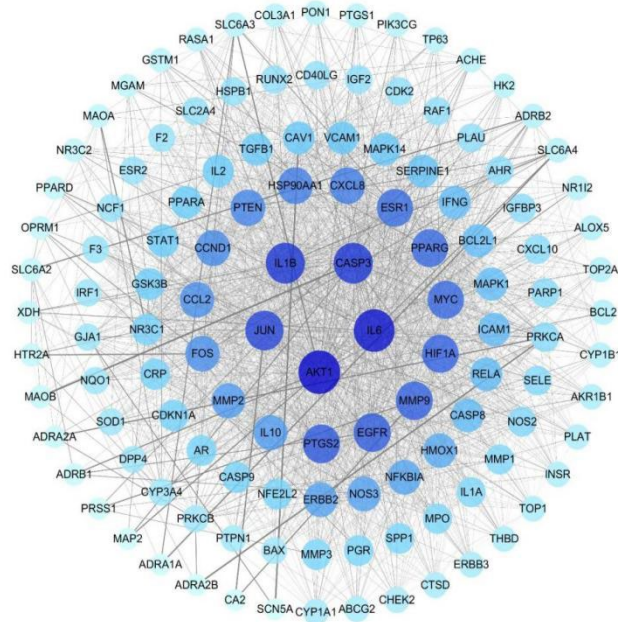*Figure 2: Network diagram of drug-disease target intersection.*

Figure 3: Cystoscape 3.8.0Degree value key target analysis map.

3.4. Component-Disease Target Network Construction

The target proteins were obtained by cytoHubba plug-in based on the Degree value, and the genes with scores greater than the average score were selected as the key targets according to the Degree sorting, and a total of 54 key targets were screened out, and the first 20 targets (Figure 4) were mainly Akt Serine/ThreonineKinase 1 (AKT1), Interleukin (IL)-6, Caspase3 (CASP3), Interleukin-1β (IL-1β), matrix metalloprotein (MMP)-9, epidermal growth factor receptor (EGFR), JUN), estrogen receptor (ESR-1), Peroxisome Proliferator Activated Receptor Gamma (PPARG), myelocytomatosis oncogene (MYC), Oestrogen receptor 1 (ESR1) etc. It is hypothesized that these nodes may be key nodes in the treatment of painful diabetic peripheral neuropathy with frankincense and myrrh. In order to better understand the complex interactions among components, diseases and corresponding targets, the screened drug components, diseases and targets were used as the basis for the component-disease-target network mapping by cytoscape 3.8.0 (Figure 5).
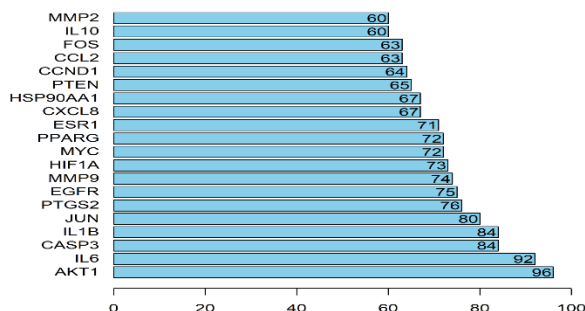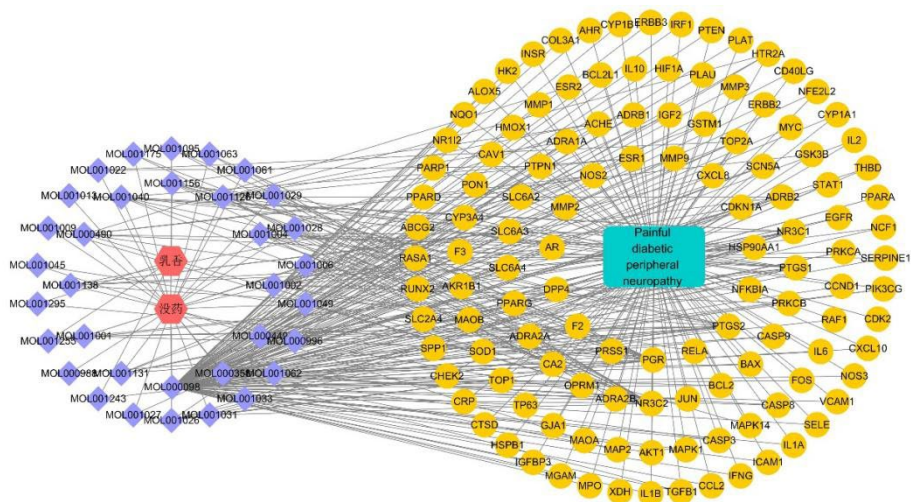
Figure 4: Topology analysis.

Figure 5:Frankincense-myrrh-painful diabetic peripheral neuropathy target-pathway network.

Note: In the network, blue color is the active ingredient, yellow color is the target of the drug acting on the disease, red color is the traditional Chinese medicine, and green rectangle is the disease.

3.5. GO functional enrichment analysis with KEGG pathway enrichment analysis

The shared drug-disease targets were enriched for biological processes (BP), molecular functions (MF), and cellular components (CC) of GO, and the String database was referenced to screen the items with corrected P-value <0.05, which enriched 2,099 biological processes, 153 molecular function-related items, and 61 cellular component-related items. The analysis reported the top 20 highest enrichment values for each of the three dimensions, and bar charts (Figure 6) and bubble charts (Figure 7) were plotted. Based on the analysis of GO results, Reactive oxygen metabolism, response to lipopolysaccharide, response to bacterial-derived molecules, cellular response to chemical stress, regulation of reactive oxygen metabolism, epithelial cell proliferation, response to drugs, response to metal ions, response to steroid hormones, exogenous apoptotic signaling pathways; Cellular components mainly included: membrane raft, membrane microstructure, membrane lateral region, cell cavity, cell membrane, organelle outer membrane, outer membrane, mitochondrial outer membrane, myelin sheath and neuronal projection cytoplasm. Molecular functions mainly include: nuclear receptor activity, ligand-activated transcription factor activity, DNA-binding transcription factor binding, RNA polymerase II-specific DNA-binding transcription factor binding, steroid hormone receptor, etc.
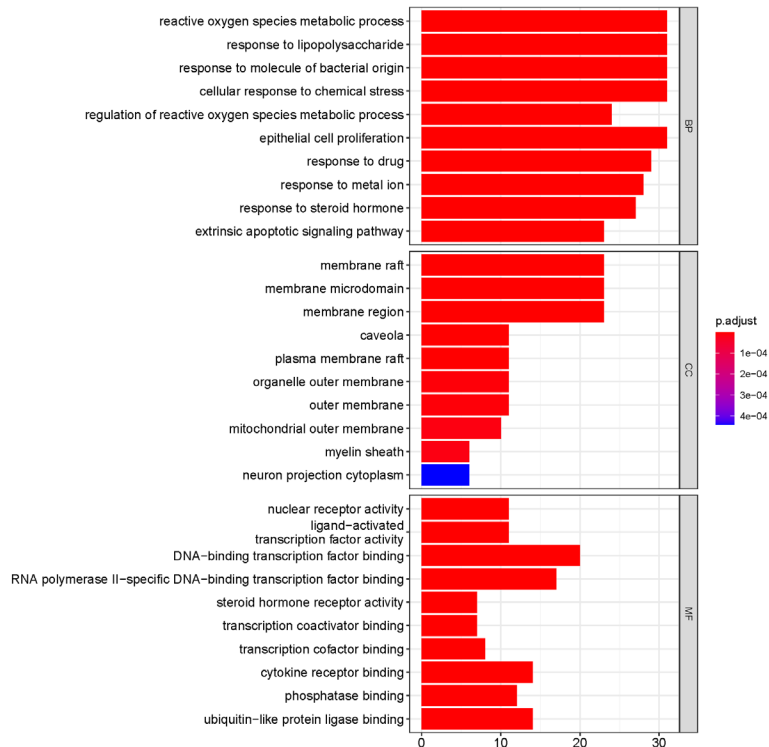
Figure 6 Histogram of GO enrichment of DPN targets by frankincense-myrrh treatment.
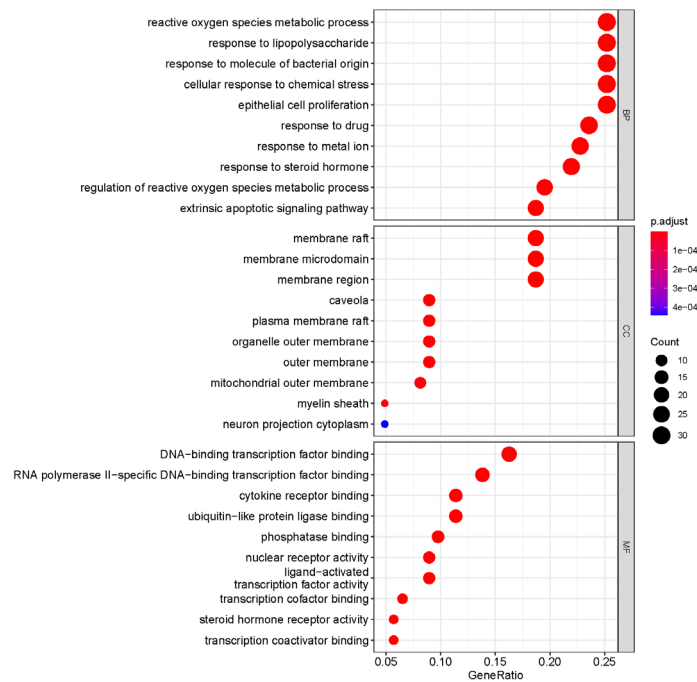

Figure 7 Bubble diagram of GO enrichment of DPN targets by frankincense-myrrh treatment.

The drug-disease shared targets were subjected to KEGG pathway enrichment analysis, citing the String database, and the items with corrected P-value < 0.05 were filtered, and a total of 166 signaling pathways were enriched. The top 20 KEGG pathways were selected for bar graph plotting (Figure 8) and bubble plots (Figure 9) using R 4.0.3 , after installing and referencing the clusterProfiler package. These pathways include, among others, the signaling pathway of AGE-RAGE in diabetic complications, lipids and atherosclerosis, fluid shear stress and atherosclerosis, PI3K-Akt signaling pathway, tumor necrosis factor signaling pathway, chemical carcinogenesis-receptor activation, hepatitis B virus, prostate cancers, Influenza A A, Hepatitis C, and PI3K-Akt signaling pathway. It is hypothesized that frankincense and

myrrh may be involved in the prevention and treatment of painful diabetic peripheral neuropathy through these pathways.
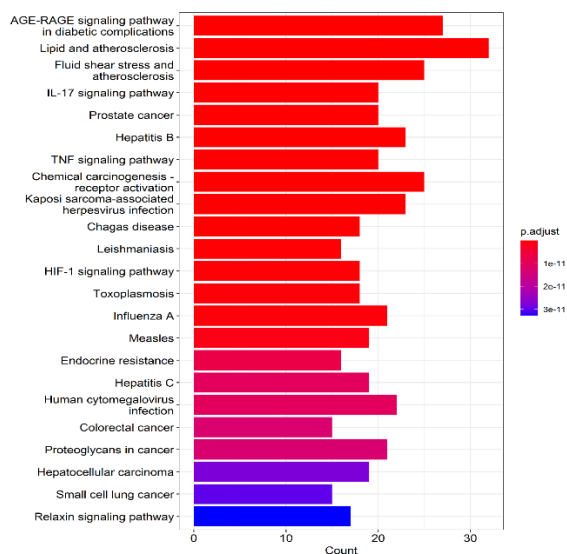

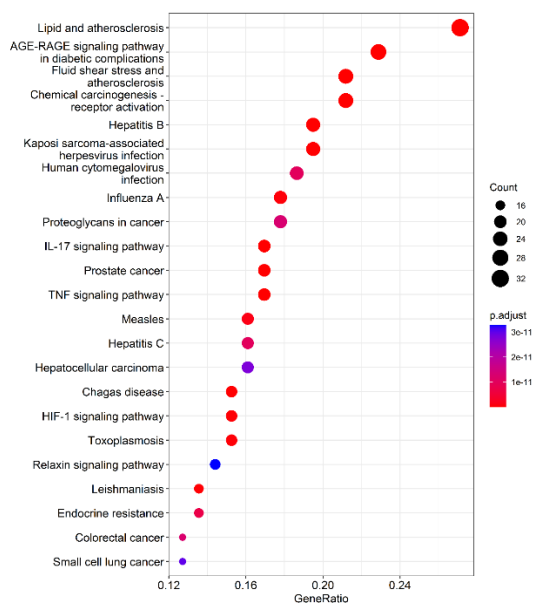*Figure 8 KEGG histogram of PDN targets treated with frankincense and myrrh.*

Figure 9 KEGG bubble plot of PDN targets treated with frankincense and myrrh.

3.7. Component-Disease-Pathway-Target Network Construction

In order to more visually demonstrate the multi-component-multi-target action characteristics of the active ingredients of traditional Chinese medicine in the treatment of painful diabetic peripheral neuropathy. The component-disease-pathway-target network file was imported into Cytoscape 3.8.0 to draw the component-disease-pathway-target network diagram (Figure 10).
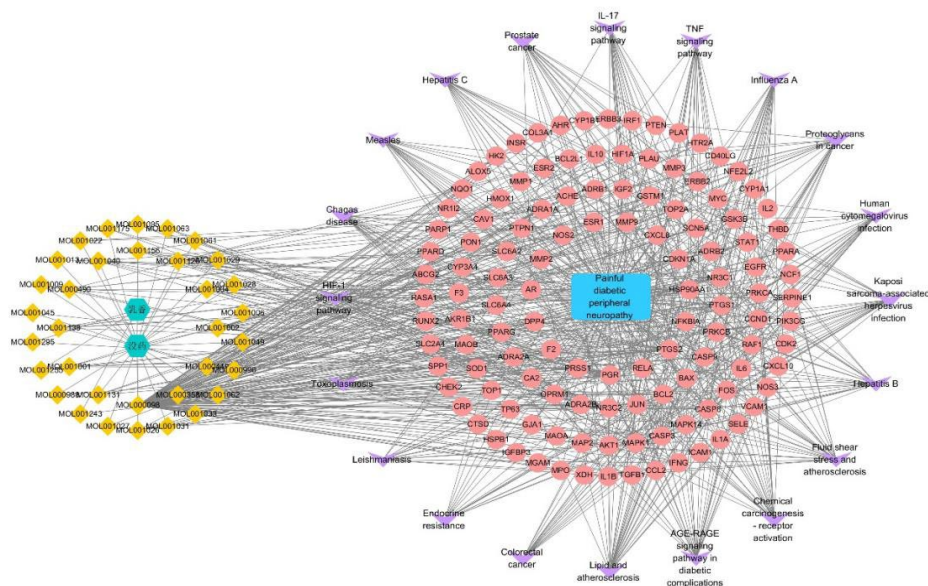
Figure10 Frankincense-myrrh-painful diabetic peripheral neuropathy target-pathway network.

Note: As shown, yellow is the compound, pink is the target of the herbal medicine acting on the disease, purple is the top 20 most significant pathways, blue is the disease i.e. painful diabetic peripheral neuropathy, and green is the herbal medicine.

4. Discussion

Painful diabetic peripheral neuropathy (PDN) is a kind of diabetes-induced peripheral neuropathic pain, the formation mechanism is complex. Its pathogenesis is not only affected by the genetic factors of diabetic patients, but also through metabolic disorders, vascular injury, oxidative stress injury, etc. to affect the patient's blood vessels and nerves, resulting in nerve degeneration and necrosis, destroying the nerve up and down to the receptor, and affecting the nerve conduction speed and sensitivity[14] . In addition, the high glucose state is also an important pain-causing factor, under the high glucose state, a large number of phospholipids accumulate, the brain intestinal peptide p substance is reduced, the patient's sensory neurons are destroyed, affecting the pain threshold, so that patients with diabetic peripheral neuropathy are over-sensitive to pain[15] . Clinical treatment for these pains lacks specificity, mainly through SNRIs, tricyclic antidepressants, anticonvulsants and opioids[16] to reduce the patient's subjective pain and improve the quality of life, the efficacy of the treatment is not good, and most of them will be accompanied by a certain degree of adverse reactions, such as nausea and vomiting, dizziness and drowsiness, skin allergies, etc., and long-term use of the drug will also be addictive and tolerant. As early as Yuan Dynasty, Zhu Danxi's Danxi Xin Fa recorded that the symptoms such as "dry and thin legs and knees, tired and painful bone joints" would appear after a long time of wasting-and-thirst, and the cognition of painful diabetic peripheral neuropathy was long. Wang Xugao Syndrome Medical Records of the Ming Dynasty also had a record that "the hands and feet were numb and cold as ice after a long time of wasting-and-thirst". "Dry mouth of Xiaoshen, dry eyes and imbecility of Yin and flapper, and irritation and pain of hands and feet" and so on. It is believed that numbness and irritation of hands and feet are complications of long-time wasting-and-thirst. Modern medical doctors combined with the clinical manifestations of this disease, most of them will be categorized in traditional Chinese medicine thirst for a long time secondary to "paralysis" "pain" category, the blood is not through the pain, not to the paralysis. The blockage of veins and channels is the main pathogenesis of paralysis of thirst, resolving blood stasis, activating blood circulation and relieving pain is the main treatment principle of paralysis of thirst.

Frankincense and myrrh were first introduced into China as spices. As plant resins, both have the effect of activating blood circulation and eliminating stasis to relieve pain, while frankincense is good at moving qi and myrrh is good at dispersing blood stasis, and the combination of the two has the best effect in relieving pain. It is found that frankincense and myrrh are most frequently used in analgesic Chinese medicines, and their effects are most complete and widely used when they are combined in a 1:1 ratio[17] . The main components of frankincense are pentacyclic triterpenoids, tetracyclic triterpenoids and

macrocyclic diterpenoids, while myrrh mainly contains terpenoids, steroids, flavonoids and lignans. Now pharmacological studies have confirmed that a large number of terpenes and volatile oils in frankincense and myrrh are the main active substances in anti-inflammatory and analgesic properties. The active ingredient of Boswellia serrata, 3-carbonyl-glycero-8,24-diene-21-carboxylic acid (KTDA), and the active ingredient of myrrh, 2-methoxy-5-acetyl-furangimane-1(10)-en-6-one (FSA), have strong anti-inflammatory and analgesic effects when used individually or when paired together.[18] , and the volatile oils and some of the extracts of myrrh also have a good analgesic effect on peripheral pains.[19] . The combination of the two is essential to get twice the result with half the effort. In this paper, the active ingredients and targets of frankincense and myrrh were collected from TCMSP[1] database, and it was found that the main active ingredients of frankincense were compound such as inoceramol, 3-oxo-tirucallic acid, acetyl-alpha boswellic acid, boswellic acid, tetracyclic triterpenoids and other compounds[20] , and that the main active ingredients of myrrh were compounds such as quercetin 3-glucuronide, ellagic acid, and vinpocetine. The interactions of frankincense and myrrh pairs with PPI proteins in painful diabetic peripheral neuropathy showed that AKT1, IL-6, CASP3, IL-1β, and JUN were the key targets of frankincense and myrrh in the treatment of PDN, and the GO functional enrichment results showed that the cellular reactive oxygen species metabolism process and response to lipopolysaccharides were the most significant signaling pathways, and the pathway that was most significantly expressed by the KEGG enrichment results was the lipid-arteriolar atherosclerosis signaling pathway. The most significant pathway expressed in the KEGG enrichment results was the lipid and atherosclerosis signaling pathway. The results of GO and KEGG enrichment suggest that Boswellia myrrha may inhibit the development of PDN at the cellular level through glycolipid metabolism, oxidative stress, and inflammation. Hyperlipidemia contributes to the increase of oxidized LDL cholesterol and free fatty acids, which increases inflammatory factors or enhances inflammatory response signaling in vivo, and the high glucose state leads to the increase of advanced glycosylation end products, which in turn causes chronic low-grade inflammation. Long-term chronic inflammatory infiltration will gradually destroy neurons, glial cells and vascular endothelial cells, etc., and eventually lead to the occurrence of PDN[21] , frankincense, myrrh and its main active substances have a strong antioxidant capacity, which can reduce the content of ROS generated by H_2O_2 induced in the HaCaT cells[22] , and then reduce the level of inflammatory factors in the body. Some studies have confirmed that[23] different ratio of frankincense-myrrh combination can reduce the expression of related neuroinflammatory factors (IL-1β, IL-6, nNOS), control neuroinflammation, improve neurological scores, and have a strong protective effect on nerve cells and functions, in addition to finding that the use of the two together can inhibit the growth of cancer cells[24] , and also has a good effect on breast hyperplasia[25] , it is a safe and effective and has good therapeutic effect on painful diabetic peripheral neuropathy. It is a safe and effective anti-inflammatory analgesic with good therapeutic effect on painful diabetic peripheral neuropathy.

5. Conclusions

Eight components and 11 targets of frankincense compounds were preliminarily found by network pharmacological technology. There were 45 bioactive components and 184 targets in myrrh. There were 2265 PDN related genes and 123 common targets of drugs and diseases. Mainly through Recombinant Nitric Oxide Synthase 2 (NOS2), factor II (F2), Recombinant Prostaglandin Endoperoxide Synthase 2 (PTGS2), Dipeptidyl peptidase-4 (DPP4), Recombinant Protease, Serine 1 (PRSS1), estrogen receptor1 (ESR1) and other key targets to treat PDN, The AGE-RAGE signaling pathway, lipid and atherosclerosis, fluid shear stress and atherosclerosis in diabetic complications are the most likely pathways of frankinsia and myrrrhiza in the treatment of PDN.

References

[1] Kioskli K, Scott W, Winkley K, et al. Psychosocial Factors in Painful Diabetic Neuropathy: A Systematic Review of Treatment Trials and Survey Studies [J]. Pain Med. 2019 Sep1；20(9): 1756-1773.
[2] Vinik AI. Diabetic Sensory and Motor Neuropathy [J]. N Engl J Med, 2016, 374(15): 1455 -1464.
[3] Iyer S, Tanenberg RJ. Pharmacologic management of diabetic peripheral neuropathic pain. Expert Opin[J]. Pharmacother, 2013, 14: 1765 ~ 1775
[4] Butler S, Eek D, Ring L, et al. The utility/futility of medications for neuropathic pain - an observational study [J]. Scand J Pain. 2019 Apr 24；19(2): 327-335.
[5] Gao R, Miao X, Sun C, et al. Frankincense and myrrh and their bioactive compounds ameliorate the multiple myeloma through regulation of metabolome profiling and JAK/STAT signaling pathway based

on U266 cells [J]. BMC Complement Med Ther. 2020 Mar 23; 20(1): 96.

[6] CHEN Hai-Bin, ZHOU Hong-Guang, LI Wen-Ting, et al. Network pharmacology-a new perspective on the mechanism of action study of Chinese medicine compounding[J]. Chinese Journal of Traditional Chinese Medicine, 2019, 34(07): 2873-2876.

[7] Ru J, Li P, Wang J, et al. TCMSP:a database of systems pharmacology for drug discovery from herbal medicines. J Cheminform. 2014 Apr 16：6: 13.

[8] The UniPort C T. UniProt: The universal protein knowledgebase[J]. Nucleic Acids Res, 2017, 45(D1): D158-D169

[9] Safran M, Dalah I, Alexander J, et al. GeneCards Version 3: the human gene integrator. database : the journal of biological databases and curation 2010, 2010: baq020. 2010: baq020.

[10] Sayers EW, Beck J, Bolton EE, et al. Database resources of the National Center for Biotechnology Information. Nucleic Acids Res. 2021 Jan 8;49(D1):D10-D17.

[11] Amberger JS, Bocchini CA, Schiettecatte F, et al. org: Online Mendelian Inheritance in Man (OMIM®), an online catalog of human genes and genetic disorders. Nucleic acids research 2015, 43(Database issue): D789-798.

[12] Doncheva NT, Morris JH, Gorodkin J, et al. Cytoscape StringApp: Network Analysis and Visualization of Proteomics Data. journal of proteome research 2019, 18(2): 623-632.

[13] Szklarczyk D, Gable AL, Lyon D, et al. STRING v11: protein-protein association networks with increased coverage, supporting functional discovery in genome-wide experimental datasets. nucleic acids research 2019, 47(D1): d607-d613.

[14] LI Ying, Leng Jinhong. Progress of research on the pathogenesis of painful diabetic peripheral neuropathy[J]. Journal of Liaoning University of Traditional Chinese Medicine, 2017, 19(11): 117-121.

[15] Frøkjaer JB, Søfteland E, Graversen C, et al. Effect of acute hyperglycaemia on sensory processing in diabetic autonomic neuropathy[J]. Eur J Clin Invest, 2010, 40(10): 883 ~ 886.

[16] Bril V, England J, Franklin GM, et al. Evidence-based guideline: Treatment of painful diabetic neuropathy: report of the American Academy of Neurology, the American Association of Neuromuscular and Electro diagnostic Medicine, and the American Academy of Physical Medicine and Rehabilitation[J]. Neurology, 2011, 76: 1758 ~ 1765.

[17] WANG Nan, GU Mengjia, YU Juanjuan, et al. Development of analgesic theory in traditional Chinese medicine and exploration of analgesic prescription medication law based on support analysis[J]. Sichuan Traditional Chinese Medicine, 2018, 36(01): 40-42.

[18] ZHAO Li-Hui, ZHAO Zi-Zhang, LI Jia-Zhou , et al. Study on the anti-inflammatory and analgesic effects and mechanism of active ingredients KTDA and FSA of frankincense and myrrh and their combinations[J]. New Chinese Medicines and Clinical Pharmacology, 2022, 33(11): 1460-1465.

[19] Wang Tuanjie. Research on the effect substance basis of myrrh in the treatment of primary dysmenorrhea [D]. Jiangsu University, 2009.

[20] LI Qiang, ZHANG Qingmin, LIU Qiang, et al. -Analysis of the effect of comprehensive treatment of knee osteoarthritis by various clinical means[J]. China Modern Drug Application, 2013, 7(3): 39-40.

[21] Yang K, Wang Y, Li YW, et al. Progress in the treatment of diabetic peripheral neuropathy. Biomed Pharmacother. 2022 Apr：148: 112717.

[22] Chen Wanning. Research on the extraction mode and antioxidant efficacy of Boswellia serrata extract [D]. Shanghai Jiaotong University, 2018.

[23] MU Xiaodong. Research on the preparation process of effective parts of frankincense-myrrh allotment and its protective effect on ischemic stroke[D]. Nanjing University of Traditional Chinese Medicine, 2020.

[24] QIN Xuhua, JIN Shenrui, XIAO Hua, et al. Effects of milk myrrh on tumor suppression and receptor-type tyrosine kinase activity[J]. Pharmacology and Clinics of Traditional Chinese Medicine, 2015, 31(06): 122-124.

[25] Ge AQ, Yang KL, Liu LF, et al. Exploring the molecular mechanism of frankincense-myrrh drug pair intervening in breast hyperplasia based on network pharmacology[J]. World Science and Technology-Modernization of Traditional Chinese Medicine, 2020, 22(04): 914-925.