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桑菊饮与德尔塔病毒

——基于网络药理学与分子对接探究桑菊饮治疗德尔塔感染机制

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摘　要

背景：桑菊饮在临床上通常用于治疗感冒、肺炎和其他类似疾病，也在治疗由德尔塔变体引起的新型冠状病毒感染中发挥了重要作用。目的：通过网络药理学和分子对接探索桑菊饮治疗德尔塔变体引起的新型冠状病毒感染的药理学机制。方法：在中药系统药理学数据库和分析平台(TCMSP)获取桑菊饮处方的有效成分和药物相关靶点，从GeneCards数据库获取德尔塔病毒相关靶点，利用CytoScape3.8.2软件和STRING数据库构建成分－靶点网络与蛋白－蛋白作用网络，通过基因本体论和京都基因百科全书呈现基因富集分析结果以及潜在信号通路。基于分子对接证实主要活性成分与病毒靶点存在相互作用。结果：最终收集到169种桑菊饮有效成分和6241个德尔塔病毒相关靶点，通过分析发现蛋白－蛋白作用网络的核心靶点JUN、MAPK3、STAT3和RELA与调节感染和转录通路关系紧密。GO和KEGG分析也揭示了PI3K-Akt、AGE-RAGE、cAMP、趋化因子和转录失调通路也许是桑菊饮治疗德尔塔病毒感染的核心通路。分子对接结果显示，桑菊饮核心活性成分与德尔塔病毒靶点ACE2、SARS-CoV-2　3CL具有良好的结合亲和力。结论：桑菊饮可通过多通路、多靶点控制德尔塔变体侵袭组织并控制细胞因子风暴的形成。

关键词

新型冠状病毒，德尔塔变种，桑菊饮，网络药理学，分子对接

Sangju Yin and Delta Variant

—Potential Mechanism of Sangju Yin for the Treating Delta Variant Based on Network Pharmacology and Molecular Docking

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Abstract

Background: Sangju Yin (SJY) is often used to treat colds, pneumonia and other similar diseases in clinical, and also plays a key role in the treatment of severe coronavirus disease 2019 caused by the Delta variant. Objective: This study was purposed to explore the pharmacological mechanism of SJY treating the Delta variant through integrating network pharmacology and molecular docking. Methods: The effective ingredients and related targets of SJY were found in the Traditional Chinese Medicine Systems Pharmacology (TCMSP) Database. The Delta-related targets were discerned from the GeneCards database. Compound-target and protein-protein interaction networks were established through CystoScape software 3.8.2 and STRING Software. Gene ontology and the Kyoto Encyclopedia of Genes and Genomes were used to perform the enrichment of genes and potential signal pathways. Molecular docking was used to confirm the main active ingredients that interact with the viral hub targets. Results: 169 active compounds of SJY, and 6241 corresponding targets related to delta were collected. The analysis revealed that hub modules of the PPI network, including JUN, MAPK3, STAT3 and RELA, were closely associated with regulating inflammation and transcription pathways. And GO and KEGG analyses also revealed that PI3K-Akt, AGE-RAGE, cAMP, chemokine and transcriptional misregulation may be a central factor for Sangju Yin to treat COVID-19. The results of molecular docking showed that core active ingredients have good binding affinities with ACE2 and SARS-CoV-2 3CLpro. Conclusion: Sangju Yin could control the process of Delta variant invading tissues and the forming of cytokine storms through multiple channels and multiple targets.

Keywords

COVID-19, Delta Variant, Sangju Yin, Network Pharmacology, Molecular Docking

1. 引言

新型冠状病毒感染是全球范围内威胁人类生命健康的主要传染病种类之一。据报道，截止到 2023 年 1 月 6 日，在超过 200 个国家或地区新型冠状病毒感染人数高达六亿多人，死亡人数已达六百多万人(Home-Johns Hopkins Coronavirus Resource Center (jhu.edu))。尽管近期各个国家对该流行病管控放开后该数据统计偏小，但其致病率和致死率依然惊人。随着新型冠状病毒的不断变异，逐渐出现了许多关注变种，如 Alpha、Beta、Gamma、Delta、Omicron 等。自 2021 年 5 月 21 日至 6 月 18 日，中国广东省共发现 167 例德尔塔(Delta)变异病毒感染病例[1]。自从德尔塔变种(B.1.617.2)首次在印度出现[2]，德尔塔变种的传播也是 2021 年新型冠状病毒感染人数激增的主要原因[3]。研究报道，德尔塔变种的特点是潜伏期短、传

播速度快、病毒载量高，它的潜伏期为 2~4 天，较之前的变种代际传播速度也有所加快[4] [5]。尽管后来被奥密克戎变种替代流行，但据报道，依然有可能爆发德尔塔变种感染的可能[6]。此外，与奥密克戎变种相比，德尔塔变种的住院率和致病率更高[7]。因此，新型冠状病毒大流行逐渐平息之后，依然需要警惕德尔塔变种的反扑。目前，依然缺乏治疗新型冠状病毒感染的特异性药物，且随着新型冠状病毒变体的增加，疫苗的有效性正在下降[8]，接种疫苗后感染新型冠状病毒的患者也比比皆是[9] [10]。因此，寻找治疗 COVID-19 的有效药物是非常有必要的。在治疗新型冠状病毒感染的浪潮中，中医中药表现的尤为突出。其中出自《温病条辩》的桑菊饮主要用于治疗风热犯肺症，具有抗炎、抗感染、抗病毒等作用，且在治疗 COVID-19 德尔塔变种的长期实践中，桑菊饮被推荐用于预防和治疗轻度 COVID-19 [11] [12]。但中药成分复杂，难以通过单一成分分析其作用机制，通过网络药理学及分子对接可以将中药处方复杂的作用机制变为多途径的信号通路展示。本文旨在研究桑菊饮治疗德尔塔病毒感染的机制，为其临床应用提供一定理论基础，相关研究方法及分析步骤如图 1 所示。

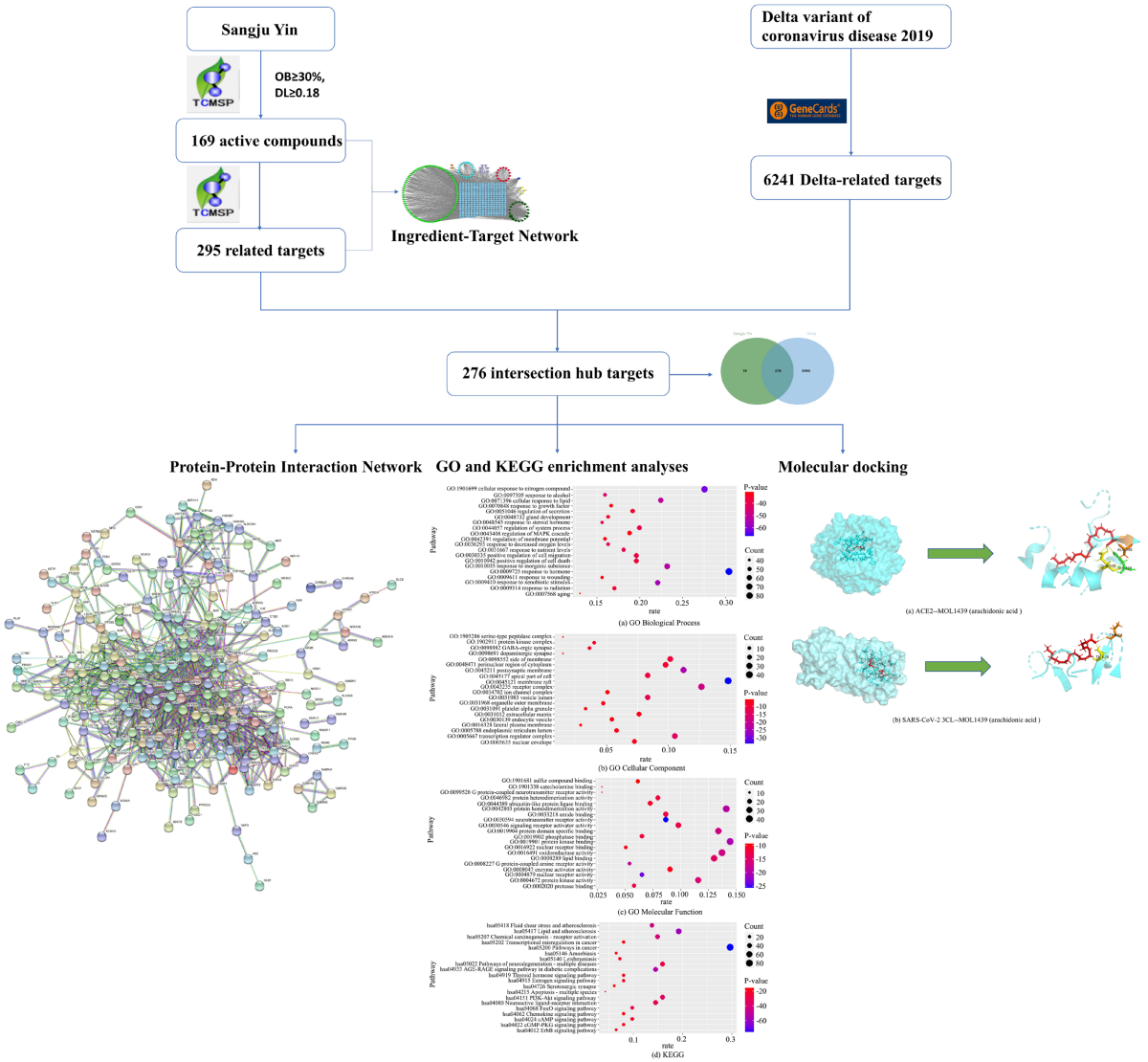


Figure 1. Graphical abstract
图 1. 图片摘要

2. 方法

2.1. 筛选活性成分

桑菊饮活性成分的获取源自中药系统药理学数据库和分析平台(TCMSP: https://old.tcmsp-e.com/)。药物成分的药代动力学特性(ADME)，包括吸收、分布、代谢和排泄，是决定药物生物活性的关键因素，在ADME 相关参数中，口服生物利用度(OB)是重要参数之一，它是指口服药物进入体循环的百分比。另一个指标是药物相似性(DL)，指的是药物库数据库中化合物与临床使用药物之间的结构相似性。本研究选用以上两个参数筛选桑菊饮主要活性成分，筛选标准为 OB ≥ 30%，DL ≥ 0.18 [13]。

获取活性成分后，从 TCMSP 数据库中查找成分相应的靶点。靶标的官方基因编号通过设置“Homo sapiens”限定物种，从 UniProt (https://www.uniprot.org/)数据库中获得。然后从 GeneCards 数据库中通过搜索关键字“Delta”检索已知的与德尔塔病毒相关的基因，将得到的成分靶点基因与病毒基因通过 Excel 软件筛选出两者共同的基因。最后，利用 Venny (http://jvenn.toulouse.inra.fr/app/example.html)工具对二者的基因重合性进行绘图展示。

2.2. 相关网络构建与分析

通过构建成分-靶点(C-T)网络和蛋白-蛋白相互作用(PPI)网络，揭示桑菊饮治疗德尔塔病毒感染的分子机制。找到桑菊饮的药物靶点和德尔塔病毒致病靶点的共同基因，得到桑菊饮治疗德尔塔病毒的潜在基因。PPI 网络是通过将这些潜在基因导入 STRING (https://cn.string-db.org/cgi/input?sessionId=bbX0BOz6VIW8&input_page_active_form=multiple_identifiers)数据库，设置置信度得分默认高于 0.9，物种限制为“智人”并进行分析而得到。将得到的 PPI 网络保存为“TSV”格式，上传到 Cytoscape 3.8.2 软件(https://cytoscape.org/)进行后续处理，利用 Cytoscap 软件插件 CytoNCA 计算网络中蛋白的度中心性(DC)、间性中心性(BC)、紧密性中心性(CC)、特征向量中心性(EC)和基于局部平均连通性的方法(LAC)，上述拓扑性质与网络中靶点的重要性相一致，从而筛选出桑菊饮治疗德尔塔病毒感染的核心基因。

2.3. 富集分析

将二者的共同基因导入 Metscape 网站(http://www.metascape.org/gp/index.html#/main/step1)，通过调整 p-value < 0.05，得到基因本体论(GO)分析和京都基因与基因组百科全书(KEGG)分析结果。将结果导出应用 R 包(https://www.rstudio.com/products/rstudio/)进行可视化处理。

2.4. 分子对接

在处方所有的活性成分中，Degree 值大于其两倍中位数的活性成分被筛选出来作为分子对接的小分子。分子结果是在 TCMSP 数据库中检索并保存为 mol2.文件，建立小分子数据库。分子对接的大分子蛋白质则是 ACE2 和 3CLpro 水解酶，二者是新型冠状病毒的重要致病靶点，其蛋白质结构可从 UniProt (https://www.uniprot.org/)和 RCSM PDB (https://www.rcsb.org/)数据库中找到，并保存为 PDB 格式文件。最终，这些文件被上传到 SYBYL-X 2.1.1 软件进行分子对接和分析，根据其参数分析其在实际环境中相互作用的潜在能力。

3. 结果

3.1. 桑菊饮治疗靶点与德尔塔病毒致病靶点

桑菊饮由桑叶、菊花、连翘、芦根、杏仁、桔梗、甘草、薄荷共 8 种中药按一定比例组成。通过设

定OB和DL值，从TCMSP数据库中筛选出169个有效成分和295个相关靶点(图2)。其中，MOL98 (querceti，槲皮素)和MOL0422 (kaempferol，山奈酚)为桑叶、菊花、连翘和甘草的常见成分。MOL06 (luteolin，木犀草素)是菊花、连翘、桔梗、薄荷的常用成分。MOL0449 (stigmasterol，豆固醇)是桑叶、芦根、杏仁的常见成分。MOL0359 (sitosterol，谷甾醇)是杏仁、甘草和薄荷的共同成分。MOL0211 (mairin，山柰素)是连翘、连翘、甘草的常用成分。MOL0358 (beta-sitosterol，β-谷甾醇)是桑叶、菊花、连翘的常用成分。MOL1689 (acacetin，金合欢素)是菊花、桔梗和薄荷的常用成分。MOL4328 (naringenin，柚皮素)是菊花、甘草、薄荷的常用成分。MOL2311 (glycyrol，甘草酚)、MOL4841 (licochalcone B，甘草查尔酮B)、MOL4903 (liquiritin，甘草苷)、MOL4908 (glabridin，甘草黄酮)、MOL5017 (phaseol，菜豆醇)是杏仁和甘草的常见成分。MOL4355 (spinasterol，菠菜甾醇)是杏仁和桔梗的常见成分。MOL1771 (poriferast-5-en-3beta-ol，γ-谷甾醇)是菊花和菊花的常用成分。MOL1790 (linarin，蒙花苷)、MOL2881 (diosmetin，香叶木素)是菊花和薄荷的常用成分。MOL0354 (isorhamnetin，异鼠李素)是菊花和甘草的常见成分。

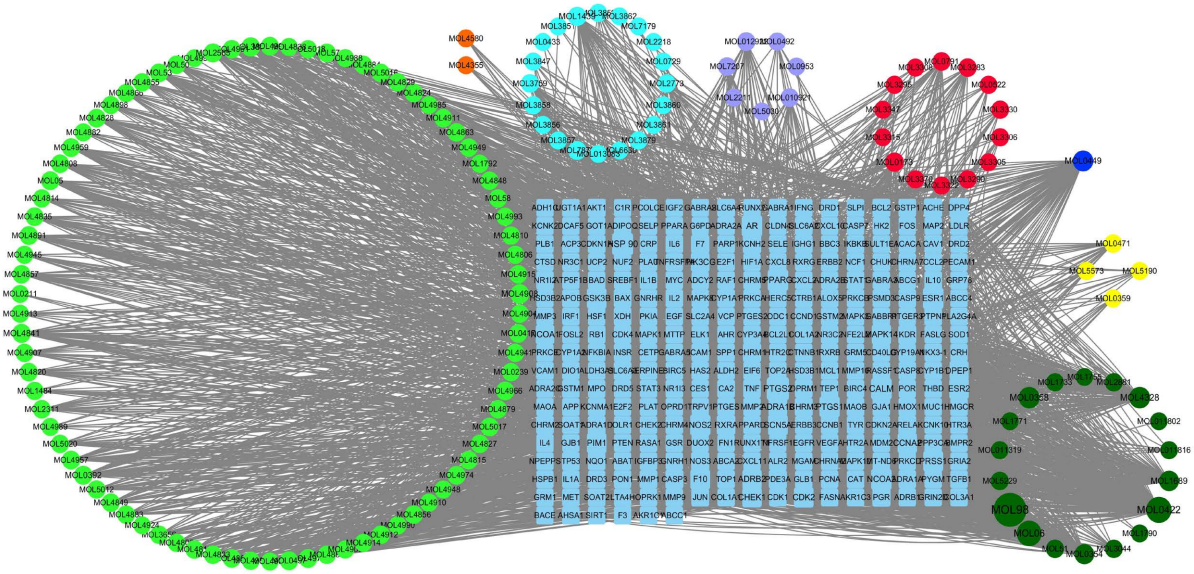


Figure 2. Ingredient-Target Network. There are 149 active ingredients, including Mori Follum (bule round nodes), Chrysanthemi Flos (dark green nodes), Forsythiae Fructus (red nodes), Phragmitis Rhizoma (dark blue nodes), Amygdalus Communis Vas (pruple nodes), *Platycodon grandiforus* (orange nodes), Licorice (green nodes), Menthae Herba (yellow nodes), and there are 19 duplicate compounds, 3 of them are found in 4 kinds of Chinese medicine, 6 of them are found in 3 kinds of Chinese medicine, 10 of them are found in 2 kinds of Chinese medicine. In addition, bule rectangular nodes represent 295 related targets of compounds

图2. 成分－靶点网络(C-T网络)共有149个活性成分，包括桑叶(浅蓝色圆点)、菊花(深绿色圆点)、连翘(红色圆点)、芦根(深蓝色圆点)、杏仁(紫色圆点)、桔梗(橙色圆点)、甘草(绿色圆点)、薄荷(黄色圆点)，共有19种重复成分，其中3种存在于4种中药，6种存在于3种中药，10种存在于2种中药。此外，蓝色方形节点表示活性成分的295个相关靶点

利用Cytoscape 3.8.2软件构建成分-靶点(C-T)网络(图2)，在C-T网络中，节点表示成分和靶点，边表示二者之间存在相互作用。然后从GeneCards数据库中获得6241个疾病相关基因。通过检索，德尔塔病毒和桑菊饮处方共276个交集基因，如图Venny所示(图3)。

3.2. 网络分析结果与核心基因

通过设置可信度 > 0.9，利用STRING数据库构建了核心靶点的蛋白-蛋白相互作用网络。该PPI网络包含276个节点和1107条边，平均度数节点为8.05，平均局部聚类系数为0.435 (如图4所示)。将PPI

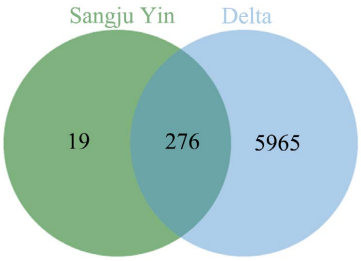


Figure 3. Intersection analysis of genes between Sangju Yin and Delta. Sangju Yin includes 295 related targets, and delta includes 6241 disease-related targets, of which 276 targets are common to both
图 3. 桑菊饮治疗靶点与德尔塔病毒的致病靶点。桑菊饮共有 295 个相关靶点，德尔塔病毒共有 6241 个疾病相关靶点，而二者共同靶点共有 276 个

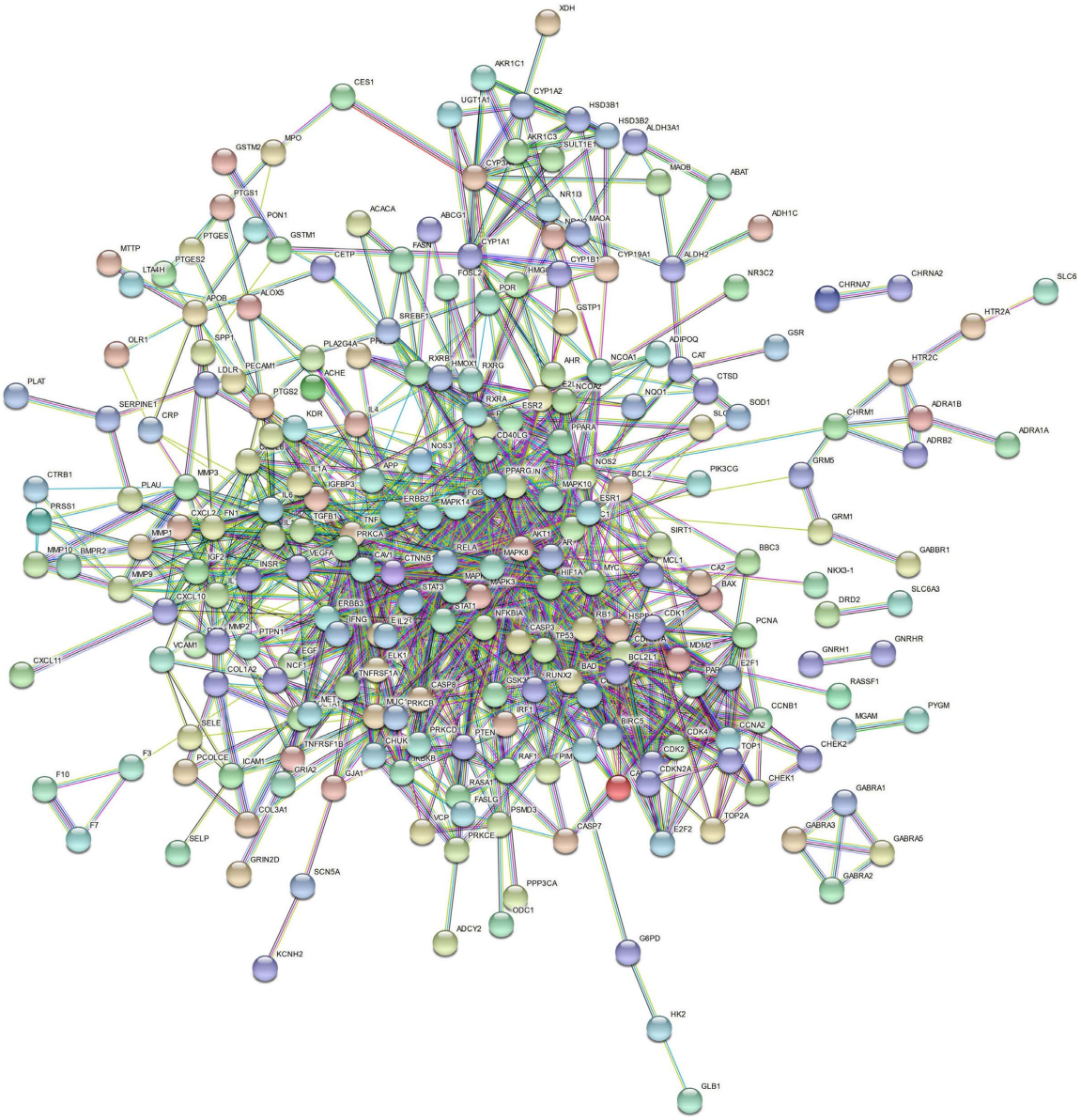


Figure 4. Protein-protein interaction network output from STRING database
图 4. STRING 数据库导出的蛋白－蛋白相互作用网络

网络保存为“TSV”格式后，导入到 Cytoscape 3.8.2 软件中，将 STRING 数据库结果的靶点进行评分可视化。通过网络分析，根据得到的 Degree 值和综合得分，设定靶点和连线的颜色和大小(图 5(a))。然后通过相关参数对 PPI 网络进行两次筛选，得到关键基因。通过设置 Degree 值中位数的 2 倍(Degree ≥ 52)进行第一次筛选得到第二个 PPI 网络(图 5(b))。接下来，根据 CytoNCA 工具得到的参数中位数进行 PPI 网络的第二次筛选，这些参数包括 Degree (≥69)、Betweenness (≥3.539285714)、Close (≥0.789473684)、LAC (≥15.66667)、Neighborhood Connectivity (≥20.12005929)。最后得到桑菊饮治疗德尔塔病毒感染的核心基因为 JUN、MAPK3、STAT3 和 RELA (图 5(c))。

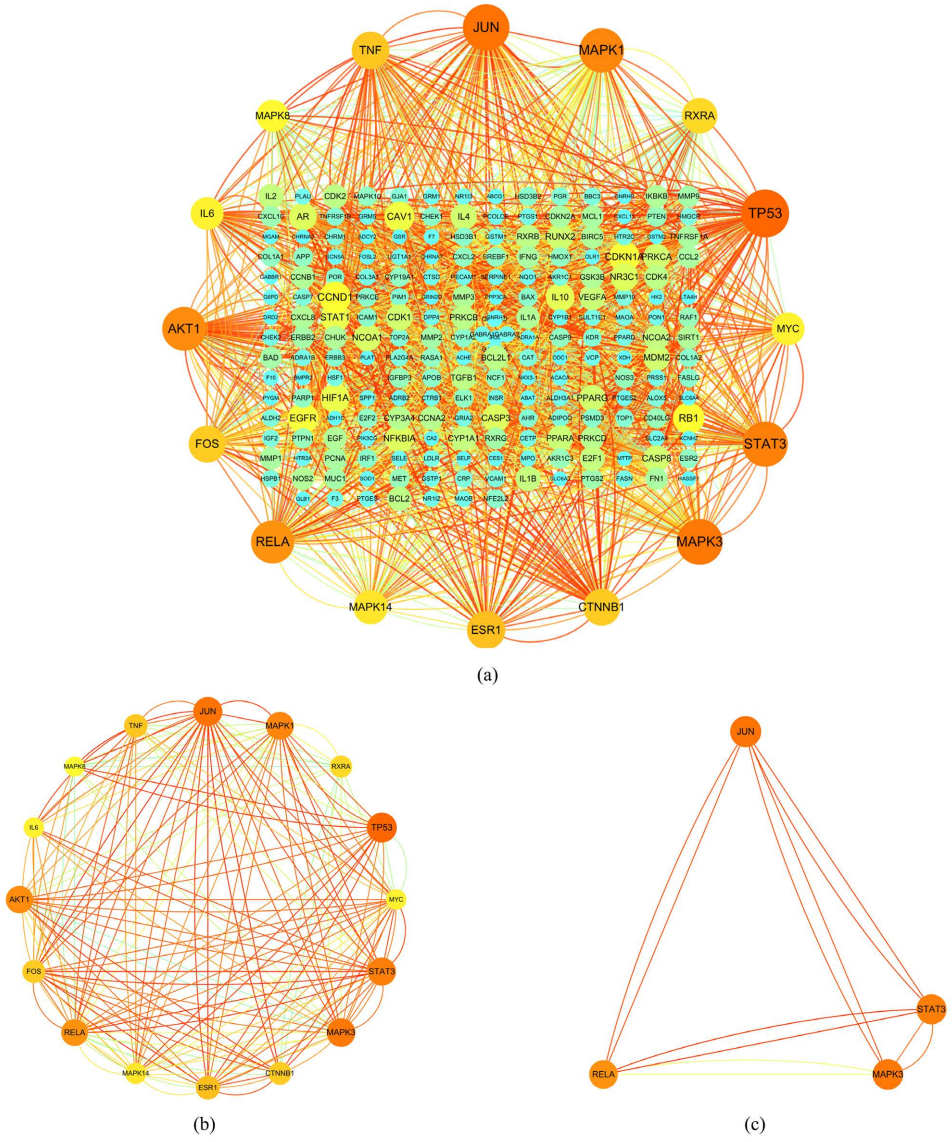


Figure 5. The protein-protein interaction network. The nodes represent target proteins and the lines represent the interaction of two proteins. And the more interacting proteins, the larger the nodes and the darker the color. The more plausible the protein interactions are, the darker the color of the line. (a) PPI Network Performed by CytoScape 3.8.2; (b) The Result of First Filtrating; (c) The Hub Genes, including JUN, MAPK3, STAT3, RELA

图 5. 蛋白－蛋白作用网络(PPI)节点表示靶点蛋白，线条表示两种蛋白之间存在相互作用。相互作用的蛋白质越多，节点越大，颜色越深。蛋白质之间的相互作用可信度越高，线的颜色就越深。(a) 由 CytoScape 3.8.2 软件绘制的 PPI 网络，(b) 第一次筛选后的 PPI 网络，(c) 核心靶点，核心靶点 JUN、MAPK3、STAT3、RELA

3.3. 富集分析结果

为确定桑菊饮治疗德尔塔病毒感染的潜在机制，将交集基因导入 metascape 数据库(https://metascape.org)，设置物种为 H. sapiens，进行 GO 和 KEGG 富集分析。最后，在 P < 0.01 的条件下，得出二者富集分析结果。其中 GO 富集分析结果共包括三个部分，分别是生化过程(Biologicalprocess，BP)、细胞成分(Cellularcomponent, CC)和分子功能(Molecularfunction, MF)。在 6420 个生化过程项目中，富集程度最高的前 20 个项目分别与激素、氧水平、营养水平有关(图 6(a))，细胞成分共有 604 项，CC 富集前 20 名分别为膜筏、突触后膜、受体复合物(图 6(b))。在 1155 项分子功能中，MF 富集前 20 位的是神经递质受体活性、核受体活性、蛋白激酶结合(图 6(c))。此外，KEGG 通路注释显示，PI3K-Akt、AGE-RAGE、cAMP、趋化因子和转录失调通路通路均排在前 20 位(图 6(d))。

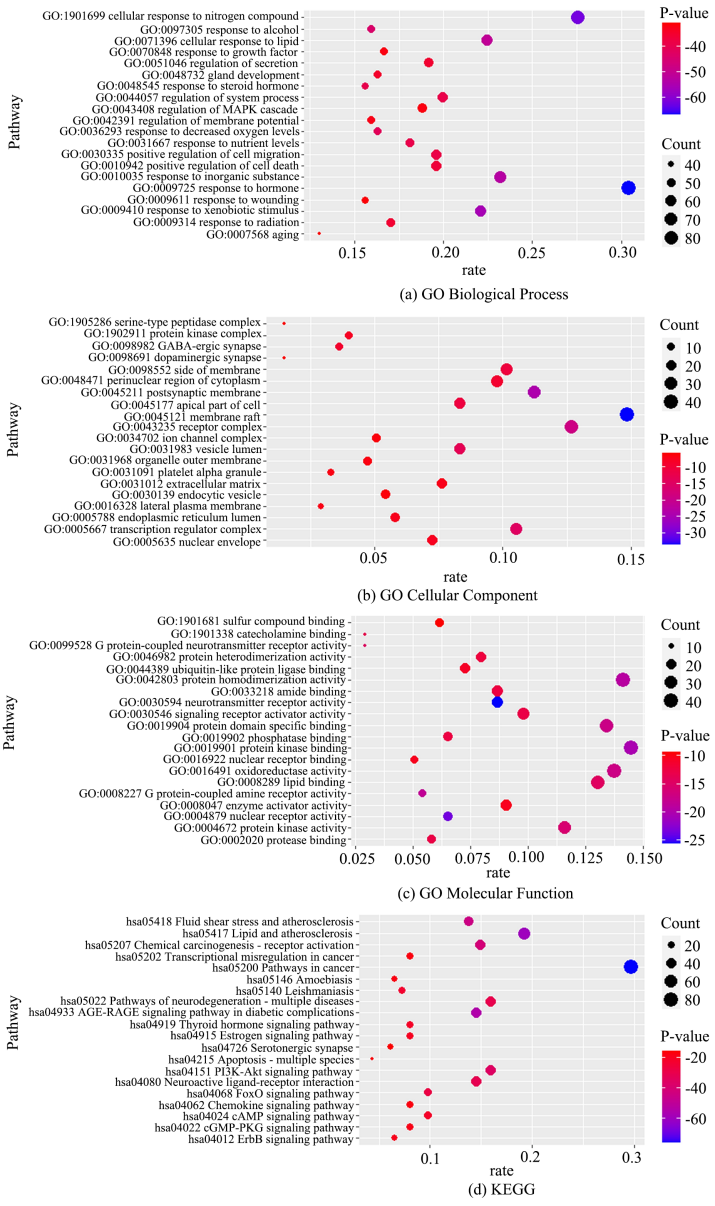


Figure 6. The results of GO and KEGG enrichment analysis
图 6. GO 和 KEGG 富集分析结果

3.4. 分子对接分析结果

最终，桑菊饮处方的 15 种核心成分分别为花生四烯酸、β-谷甾醇、甘草查尔酮 B、豆甾醇、木犀草素、苜蓿素、异鼠李素、刺槐素、槲皮素、刺芒柄花素、山柰酚、汉黄芩素、光甘草定、7-甲氧基-2-甲基异黄酮和柚皮素(表 1)。分子对接靶点蛋白结构则是从 Uniprot 和 RCSM PDB 数据库中选择高分辨率、小分子量作为对接配体。在 Sybyl X2.1.1 软件中与 ACE2 (1r4l)和 SARS-CoV-2 3CLpro (6lu7)进行分子对接。总分(Totalscore, TS)是由碰撞、极值、相似度和 C_score 得出的综合分数，当 TS ≥ 3 时，说明分子与蛋白质之间具有一定的亲和力。表 2 显示了各组分与蛋白质之间具有一定的结合亲和力。根据此结果，将分子对接结果导入 PyMol 软件，将分子与蛋白质的结合位点可视化处理(图 7)。

Table 1. The information of main ingredients
表 1. 主要成分信息

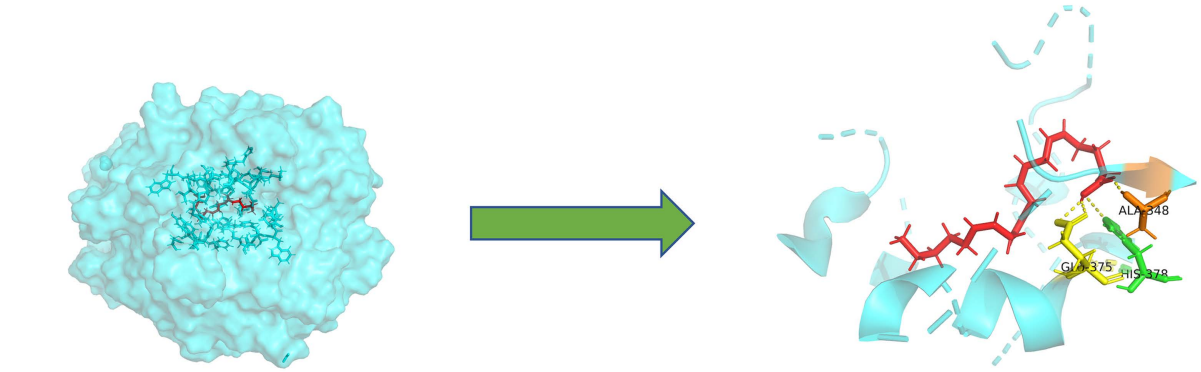
Mol ID	成分名称	中药来源	口服生物利用度 OB (%)	药物相似性 DL (%)
MOL1439	花生四烯酸(Arachidonic acid)	桑叶	45.57	0.2
MOL0358	β-谷甾醇(Beta-sitosterol)	桑叶，菊花，连翘	36.91	0.75
MOL4841	甘草查尔酮 B (Licochalcone B)	杏仁，甘草	76.76	0.19
MOL0449	豆甾醇(Stigmasterol)	桑叶，芦根，杏仁	43.83	0.76
MOL06	木犀草素(Luteolin)	菊花，连翘，桔梗，薄荷	36.16	0.25
MOL2565	苜蓿素(Medicarpin)	甘草	49.22	0.34
MOL0354	异鼠李素(Isorhamnetin)	菊花，甘草	49.6	0.31
MOL1689	刺槐素(Acacetin)	菊花，桔梗，薄荷	34.97	0.24
MOL98	槲皮素(Quercetin)	桑叶，菊花，连翘，甘草	46.43	0.28
MOL0392	刺芒柄花素(Formononetin)	甘草	69.67	0.21
MOL0422	山柰酚(Kaempferol)	桑叶，菊花，连翘，甘草	41.88	0.24
MOL0173	汉黄芩素(Wogonin)	连翘	30.68	0.23
MOL4908	光甘草定(Glabridin)	杏仁，甘草	53.25	0.47
MOL3896	7-甲氧基-2-甲基异黄酮 (7-Methoxy-2-methyl isoflavone)	甘草	42.56	0.2
MOL4328	柚皮素(Naringenin)	菊花，甘草，薄荷	59.29	0.21

Table 2. The results of molecular docking
表 2. 分子对接结果

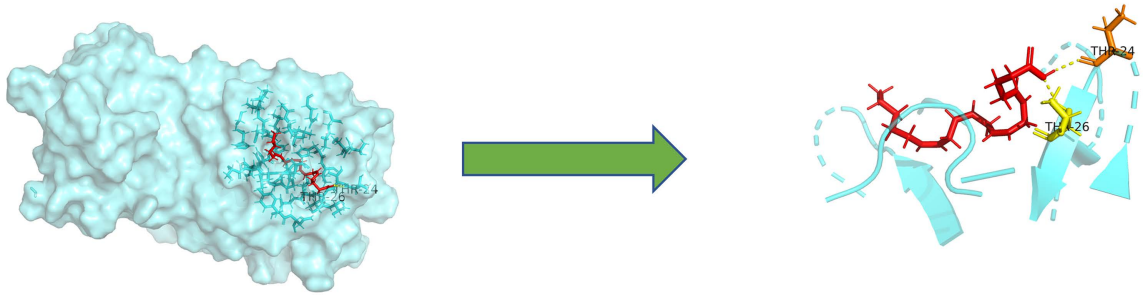
No.	MOL ID	PubChem ID	Name	TOTAL SCORE	
				ACE2	3CLpro
1	MOL1439	444899	花生四烯酸(Arachidonic acid)	10.0006	6.6403
2	MOL0358	222284	β-谷甾醇(Beta-sitosterol)	9.2023	4.6254
3	MOL4841	5318999	甘草查尔酮 B(Licochalcone B)	7.1621	4.7144
4	MOL0449	5280794	豆甾醇(Stigmasterol)	6.7287	4.193

Continued

5	MOL06	5280445	木犀草素(Luteolin)	6.6322	5.0247
6	MOL2565	336327	苜蓿素(Medicarpin)	6.0877	4.3476
7	MOL0354	5281654	异鼠李素(Isorhamnetin)	5.9138	5.9557
8	MOL1689	5280442	刺槐素(Acacetin)	5.5984	4.8989
9	MOL98	5280343	槲皮素(Quercetin)	5.5231	4.8266
10	MOL0392	5280378	刺芒柄花素(Formononetin)	5.4725	3.6613
11	MOL0422	5280863	山柰酚(Kaempferol)	5.4543	4.4783
12	MOL0173	5281703	汉黄芩素(Wogonin)	5.4323	4.3883
13	MOL4908	124052	光甘草定(Glabridin)	5.1793	5.8481
14	MOL3896	354368	7-甲氧基-2-甲基异黄酮(7-Methoxy-2-methyl isoflavone)	4.7083	4.0094
15	MOL4328	439246	柚皮素(Naringenin)	4.4374	4.1014



(a) ACE2--MOL1439 (arachidonic acid)



(b) SARS-CoV-2 3CL--MOL1439 (arachidonic acid)

Figure 7. The results of molecular docking. Red small molecules represent active ingredients, blue structures represent target proteins, and amino acid residues that interact with small molecules are marked with different color
图 7. 分子对接结果：红色结构代表活性小分子成分，蓝色结构代表靶蛋白，与小分子相互作用的氨基酸残基分别用不同颜色标记

4. 讨论

新型冠状病毒感染于 2019 年 12 月在我国武汉爆发[14]，最终在世界范围内流行。这是一种由新型冠

状病毒引起的流行性传染病，曾一度威胁了世界范围内人类的生命安全。有描述性研究显示[15]，该传染病最常见的症状为发热、咳嗽、流涕。尽管新型冠状病毒感染最初表现为流感样症状，但它会通过病程发展大量释放炎症因子引起细胞因子风暴[16] [17]，从而最终导致全身炎症及多器官衰竭[18]。从中医理论来讲，新型冠状病毒感染可以看作是湿、热、淤血引起的疾病，而桑菊饮具有散风清热、散肺止咳的功效，能够解热、抗炎、排汗、抗菌、抑制肠道亢进，是清热解毒的代表方之一，主要用于治疗感冒、急性支气管炎、上呼吸道感染、肺炎、急性结膜炎、角膜炎等疾病。因此，桑菊饮被推荐为治疗 COVID-19 轻症和普通型症状的常用中药处方[11]。

据报道，德尔塔变异株的潜伏期为 2~3 天，而野生株的潜伏期为 3~7 天[4] [5]。这种变化的原因是德尔塔病毒通过刺突蛋白与血管紧张素转换酶-2 (ACE-2)的结合入侵宿主细胞。在病毒变异过程中，刺突蛋白中的氨基酸发生突变，从而增加了它们的结合亲和力[3]。更有流行病学研究报道，德尔塔变异株并未随着奥密克戎变异株的出现而消失，再次暴发感染的可能性很大[6]。目前，关于德尔塔-奥密克戎共突变病毒的报道层出不穷，虽然可信度有待商榷，但不可怀疑的是新型冠状病毒的突变将会继续[19]，因此，研究桑菊饮治疗德尔塔病毒的作用机制更具有重要意义。

作为病毒进入细胞的通道，病毒通过与 ACE2 蛋白结合打破 Ang II 的动态平衡，加重肺损伤，甚至引起细胞因子风暴，导致多器官功能障碍综合征(MODS) [20]，从而危及生命；另一方面，病毒在体内感染的基础在于 SARS-CoV-2 3CLpro 将病毒多聚蛋白裂解为单一蛋白，从而发挥病毒侵袭作用[21]。此外，与病毒突变率较高的 RNA 遗传产物相比，SARS-CoV-2 3CLpro 的编码序列较为保守[22]，因此，以 ACE2 和 SARS-CoV-2 3CLpro 作为靶点寻找药物具有重要意义。

最终，通过网络药理学筛选出 169 个处方活性成分以及 6241 个德尔塔病毒致病靶点。从图 2 可以看出，桑菊饮是通过多成分多靶点多通路来治疗德尔塔病毒感染，而桑菊饮的 15 种核心成分花生四烯酸、*β*-谷甾醇、甘草查尔酮 B、豆甾醇、木犀草素、苜蓿素、异鼠李素、刺槐素、槲皮素、刺芒柄花素、山柰酚、汉黄芩素、光甘草定、7-甲氧基-2-甲基异黄酮和柚皮素均具有抗炎作用[22]-[37]，可以通过不同的途径减轻机体的炎症反应。尽管花生四烯酸是几种炎症因子的前体，但其与两种靶蛋白的结合能力最高，且有文献报道，花生四烯酸可通过其衍生代谢物抑制炎症的形成，并促进炎症因子的分解[38]。此外，从分子对接结果来看，木犀草素、槲皮素、山柰酚、汉黄芩素、光甘草定和柚皮素对 3CLpro 和 ACE2 的亲和力较高，因此推测其具有抗病毒作用[22] [39]-[44]，而这一点不仅在临床前研究中得到证实，在临床实践中也得到印证。上述几种成分是新型冠状病毒肺炎诊疗方案中六种中药处方的共同成分，曾用于治疗 H1N1 等病毒感染且非常有效[45]；而在目前的 COVID-19 治疗中，在一项包含 80 例新冠肺炎病例的研究中，中药治疗组的 7 天病毒清除率显著高于对照组，而肺炎痊愈时间显著缩短[46]。通过两次筛选，桑菊饮治疗德尔塔病毒的关键靶点为 JUN、MAPK3、STAT3 和 RELA，通过 GO 和 KEGG 富集分析最终得到桑菊饮的潜在抗病毒机制。GO 生物过程富集分析结果显示，关键靶点主要参与对氧水平降低、类固醇激素、营养水平、生长因子和损伤等过程的反应过程，而 GO 细胞成分分析结果主要与膜筏、突触后膜、侧膜、核膜、侧质膜、细胞器外膜等膜结合有关，另外，它还与突触和复合体相关，如 GABA 能突触、多巴胺能突触、受体复合体、转录调节复合体、蛋白激酶复合体、离子通道复合体、丝氨酸型肽酶复合体等。GO 分析分子功能分析结果主要与结合功能有关，包括蛋白激酶结合、蛋白结构域特异性结合、脂质结合、儿茶酚胺结合、蛋白酶结合、磷酸酶结合、酰胺结合、泛素样蛋白连接酶结合、核受体结合、硫化合物结合等。此外，还与蛋白活性相关，包括神经递质受体活性、核受体活性、蛋白同源二聚化活性、G 蛋白偶联胺受体活性、氧化还原酶活性、蛋白激酶活性。而 KEGG 富集分析结果显示，桑菊饮治疗德尔塔病毒的信号通路主要为 has04151 (PI3K-Akt 信号通路)、hsa04933 (糖尿病并发症中 AGE-RAGE 信号通路)、hsa04024 (cAMP 信号通路)、hsa04062 (趋化因子信号通路)、hsa05202 (肿瘤中转录异常)，表

明桑菊饮可通过调节炎症和转录来治疗德尔塔病毒感染。

在最终筛选出的关键靶点中，JUN (AP-1)作为转录因子可与 DNA 结合调节病毒在宿主体内的转录[47]，MAPK3 可通过磷酸化参与白细胞介素-1 (IL-1)介导的信号通路，诱导炎性因子的募集和释放，最终形成细胞因子风暴起到抗病毒作用[48] [49]，STAT3 通过去磷酸化激活白细胞介素调节白细胞介素-6 (IL-6)介导的信号通路[50]，也通过乙酰化调节炎症反应[51]。研究表明 STAT3 介导信号通路中的 IL-6 水平与疾病严重程度相关，多数患者治疗前有不同程度的 IL-6 升高，治疗后均较前有所下降[22]。而 RELA 作为细胞因子的关键转录因子参与调节炎症反应[52]。此外，我们发现 JUN (AP-1)、MAPK3 (ERK1)、STAT3 和 RELA 在 hsa05171 (冠状病毒感染) (https://www.genome.jp/pathway/hsa05171) COVID-19 的 KEGG 分析中参与多个信号通路调控细胞因子风暴的形成，这也证明桑菊饮具有控制新型冠状病毒感染发展为重症的可能。在后续的分子对接中，表 2 显示，桑菊饮的核心成分与德尔塔病毒的核心蛋白分子对接结果 Totalscore 均>3 分，表明处方核心成分与病毒核心靶点 ACE、3CLpro 存在一定结合能力，这也在一定程度上为桑菊饮的临床应用提供了理论基础。同时，为了更直观的展示二者的结合能力，图 7 为由 PyMol 软件处理展示的分子对接结果，以便进行进一步研究。

5. 结论

总之，通过网络药理学和分子对接进行分析，桑菊饮可以通过多靶点和多通路治疗德尔塔病毒感染。然而，该结果需进一步实验和临床验证。

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