

Anti-inflammatory Mechanism of Rhein in Treating Asthma Based on Network Pharmacology

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Research

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Abstract

Background

Network pharmacology was used to study Rhein -target-pathway and to clarify its anti-inflammatory mechanism in the treatment of asthma.and provide a new idea for the treatment of asthma

Methods

This method, which allows using network pharmacology to figure out the operational mechanism of Rhein-Target-Pathway, defines the effect of anti-inflammatory in treating asthma. The platform of Traditional Chinese Medicine Molecular Mechanism Bioinformatics, a web server for network, is used to get the corresponding target of Rhein and permit molecular docking. Cytoscape3.7.1, a kind of network software, is used to construct Rhein-predicted target network and analyse network topology. Search anti-inflammatory targets in the database of TTD and then construct the PPI network as well as create protein interaction networks that are combined with the Rhein-predicted target network. The anti-inflammatory targets of Rhein should be presented. The asthma genes of human being can be attained from the database of NCBI Gene Database and construct correspondence vivo response network model. Find Anti-inflammatory targets of Rhein against asthma, screen anti-inflammatory targets of Rhein related with Pathogenesis of asthma. Enrichr database is used to analyse signal pathway from anti-inflammatory targets of Rhein KEGG.

Results

According to the study, Rhein corresponds to 17 target proteins, four anti-inflammatory targets of Rhein related to asthma(MAPK14, EGFR, ERBB2, TNFRSF1A) are probably the most important targets where asthma is treated by Rhein.

Conclusions

These four anti-inflammatory targets of Rhein related to asthma are probably the key targets in the treatment of asthma by using Rhein. For the purpose of preventing the occurrence as well as development of asthma and delaying the progress of the disease, one or some of the four anti-inflammatory targets of Rhein related to asthma can be controlled.

Background

Asthma is abbreviated for Bronchial Asthma. It is a kind of chronic inflammatory disorder of the airways involved of cells and corresponding components, which is characterized by inflammation, hyper responsiveness, stenosis and remodelling of the airway. The chronic inflammation is considered as the hypostasis of asthma [1, 2].

Rhein is one of the effective monomer components that are isolated and then purified from rhubarb which belongs to Chinese herbs. Rhein, which is a monanthraquinone 1, 8-dihydroxyl anthraquinone derivative, permits anti-inflammatory, anti-bacterial, anti-tumour and other effect [3, 4, 5, 6]. As far as it goes, we have probably not found any report that Rhein can be used to treating asthma by its anti-inflammatory.

Network pharmacology, which is an emerging research method for the past few years, is considered to be a new model for the next generation of drug research. While the construction of biomolecular networks such as “drug-target-path” are the basis of network pharmacology [7, 8].

We can explore the efficacy of drugs from the component-target-pathway, and can also reverse the pathogenesis of the disease through drugs with known therapeutic effect [9, 10]. Hence, by exploring the anti-inflammatory mechanism of rhubarb acid in treating asthma with network pharmacology method, a newer and more advanced view may be provided to treat asthma.

Materials And Methods

Tools used and data sources

Search for chemical composition of the drug and its targets on the web of TCMSP, PubChem, TTD, STITCH, Drugbank Database; It is possible to get the genes of related diseases and the Information on gene interactions and protein-protein interactions from Genebank Database, String Database; Signal pathways including biomolecules could be found by searching Biocarta and KEGG; Cytoscape 3.7.1 and Systemsdock are respectively for network construction and molecular docking.

Prediction of the target of Rhein prediction

Three-dimensional chemical structure data of Rhein, where it can be searched and exported from TCMSP and PubChem, is imported into the database of Swiss prediction target and do the reverse molecular docking. Predictive target can be gotten by setting the target set “homo sapiens”. The result can be used for further study.

Molecular docking to Rhein-Target protein

The PDB-ID in PDB Database is imported into the Systemsdock where the molecular docking can be processed. The value of the Docking Score is used to judge the matching degree between rhubarb acid and target. The score ranges from 0-10, the greater the value, the more stable the ligand binds to the receptor [11].

Construction of Rhein-Target Network

Construct a ‘drug-target’ interaction network with Rhein and its potential targets by using the software Cytoscape.

Construction of Rhein-Target Protein Interaction Network

By using STRING, construct Rhein-Target Protein Interaction Network by setting Protein type into Homo sapiens, the minimum interaction threshold is set to medium “medium confidence” and the remaining parameters are kept silent.

Anti-inflammatory related target protein screening

In the database TTD, search for information of anti-inflammatory target protein by using the keyword “anti-inflammation” which is put into Cytoscape3.7.1 to construct anti-inflammatory target protein PPI network.

Screening of anti-inflammatory targets for Rhein-effect and network construction of anti-inflammatory targets

Combine the Rhein-Target Protein Network with the Anti-inflammatory Protein PPI Network and import the result, which is called anti-inflammatory targets for Rhein-effect, into the database of String and construct the network. Screen values which are beyond 0.7 as high confidence basis for protein interactions.

Search for asthma related genes of human being

In NCBI Gene Database (<http://www.ncbi.nlm.nih.gov>), ‘asthma’ and ‘homo sapiens’ are searchable keywords, the asthma-related genes of a human being can be acquired.

Network of the anti-inflammatory target of Rhein against asthma

Import the anti-inflammatory target genes of Rhein and human asthma-related genes into the String database to construct the response network of drug anti-inflammatory targets against asthma in vivo and screen the anti-inflammatory targets related to asthma incidence.

Enrichment pathways of the target genes to KEGG

Use the Enrichr database to analyse the biological enrichment pathways of the target genes to KEGG for the prediction of rhein anti-inflammatory targets.

Results

The reverse docking score of Rhein and related targets, which are all beyond 5, verifying the reliability of the predicted target (Tab. 1).

Rhein-Anti-inflammatory target PPI network analysis

Nine anti-inflammatory proteins are detected in TTD, and in PPI network (Fig.3), there were nine interacting targets, constituting seven interacting relationships.

The Cytoscape 3.7.1 merge function is used to fuse the Rhein-Predicted target network to the anti-inflammatory target PPI network in the TTD, taking the intersection (Fig. 4), taking a high confidence interval beyond 0.7 to obtain Rhein resistance inflammatory targets, including mitogen-activated protein kinase (MAPK14), receptor tyrosine-protein kinase erbB-2 (ERBB2), tumor necrosis factor receptor superfamily member 1A (TNFRSF1A), epidermal growth factor receptor (EGFR).

Reaction network of Rhein anti-inflammatory targets against asthma (Fig. 5)

The Cytoscape 3.7.1 merge function was used to fuse the Rhein-predicted target network to the anti-inflammatory target PPI network in the TTD, taking the intersection (Fig. 5), taking a high confidence interval greater than 0.7 to obtain rhein resistance inflammatory targets, including mitogen-activated protein kinase (MAPK14), receptor tyrosine-protein kinase erbB-2 (ERBB2), tumor necrosis factor receptor superfamily member 1A (TNFRSF1A), epidermal growth factor receptor (EGFR).

Enrichment pathways of the target genes to KEGG

According to enrichment pathways of the target genes to KEGG, anti-inflammatory target protein of Rhein associated with asthma involves 87 signalling pathways. Enrichr analysis results are sorted in the descending order of Combined Score. The top ten pathways contain MAPK signalling pathway Hepatitis C \square Epithelial cell signalling in Helicobacter pylori infection, immune signalling pathway, and cancer signalling pathway proteoglycans in cancer, Bladder cancer, Non-small cell lung cancer, Endometrial cancer, Pancreatic cancer, Central carbon metabolism in cancer, Amyotrophic lateral sclerosis (ALS) (Fig. 6) .

Discussion

It has been reported that Rhein has anti-inflammatory activity [12, 13]. The results of this study show the anti-inflammatory effects of Rhein. Youdong Xu et al. explained the anti-inflammatory effects of Rhein from the molecular mechanism [14]. MAPK14 \square EGFR \square EERB2 \square TNFRSF1A et al. are the main target of the anti-inflammatory of Rhein. Rhein can directly act on EGFR \square MAPK14 \square EERB2 \square TNFRSF1A to exert anti-inflammatory effects, and can also act indirectly on other targets to exert its anti-inflammatory effects[15].

EGFR is an epidermal growth factor receptor, which is widely distributed in epithelial tissues and plays an important regulatory role in the development of respiratory inflammation [16]. GFR inhibitor can effectively inhibit acute inflammation of the rat respiratory tract caused by exogenous zinc ion [17]. EGFR inhibitors reduce the symptoms of allergic asthma caused by dust mites by reducing the production of pro-inflammatory factors such as IL-6 and IL-8 [18]. Thus, it is speculated that Rhein interacts with EGFR, thereby blocking the binding of EGFR to pro-inflammatory cytokines to exert an anti-inflammatory effect.

MAPK14 plays an important role in the cellular cascade triggered by pro-inflammatory cytokines or extracellular stimuli and is a key signalling molecule for lung tissue inflammation induced by S.

pneumonia [19, 20]. Inflammatory factors such as IL-1 β , IL-6 and TNF- α can positively feedback the signalling pathways such as ERK and nuclear factor- κ B (NF- κ B) by activating the p38MAPK signalling pathway and cascading amplifying the inflammatory response [21, 22]. The transcriptional cascade regulated by P38MAPKs leads to the production of pro-inflammatory factors such as TNF- α and IL-1 β , which in turn leads to the activation of enzymes involved in inflammation [23]. It is speculated that Rhein exerts its anti-inflammatory effect by inhibiting the release of pro-inflammatory factors such as TNF- α and IL-1 β by inhibiting MAPK14.

TNFRSF1A is a type 1 TNF receptor that mediates inflammatory responses mainly by activating NF- κ B, p38, ERK1/2 to induce IL-6, IL-8 synthesis and apoptosis [24, 25]. It is thus concluded that Rhein interacts with TNFRSF1A to reduce the release of anti-inflammatory factors such as IL-6 and exert an anti-inflammatory effect.

EERB2, also called HER2, NEU, CD340, is a 185kDa cell membrane receptor encoded by the proto-oncogene erbB-2 and is a member of the EGFR family. EERB2 can induce IL-6 autocrine, which in turn affects JAK-STAT or NF- κ B pathway-mediated inflammatory response [26]. It is speculated that Rhein interacts with EERB2 and may exert anti-inflammatory effects by regulating the secretion of IL-6 [27, 28, 29].

In this experiment, the anti-inflammatory mechanism of Rhein was confirmed by the method of network pharmacology. The main target of the anti-inflammatory effect of Rhein and related signalling pathways were predicted. It was found that Rhein exerted an anti-inflammatory effect by acting on multiple targets. However, network pharmacology research is based on network modelling, database resource development and software application. The network model has certain differences with the internal environment. Therefore, the research results of the anti-inflammatory effects of Rhein need to be confirmed by further experiments.

Declarations

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Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request [Supplementary Table SII](#).

Authors' contributions

All authors read and approved the final manuscript.

Ethics approval and consent to participate

The experimental protocol was established, according to the ethical guidelines of the Helsinki Declaration and was approved by the Human Ethics Committee. Written informed consent was obtained from individual or guardian participants.

Patient consent for publication

Not applicable

Publication of clinical datasets

Not applicable

Competing interests

The authors declare that they have no competing interests.

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Table

Table1. The reverse docking score of Rhein and related targets

Targets	Rhein
ERK1	8.476
MEK1	5.643
MP2K1	6.972
EGFR	6.032
PAK1	8.012
CREB1	8.785
Grb2	7.085
HRas	7.506
Myc	6.446
PDK1	7.408
RasH	8.015
SRC	8.254

Figures

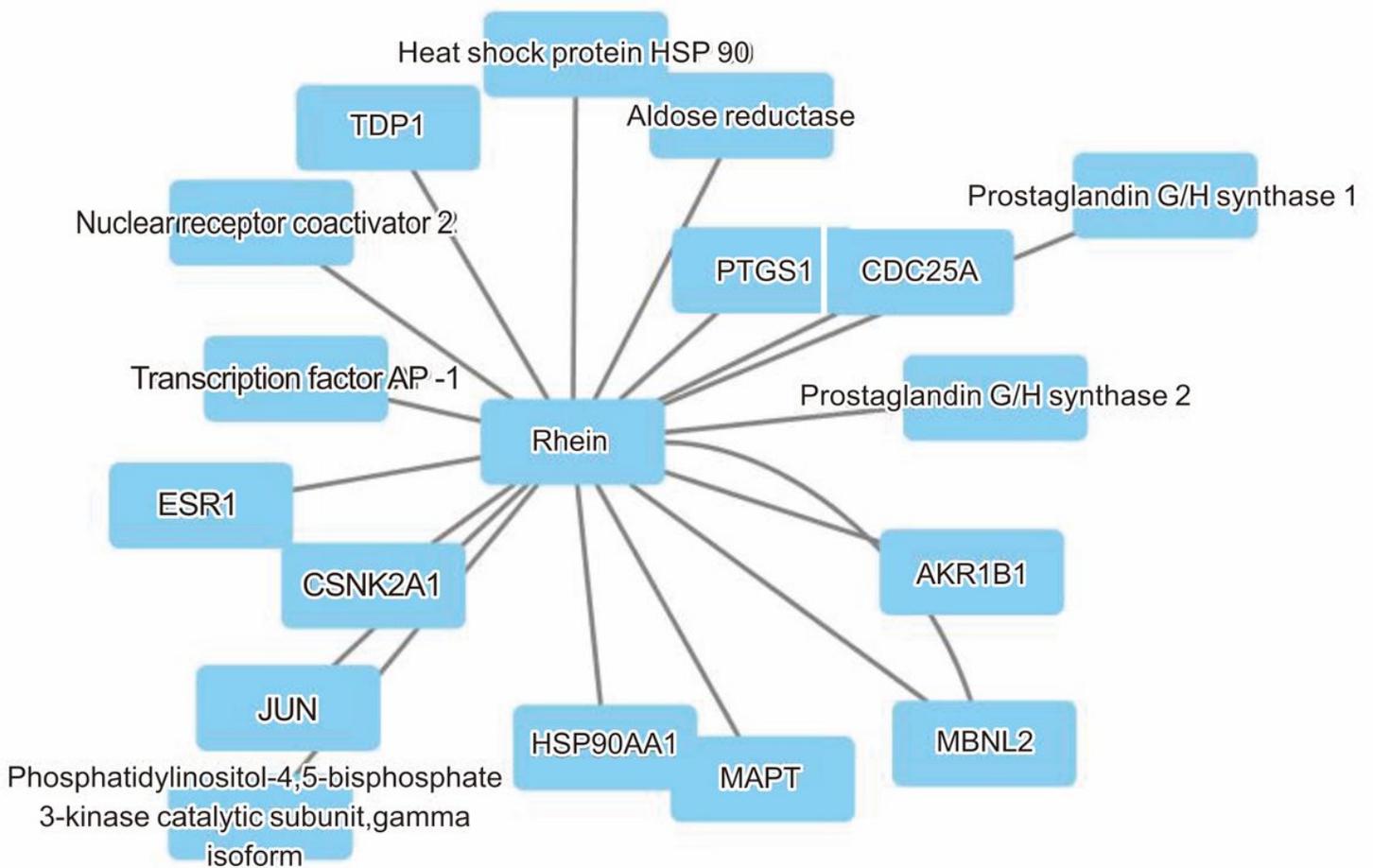


Figure 1

Seventeen predictive targets of Rhein.

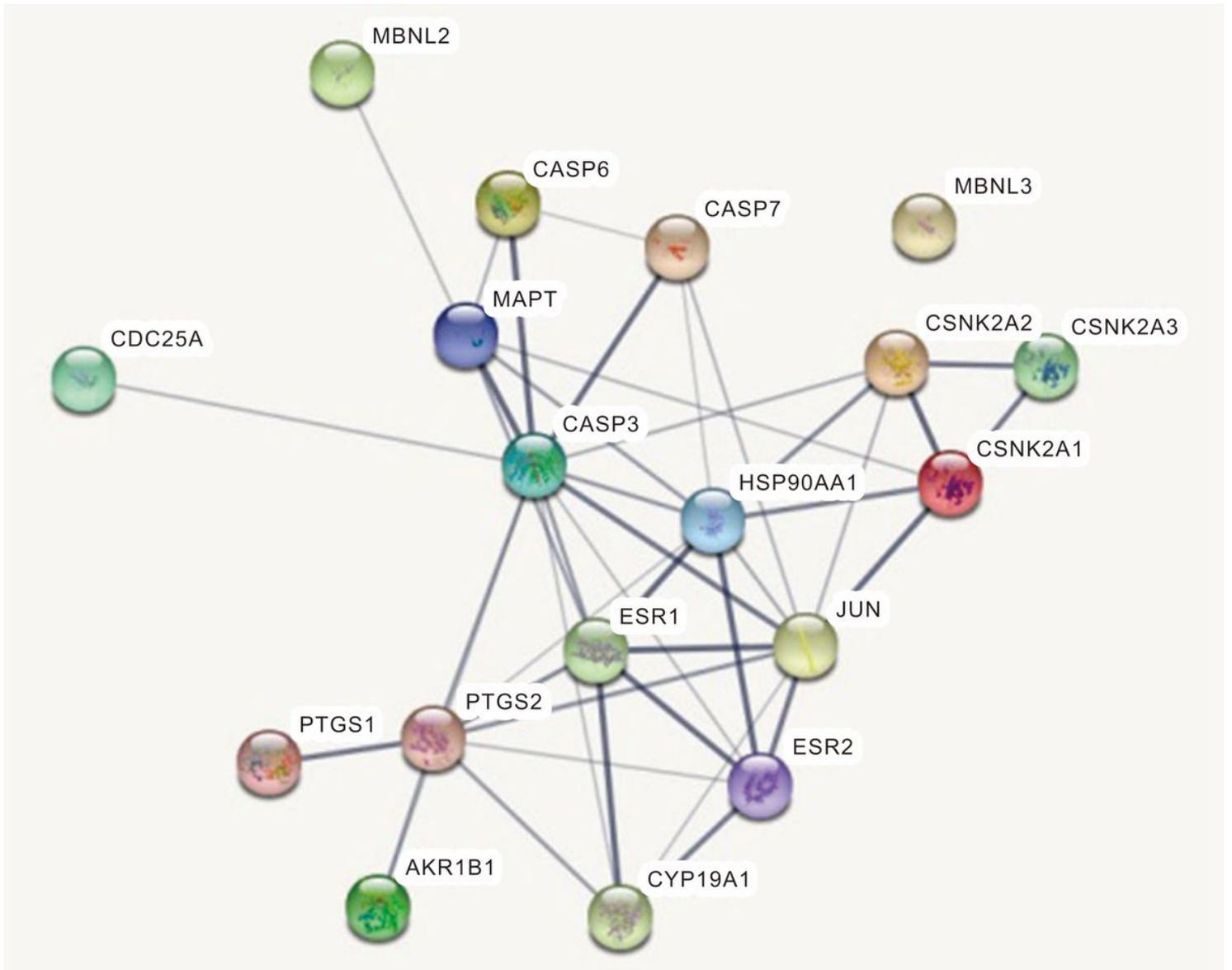


Figure 2

Network analysis map of interactions of Rhein target protein.

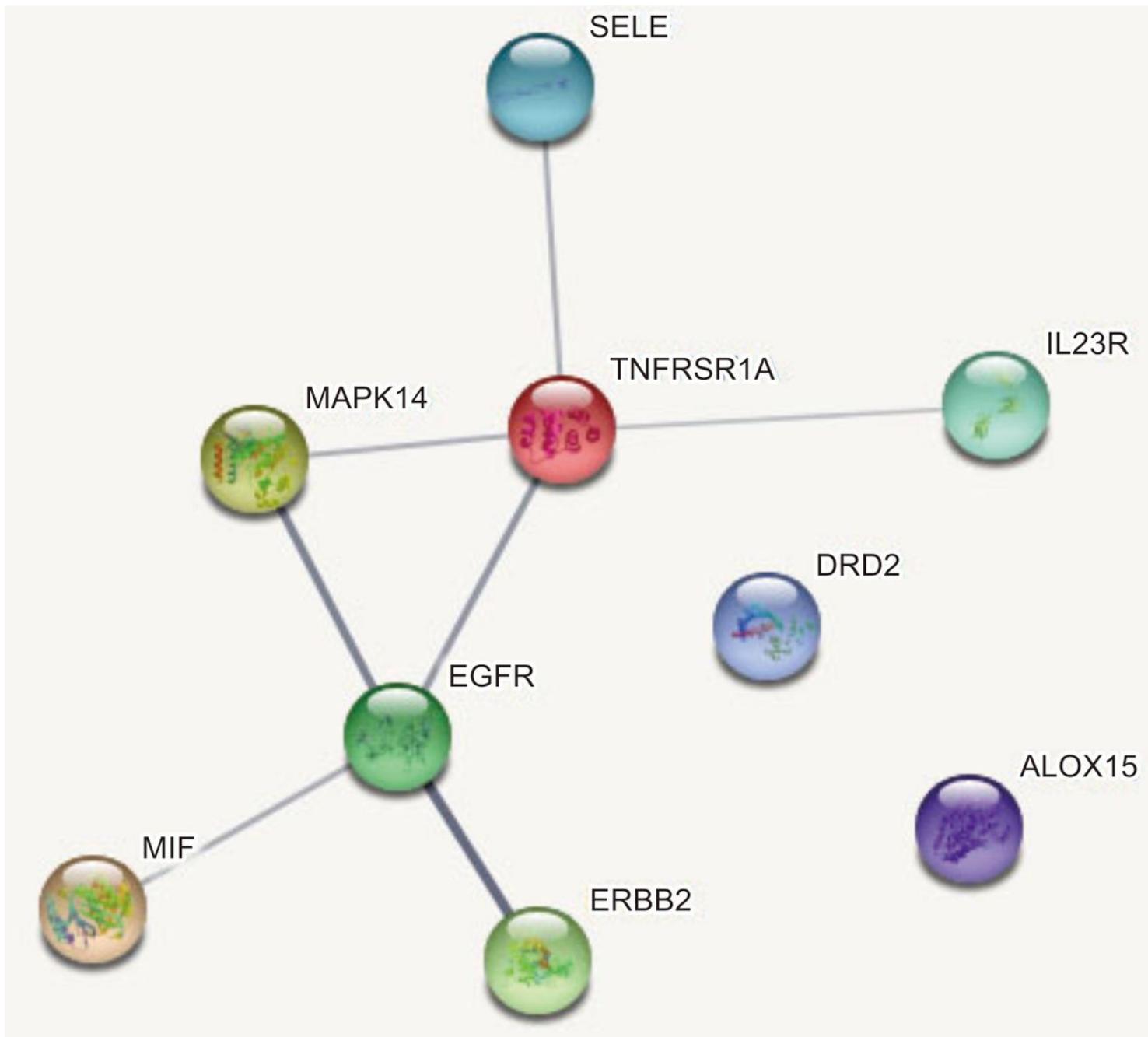


Figure 3

Seven proteins (SELE, MAPK14, TNFRSF1A, IL23R, EGFR, MIF, ERBB2) of nine proteins (SELE, MAPK14, TNFRSF1A, IL23R, EGFR, MIF, ERBB2, DRD2, ALOX15) interact with Rhein target protein.

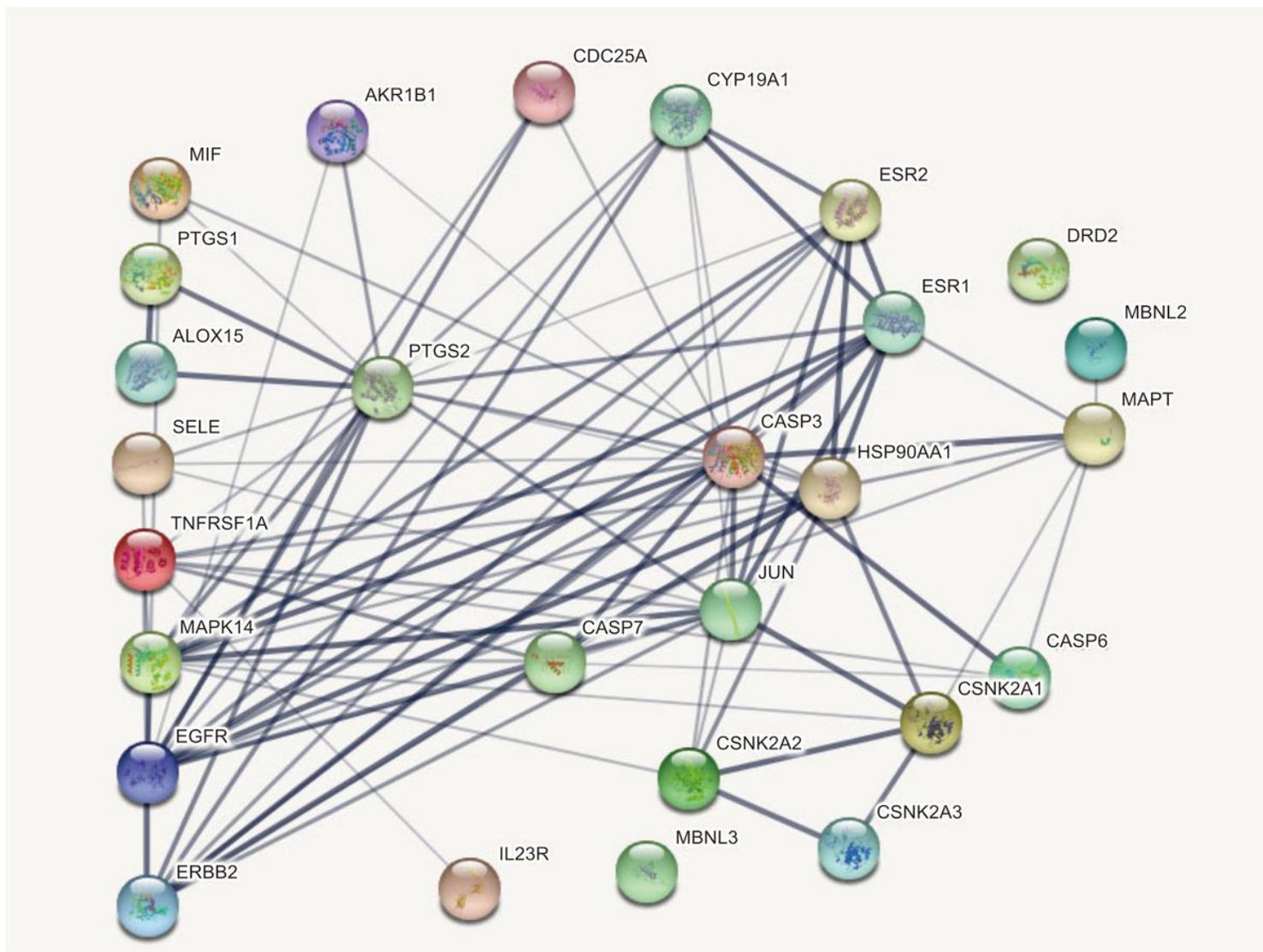


Figure 4

Anti-inflammatory targets of Rhein: MAPK14; ERBB2; TNFRSF1A; EGFR.

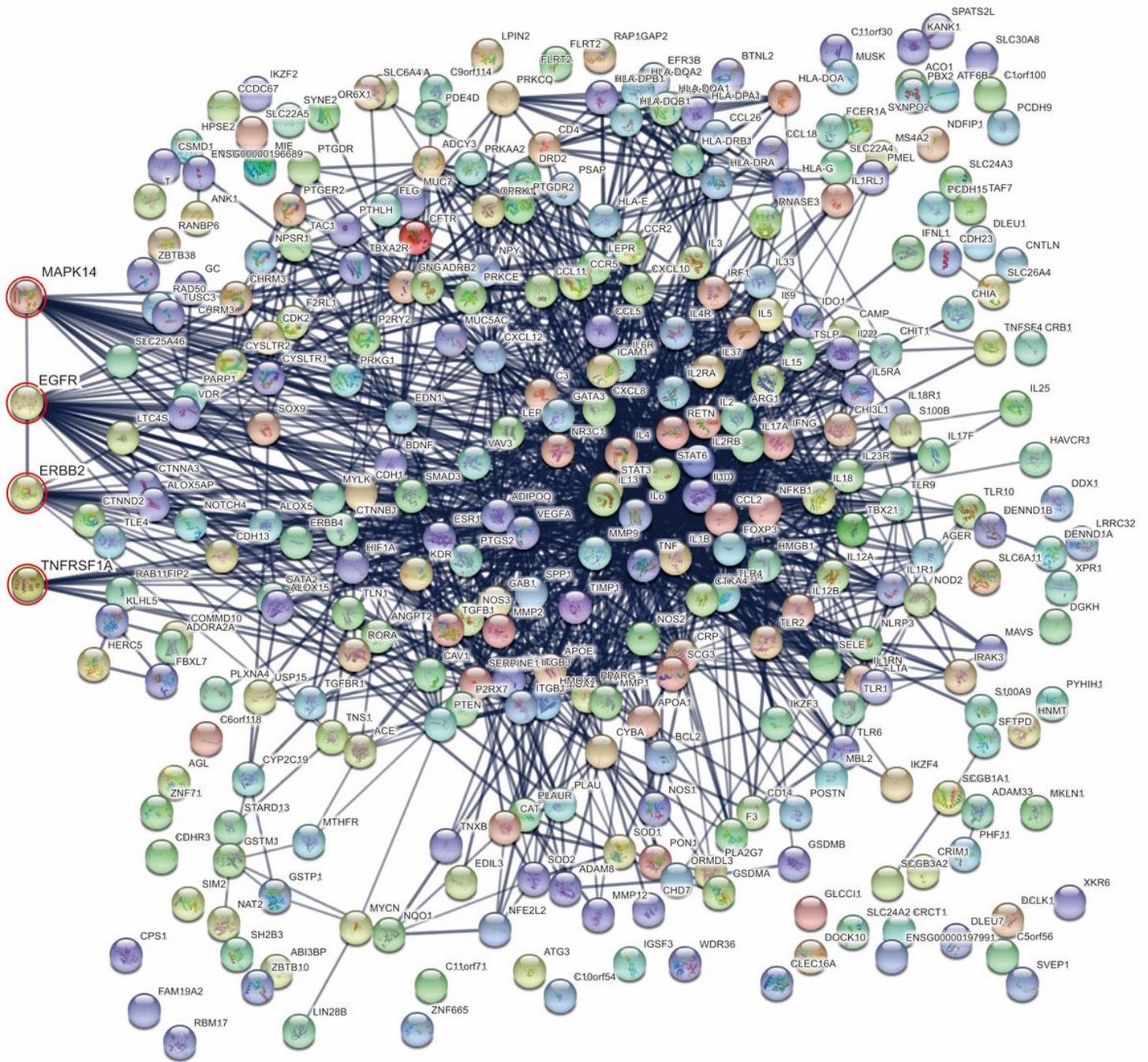


Figure 5

Four anti-inflammatory targets of Rhein related to asthma: MAPK14; EGFR; EERB2; TNFRSF1A.



Enriched Terms

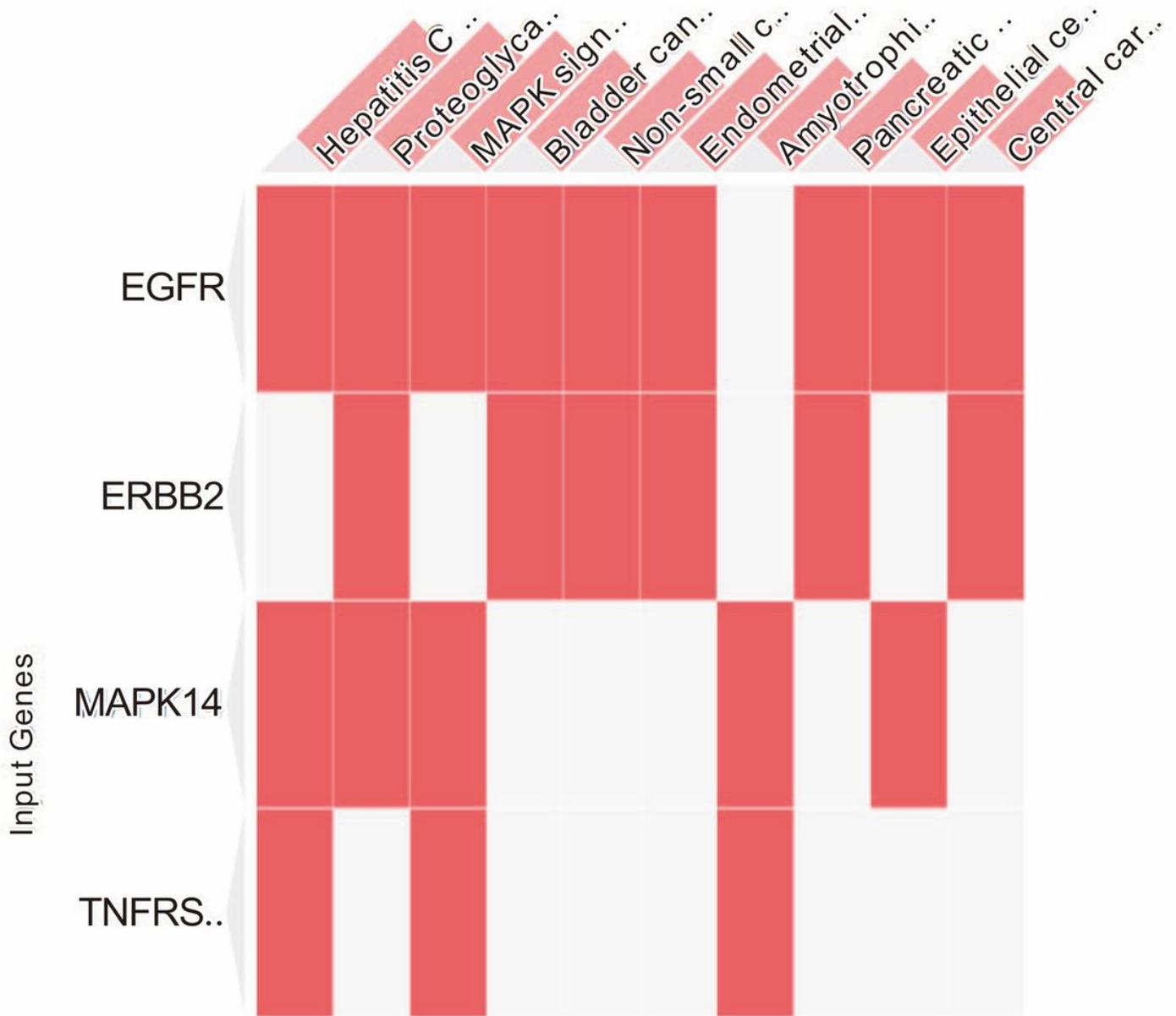


Figure 6

Top ten signaling pathways among Anti-inflammatory target protein of Rhein related to asthma: MAPK signaling pathway; Hepatitis C signaling pathway; Epithelial cell signaling in Helicobacter pylori infection signaling pathway; Proteoglycans in cancer signaling pathway; Bladder cancer signaling pathway; Non-small cell lung cancer signaling pathway; Endometrial cancer signaling pathway; Pancreatic cancer signaling pathway; Central carbon metabolism in cancer signaling pathway; Amyotrophic lateral sclerosis (ALS) signaling pathway.

Supplementary Files

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