Investigating Action Mechanism of a Compound Preparation Containing Calamine, Zinc Oxide, and Plant Extracts in Eczema Intervention Based on Network Pharmacology

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Abstract [**Objectives**] Using Calamine Zinc Oxide Phytocomplex Cream as an example, this study employed network pharmacology to investigate the therapeutic potential and mechanism of action of the combination of calamine, zinc oxide, and plant extracts in eczema intervention. [**Methods**] Active constituents of Calamine Zinc Oxide Phytocomplex Cream were identified through screening using the HIT2.0, HERB, and TCMSP databases. Corresponding targets of the active constituents were predicted using NetInfer. The collected targets were intersected with eczema and atopic dermatitis (AD)-related targets obtained from the GeneCards database to identify the effective therapeutic targets of Calamine Zinc Oxide Phytocomplex Cream. The network diagram of effective active constituents versus therapeutic targets for Calamine Zinc Oxide Phytocomplex Cream was constructed and subjected to topological analysis using Cytoscape software. The Protein-Protein Interaction (PPI) network was established and analyzed using the String database, Cytoscape software, and the cytoHubba plugin to identify key hub genes. Gene Ontology (GO) enrichment analysis and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analysis were performed on the therapeutic targets using the DAVID database. [**Results**] Screening identified 57 active constituents in Calamine Zinc Oxide Phytocomplex Cream, corresponding to 601 potential targets. Subsequent analysis revealed 72 core therapeutic targets of Calamine Zinc Oxide Phytocomplex Cream specifically relevant to eczema and AD. Analysis of the network diagram suggested that Calamine Zinc Oxide Phytocomplex Cream may exert anti-inflammatory and immunomodulatory effects through active constituents such as quercetin, luteolin, and apigenin, while concurrently repairing skin barrier function by acting on targets including AKT1, NF-κB, and STAT3. Furthermore, the inclusion of mineral-based medicines provides additional functions such as itch relief and reinforcement of the skin barrier. [**Conclusions**] Calamine Zinc Oxide Phytocomplex Cream combines organic and inorganic constituents, synergistically alleviating the adverse symptoms of eczema and AD through multiple pathways.

Key words Calamine lotion, Plant extracts, Eczema, Atopic dermatitis, Network pharmacology

1 Introduction

Eczema is an inflammatory skin disease that can be classified into types such as eyelid eczema, perioral eczema (drool rash), hand-foot eczema, and leg eczema. It often manifests with undesirable symptoms including skin redness, swelling, scaling, and exudation[1-2]. The most common form of eczema is atopic dermatitis (AD), sometimes referred to as atopic eczema[1]. AD is characterized by pruritus (itching), which often intensifies at night. This excessive itching can further lead to sleep disturbances, anxiety, hyperactivity, and depression[3]. The age of onset for AD primarily ranges from three months to 60 years, with a higher prevalence among children; however, onset after 60 years of age is still possible[4]. Its pathogenesis involves dysregulation of both the innate and adaptive immune systems. Genetically, filaggrin mutations lead to increased skin barrier permeability, facilitating the penetration of allergens into deeper skin layers. This heightens the chance of allergen encounter with antigen-presenting cells, thereby initiating a complex immune response[5]. Due to genetic factors and certain unavoidable environmental triggers, the susceptibility to and recurrent nature of eczema necessitate long-term therapy focused on anti-inflammation, itch relief, and barrier repair.

For centuries, zinc, whether in elemental or salt form, has played a pivotal role in biology and medicine. Topical formulations containing zinc oxide, calamine, and others have been used as active ingredients in photoprotective agents, soothing preparations, or anti-dandruff shampoos[6]. The clinically common Calamine Lotion (CL), primarily composed of calamine, zinc oxide, and glycerol, possesses astringent, antipruritic, antiseptic, hemostatic, antibacterial, moisturizing, and protective properties. Consequently, it is used to manage skin inflammation, rashes, and pruritus[7]. On the other hand, numerous traditional Chinese medicines (TCM) have demonstrated good efficacy in eczema treatment, with the unique advantage of relatively low adverse effects[8]. The Renhe Calamine Zinc Oxide Phytocomplex Protective Cream (hereinafter referred to as Calamine Zinc Oxide Phytocomplex Cream), which combines plant extracts (such as sunflower seed, *Centella Asiatica*, and purslane) with calamine and zinc oxide, can simultaneously ameliorate eczema inflammation and skin dryness symptoms. However, the specific synergistic mechanisms remain unclear. This study, based on network pharmacology, takes Calamine Zinc Oxide Phytocomplex Cream as the analytical subject to investigate the molecular mechanisms underlying the combined use of calamine, zinc oxide, and plant extracts. The aim is to further optimize product concentration ratios and promote the development and utilization of natural medicinal resources.

Received: September 13, 2025 Accepted: November 19, 2025

Supported by Putuo District Science and Technology R&D Platform Project, Shanghai (2024QX04).

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2 Materials and methods

2.1 Screening of active constituents and corresponding targets Active constituents of Calamine Zinc Oxide Phytocomplex Cream were collected using the Herbal Ingredients' Targets Platform (HIT2.0, http://www.badd-cao.net:2345/) and the HERB database (http://herb.ac.cn/). To enhance data authenticity and reliability, the collected active constituents were input into the Traditional Chinese Medicine Systems Pharmacology Database and Analysis Platform (TCMSP). Screening criteria were set as Topological Polar Surface Area (*TPSA*) ≤ 140 and Drug-likeness (*DL*) ≥ 0.18. The active constituents obtained from screening, along with zinc oxide and the main component of calamine (zinc carbonate), were input into NetInfer (https://lmmd.ecust.edu.cn/netinfer/) to predict their corresponding targets. For each compound, the top 50 predicted targets by score were selected to enhance relevance.

2.2 Screening of eczema-related targets Using the keywords "Eczema" and "Atopic dermatitis", relevant targets were retrieved from the GeneCards database (https://www.genecards.org/). Targets with a Relevance score greater than the average value, indicating high relevance to disease pathogenesis and development, were selected. Target information was imported into the Hiplot biomedical data online visualization tool (https://hiplot.cn) to generate a Venn diagram for identifying targets with potential regulatory effects on eczema.

2.3 Construction of the effective active constituents-therapeutic targets network Cytoscape 3.10.1 software was used for the visual analysis of the effective active constituents and therapeutic targets obtained in Section **2.2**, constructing the effective active constituents-therapeutic targets network. Nodes represented the active compounds of Calamine Zinc Oxide Phytocomplex Cream and their corresponding targets, while edges represented the interaction relationships between them. Topological analysis was performed on the established network graph. The importance of nodes within the network was evaluated using Degree and Betweenness Centrality metrics.

2.4 Construction and analysis of the Protein-Protein Interaction (PPI) network The genes corresponding to the therapeutic targets of Calamine Zinc Oxide Phytocomplex Cream relevant to eczema, obtained in Section **2.2**, were input into the String database (https://string-db.org) to establish the PPI network. The established PPI network information was imported into Cytoscape 3.10.1 software. The cytoHubba plugin's Maximal Clique Centrality (MCC) algorithm was utilized to analyze and identify hub gene targets within the PPI network.

2.5 Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analysis The coding gene information for the therapeutic targets of Calamine Zinc Oxide Phytocomplex Cream relevant to eczema, acquired in Section **2.2**, was input into the DAVID database (https://david.ncifcrf.gov). GO functional analysis and KEGG pathway enrichment analysis were performed using "Homo sapiens" as the analysis species, under the condition of $P < 0.01$. The output analysis data were visualized using Hiplot to obtain information on biological functions and pathways highly correlated with pharmacological activity.

3 Results and analysis

3.1 Analysis of active constituents and therapeutic targets Based on the screening criteria, 55 active compounds derived from medicinal plants were identified in the TCMSP database, as listed in Table 1. The targets corresponding to the 55 active constituents, zinc oxide, and the main component of calamine (zinc carbonate) were input into the UniProt database for name standardization and deduplication, resulting in 601 corresponding therapeutic targets.

Table 1 Active constituents (*TPSA* ≤ 140, *DL* ≥ 0.18)

No.	CAS	Molecule name	TPSA	DL	Medicinal plants
1	83-48-7	Stigmasterol	20.23	0.76	*Ophiopogon japonicus*
2	508-02-1	Oleanolic acid	57.53	0.76	*O. japonicus*; *Olea europaea*
3	485-72-3	Formononetin	59.67	0.21	*Sophora flavescens*
4	520-36-5	Apigenin	90.90	0.21	*S. flavescens*; *Portulaca oleracea*; *Centella asiatica*; *Chrysanthellum indicum*
5	117-39-5	Quercetin	131.36	0.28	*S. flavescens*; *P. oleracea*; *C. asiatica*; *C. indicum*
6	491-80-5	Biochanin	79.90	0.24	*S. flavescens*
7	520-33-2	Hesperetin	96.22	0.27	*S. flavescens*
8	6754-58-1	Xanthohumol	86.99	0.35	*S. flavescens*
9	53846-50-7	8-Prenylnaringenin	90.90	0.38	*S. flavescens*
10	491-70-3	Luteolin	111.13	0.25	*S. flavescens*; *P. oleracea*; *C. indicum*
11	855746-98-4	Kurarinol	116.45	0.67	*S. flavescens*
12	2035-15-6	Maackiain	57.15	0.54	*S. flavescens*
13	70872-29-6	Isoxanthohumol	75.99	0.39	*S. flavescens*
14	20575-57-9	Calycosin	79.90	0.24	*S. flavescens*
15	83-46-5	Beta-Sitosterol	20.23	0.71	*S. flavescens*; *P. oleracea*; *C. asiatica*
16	545-47-1	Lupeol	0.00	0.77	*S. flavescens*; *P. oleracea*; *C. asiatica*
17	446-72-0	Genistein	90.90	0.21	*S. flavescens*
18	519-02-8	Matrine	23.55	0.25	*S. flavescens*
19	16837-52-8	Oxymatrine	37.38	0.28	*S. flavescens*
20	6483-15-4	Sophocarpine	23.55	0.25	*S. flavescens*
21	6882-68-4	Sophoridine	23.55	0.25	*S. flavescens*
22	6882-66-2	Sophoramine	25.24	0.25	*S. flavescens*
23	13392-26-2	Catechin	110.38	0.24	*P. oleracea*; *C. asiatica*
24	208-033-4	Arachidonic Acid	37.30	0.20	*P. oleracea*
25	520-18-3	Kaempferol	111.13	0.24	*P. oleracea*; *C. asiatica*; *C. indicum*

(To be continued)

(Continued)

No.	CAS	Molecule name	TPSA	DL	Medicinal plants
26	480-19-3	Isorhamnetin	120.36	0.31	*P. oleracea*; *C. indicum*
27	970-73-0	Gallocatechin	130.61	0.27	*P. oleracea*
28	7235-40-7	Beta Carotene	0.00	0.58	*P. oleracea*
29	77-52-1	Ursolic Acid	57.53	0.75	*C. asiatica*
30	4547-24-4	Corosolic Acid	77.76	0.74	*C. asiatica*
31	464-92-6	Asiatic Acid	97.99	0.72	*C. asiatica*
32	57-88-5	Cholesterol	20.23	0.68	*C. asiatica*
33	490-46-0	(-)-Epicatechin	110.38	0.24	*C. asiatica*; *C. japonica*
34	34157-83-0	Celastrol	74.60	0.78	*C. asiatica*
35	472-15-1	Betulinic Acid	41.63	0.78	*C. asiatica*
36	111-02-4	Squalene	0.00	0.42	*C. asiatica*
37	154-23-4	Cianidanol	110.38	0.24	*C. asiatica*
38	22368-21-4	Eupatilin	98.36	0.38	*C. indicum*
39	480-44-4	Acacetin	79.90	0.24	*C. indicum*
40	520-34-3	Diosmetin	100.13	0.27	*C. indicum*
41	520-32-1	Tricin	109.36	0.34	*C. indicum*
42	24181-77-9	Fumaricine	60.39	0.72	*Helianthi annui*
43	478-01-3	Nobiletin	85.59	0.52	*H. annui*
44	52211-63-9	Quinicine	51.22	0.33	*H. annui*
45	3763-55-1	Rubixanthin	20.23	0.53	*H. annui*
46	531-44-2	Scopolin	138.82	0.39	*H. annui*
47	6451-72-5	Scoulerine	62.16	0.54	*H. annui*
48	4373-41-5	Maslinicacid	77.76	0.74	*H. ennui*
49	148-03-8	β-tocopherol	133.06	0.70	*H. ennui*
50	79-63-0	Lanosterol	20.23	0.75	*H. ennui*
51	14162-53-9	Oleanolic acid-28-O-beta-D-glucopyranoside	136.68	0.41	*H. ennui*
52	2955-23-9	Olivil	108.61	0.41	*H. ennui*
53	474-62-4	Campesterol	20.23	0.71	*Camellia japonica*
54	20853-07-0	Protoaescigenin	121.38	0.69	*Macadamia ternifolia*
55	2181-75-1	Indicaxanthin	129.77	0.22	*Opuntia ficusIndica*

3.2 Intersection of therapeutic targets and disease targets

Using the keywords "Eczema" and "Atopic Dermatitis" in the GeneCards database, targets associated with eczema were retrieved. Targets with Relevance scores exceeding the mean value were selected, yielding 606 eczema-related targets and 380 atopic dermatitis-related targets. The therapeutic targets of active constituents from Calamine Zinc Oxide Phytocomplex Cream and eczema-related targets were imported into Hiplot for intersection analysis, as depicted in Fig. 1. Analysis identified 29 eczema-specific targets, 16 atopic dermatitis-specific targets, and 27 overlapping core targets—totaling 72 core therapeutic targets for eczema and AD intervention by Calamine Zinc Oxide Phytocomplex Cream.

3.3 Analysis of the effective active constituents-therapeutic targets network Cytoscape 3.10.1 software was employed to visualize effective active constituents and their therapeutic targets, constructing the network diagram shown in Fig. 2. The network comprised 139 nodes and 403 edges. Topological analysis revealed an average node degree of 5.80, with 67 nodes exceeding this value. The average betweenness centrality was 0.016, surpassed by 38 nodes. 37 nodes satisfied both criteria simultaneously (Table 2). Table 2 identifies *S. flavescens*, *C. asiatica*, and *P. oleracea* as core medicinal plants; Quercetin, Luteolin, and Apigenin as pivotal active compounds; and ADRB2, MAOA, NR3C1 among others as key therapeutic targets.

AD severity correlates with immune-inflammatory dysregulation and elevated cytokine expression. IL-4 and IL-13 lower sensory neuron thresholds to pruritogenic stimuli, activating immune cells and itch-sensing nerve fibers to induce pruritus[9]. *S. flavescens*, a traditional Chinese herb, demonstrates efficacy against eczema and atopic dermatitis[10]. Pharmacological studies reveal that oxymatrine—a primary active constituent—inhibits mast cell degranulation and reduces release of inflammatory mediators (histamine, LTB4, IL-2, IL-4), exerting therapeutic effects in AD[11].

Table 2 Key nodes and topological features of the active compounds-targets network

No.	Name	Type	Degree	Betweenness centrality
1	ADRB2	Target	24	0.156
2	*Sophora flavescens*	Medicinal Plant	20	0.106
3	MAOA	Target	19	0.096
4	Quercetin	Compound	19	0.095
5	NR3C1	Target	18	0.083
6	TRPV1	Target	17	0.088
7	VDR	Target	16	0.068
8	Luteolin	Compound	16	0.061
9	Apigenin	Compound	16	0.054
10	*Centella asiatica*	Medicinal Plant	15	0.072
11	LCK	Target	15	0.047
12	Beta-Sitosterol	Compound	11	0.045
13	*Portulaca oleracea*	Medicinal Plant	11	0.043
14	Oleanolic acid	Compound	10	0.071
15	Protoaescigenin	Compound	10	0.047
16	Arachidonic Acid	Compound	10	0.043
17	Calycosin	Compound	9	0.036
18	Sophocarpine	Compound	9	0.035
19	ALOX5	Target	9	0.030
20	Genistein	Compound	9	0.030
21	TBXA2R	Target	9	0.027
22	Lupeol	Compound	8	0.032
23	Beta Carotene	Compound	8	0.030
24	Catechin	Compound	8	0.029
25	Stigmasterol	Compound	8	0.026
26	Rubixanthin	Compound	8	0.025
27	Campesterol	Compound	8	0.022
28	ELANE	Target	8	0.021
29	Corosolic Acid	Compound	7	0.031
30	Nobiletin	Compound	7	0.024
31	Tricin	Compound	7	0.019
32	Ursolic Acid	Compound	7	0.019
33	Cholesterol	Compound	6	0.033
34	Olivil	Target	6	0.025
35	Cianidanol	Compound	6	0.022
36	Indicaxanthin	Compound	6	0.019
37	Acacetin	Compound	6	0.019

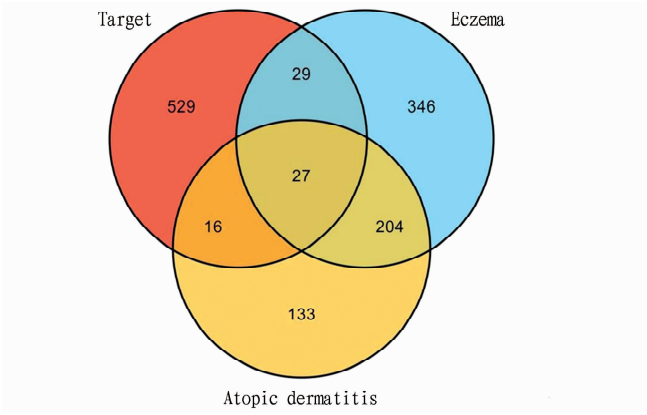
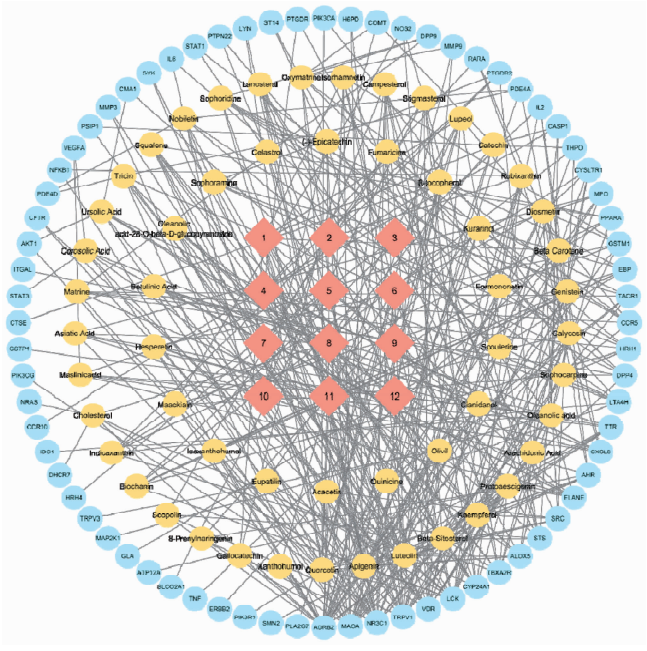


Fig. 1 Venn diagram of eczema targets, atopic dermatitis targets, and therapeutic targets of Calamine Zinc Oxide Phytocomplex Cream



NOTE 1. *Portulaca oleracea*; 2. Sunflower seed; 3. Opuntia ficus-indica; 4. Calamine; 5. Zinc oxide; 6. *Matricaria chamomilla*; 7. *Ophiopogon japonicus*; 8. *Centella asiatica*; 9. *Macadamia ternifolia*; 10. *Camellia japonica*; 11. *Sophora flavescens*; 12. *Olea europaea*.

Fig. 2 Active constituents-targets network of Calamine Zinc Oxide Phytocomplex Cream

Asiatic acid from *C. asiatica* exhibits potent antioxidant and anti-inflammatory activity by activating PPAR-γ, thereby suppressing LPS-induced NF-κB activation and production of PGE2, NO, IL-6, and IL-8[12]. Ethanol extracts of *C. asiatica* display anti-inflammatory and immunomodulatory effects in AD via both topical and oral administration[13]. Wei *et al.* [14] demonstrated that *P. oleracea* aqueous extract inhibits JAK1 enzymatic activity and JAK1-mediated STAT signaling, ameliorating skin lesions and restoring barrier function in AD murine models. Quercetin possesses notable antioxidant and anti-inflammatory properties; liposomal gels containing quercetin exert preventive and therapeutic effects against eczema when applied topically[15]. Luteolin modulates inflammatory skin diseases like AD by suppressing cytokines including IL-1β, IL-6, IL-8, IL-17, IL-22, TNF-α, and COX-2[16]. Apigenin alleviates AD symptoms by downregulating MAPK, NF-κB, and JAK/STAT signaling pathways, thereby reducing pro-inflammatory cytokines and mediators[17]. Additionally, calamine and zinc oxide, as mineral medicines, leverage physical properties to reduce exudate in acute eczema and inhibit bacterial colonization[18].

3.4 PPI network analysis and hub gene identification The PPI network for eczema/AD therapeutic targets comprised 72 protein nodes and 456 edges (Fig. 3). Edges represent interactions; nodes with more connections indicate greater biological significance. The cytoHubba plugin's MCC algorithm identified hub genes, visualized in Fig. 4.

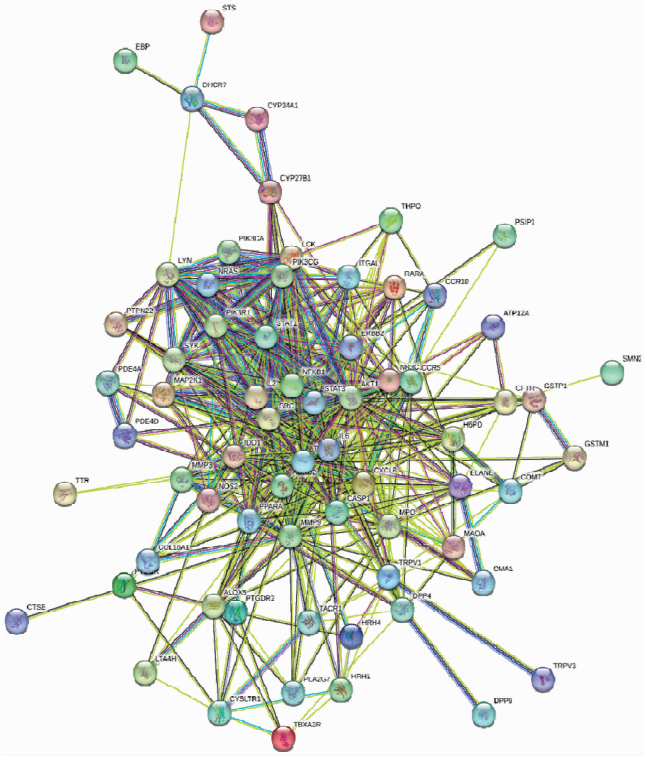


Fig. 3 PPI network of therapeutic targets

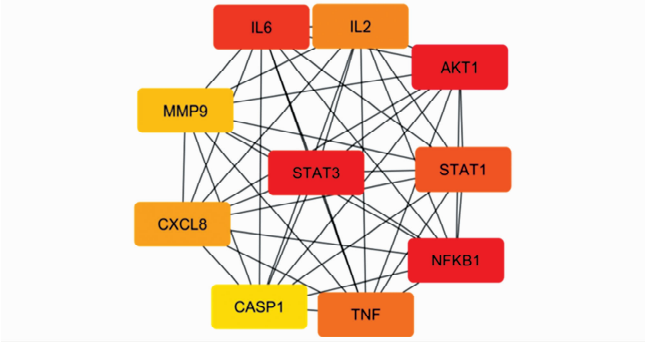
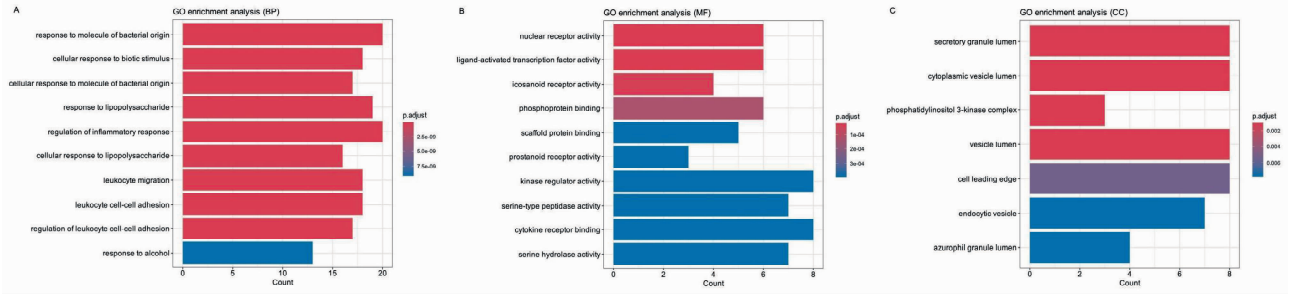


Fig. 4 Hub genes of Calamine Zinc Oxide Phytocomplex Cream

The top five hub genes were AKT1, NFKB1, STAT3, IL-6, and STAT1 (Fig. 4). AKT1, a key PI3K/AKT pathway protein,

regulates cell proliferation and inflammation, implicating it in eczema-related inflammation and barrier repair[19]. NF-κB1 (NFKB1), a member of the NF-κB family, is a master regulator of inflammation. In AD pathogenesis, this pathway amplifies inflammatory cascades by promoting pro-inflammatory factors (*e.g.*, IL-6, TNF-α) and chemokines, exacerbating barrier disruption[20]. Thus, NFKB1 inhibition may mitigate AD-associated damage. STAT3 and STAT1 mediate JAK-STAT signaling, playing central roles in Th17-mediated immune responses in AD[21]. JAK-STAT signaling also modulates epidermal barrier function and peripheral nerve activity involved in itch transduction[22]. Targeting this pathway may attenuate these signals. IL-6, a pro-inflammatory cytokine elevated in AD patient serum, correlates positively with disease severity[23]. Collectively, active constituents likely modulate eczema/AD inflammatory processes and preserve epidermal barrier integrity by targeting these hubs.

3.5 Gene Ontology (GO) functional analysis The 72 therapeutic targets of active compounds were subjected to GO functional analysis using the DAVID database under the threshold of $P < 0.01$, yielding 372 significant GO terms. These comprised 270 Biological Process (BP), 68 Molecular Function (MF), and 34 Cellular Component (CC) terms. Data were imported into Hiplot for visualization. Based on *P*-value, *Q*-value ($Q < 0.05$), and gene count enrichment, the top 10 entries for BP, MF, and CC were selected to generate bar charts (Fig. 5). Eczema-relevant BP terms included: regulation of inflammatory response, cellular response to lipopolysaccharide, leukocyte migration, response to molecule of bacterial origin, and cellular response to biotic stimulus. MF terms associated with eczema encompassed: cytokine receptor binding, prostanoid receptor activity, eicosanoid receptor activity, serine-type peptidase activity, and kinase regulator activity. CC terms potentially involved in eczema pathogenesis included: secretory granule lumen, cytoplasmic vesicle lumen, phosphatidylinositol 3-kinase complex, cell leading edge, and azurophil granule lumen.



NOTE A. BP enrichment results; B. MF enrichment results; C. CC enrichment results.

Fig. 5 GO Functional analysis of therapeutic targets

3.6 KEGG pathway enrichment analysis KEGG pathway enrichment analysis identified 133 significant pathways ($P < 0.01$). Pathway data were imported into Hiplot. Based on *P*-value and gene enrichment count, the top 20 pathways were visualized in a bubble plot. Fig. 6 indicates potential modulation of eczema through inflammation-related pathways, including: Chemokine signaling pathway, C-type lectin receptor signaling pathway, AGE-RAGE signaling pathway in diabetic complications, Fc epsilon RI signaling pathway, Toll-like receptor signaling pathway, and T cell receptor signaling pathway.

Chemokines are cytokines directing cellular migration. Recent studies reveal chemokines can excite nerve terminals, inducing pruritus while exacerbating inflammation and compromising skin barrier integrity[24]. C-type lectin receptors (CLRs) orchestrate cellular responses through complex signaling, mediating recognition of bacteria, fungi, and pathogens while regulating inflammatory immune responses[25]. Elevated Immunoglobulin E (IgE) levels are frequently associated with allergic diseases like AD[26]. Activation of the IgE-FcεRI pathway triggers mast cell degranulation, releasing histamine and other mediators that induce pruritus, vasodilation, and inflammation—a key mechanism in acute AD flares[27]. Thus, active constituents may suppress eczema and AD progression by modulating inflammatory immune responses and pruritic neural signaling pathways.

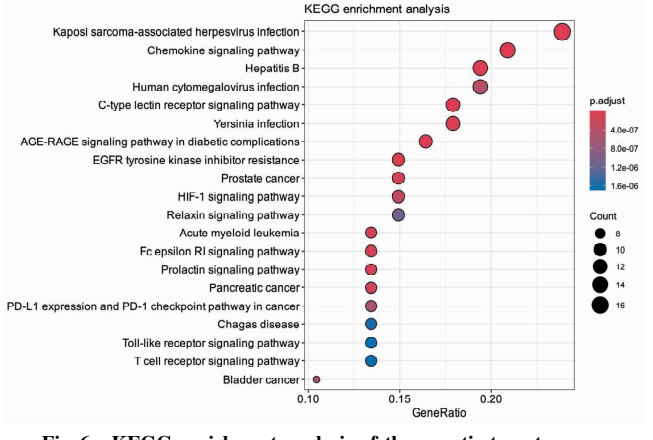


Fig. 6 KEGG enrichment analysis of therapeutic targets

4 Discussion

The pathogenesis of eczema/AD is multifactorial, involving genetic skin barrier defects, immune dysregulation, sensory neuron hypersensitivity to pruritogens, skin microbiome disturbances, and environmental triggers[28]. Barrier defects from filaggrin mutations, scratching-induced damage, microbial dysbiosis, and environmental insults can increase transepidermal water loss, cause xerosis, elevate skin surface pH, enhance epidermal permeability, and

stimulate pro-inflammatory cytokines. Network pharmacology suggests core components—particularly plant extracts—may preserve skin barrier function by modulating inflammation, balancing immune responses, and alleviating pruritus. Liquid lipid extracts (sunflower, olive, camellia, macadamia) provide emollient and moisturizing effects, help normalize sebum composition, and restore barrier function, offering adjunctive management and prevention for chronic eczema[29]. Furthermore, Meng *et al.* [7] reported calamine lotion (containing calamine/zinc oxide) combined with mometasone furoate ointment was more effective than monotherapy for infantile eczema. Calamine and zinc oxide reduce exudation, alleviate pruritus, inhibit bacterial colonization, and form a physical barrier against irritants and moisture loss. This complements plant extract activities, enabling multi-pathway eczema mitigation.

5 Conclusions

Botanical and mineral medicines are integral to Traditional Chinese Medicine. Using network pharmacology, this study analyzed the multi-target orientation and synergism of mineral-plant complexes in Calamine Zinc Oxide Phytocomplex Cream. Screening identified 55 plant-derived active constituents via HIT2.0, HERB, and TCMSP databases. Intersection analysis of 601 eczema/AD targets from GeneCards with predicted targets of zinc oxide, zinc carbonate (calamine's main component), and the 55 plant-derived compounds yielded 72 potential therapeutic targets. Network analysis identified quercetin, luteolin, and apigenin as primary active compounds. PPI, GO, and KEGG analyses indicate modulation of eczema symptoms via action on AKT1, NFKB1, STAT3, IL-6, and STAT1—affecting cytokines/chemokines, regulating inflammation, and altering immune responses to exogenous stimuli like bacteria. Notably, emollient plant oils and astringent minerals contribute to multi-pathway symptom relief, complementing traditional plant extracts. Network pharmacology predicts the therapeutic applicability of this mineral-botanical complex for eczema, warranting further investigation into its *in vivo* synergism.

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cessfully overcoming the technical bottleneck in isolating active components from *E. americana*. Utilizing an innovative solvent system composed of petroleum ether-ethyl acetate-ethanol-water (5∶5∶6∶4, *V/V/V/V*), this study successfully achieved high-efficiency separation of eleutherol, eleutherin, and isoeleutherin. Compared to traditional silica gel column chromatography, this method reduces the separation time to 3 h and simultaneously isolates three active components from the ethnomedicine *E. americana*, significantly improving efficiency. More importantly, by establishing a coupled technology integrating HSCCC and silica gel column chromatography, large-scale preparation from 2 kg of raw material yielded 20 mg of eleutherol (purity 99%), 310 mg of eleutherin (purity 98%), and 38 mg of isoeleutherin (purity 98%). This approach demonstrated enhanced production yields, reduced costs, and promising industrial application potential. This study not only provides a scientific basis for improving the quality standards of *E. americana*, but also establishes an eco-friendly separation strategy that offers new insights for the large-scale preparation of naphthoquinone natural products, which will significantly promote the modern development and utilization of ethnomedicine resources. Future research should further explore the combined application of HSCCC with other separation technologies, as well as investigate the *in vivo* metabolic mechanisms of active components from *E. americana* and develop novel drug delivery systems.

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