

Supplementary File

Network pharmacology and molecular docking reveal anti-asthmatic potential of *Zephyranthes rosea* in an ovalbumin-induced asthma model

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Abstract: The present study aimed to evaluate the anti-inflammatory effects of *Zephyranthes rosea* in an ovalbumin-induced asthma model. Allergic asthma was induced in mice via intraperitoneal injection followed by intranasal ovalbumin challenge. The methanolic extract of *Z. rosea* (200-600 mg/kg) was orally administered to asthmatic mice for 14 days. Hematological parameters for bronchoalveolar lavage fluid (BALF) and blood were analyzed. The mRNA expression levels of interleukins and transforming growth factor-beta (TGF- β 1) in lung tissues were determined using reverse transcriptase-polymerase chain reaction (RT-PCR). Analysis of network pharmacology was used to find possible *Z. rosea* targets. After building a protein-protein interaction network to find hub genes, GO and KEGG enrichment analyses were carried out to determine the potential mechanism. *In silico* analysis was performed by Molecular Operating Environment. The phytochemical and GCMS analysis of *Z. rosea* extract indicated major classes of phytochemicals. Hematological parameters in blood, and BALF from *Z. rosea* extract-treated animals were significantly reduced in a dose-dependent fashion. Histopathology revealed that *Z. rosea* had an ameliorative effect on lung tissues. Moreover, treatment with *Z. rosea* extract significantly restored the normal levels of IL-4, IL-6, IL-1 β , IL-10, IL-13, and TGF- β 1 in allergic asthmatic mice compared to diseased group. *In-silico* analysis, particularly of the binding affinities of *Z. rosea* phytoconstituents for IL6, AKT1, and Src, supported *in-vivo* results. These findings indicated that *Z. rosea* extract significantly ameliorates cellular and molecular biomarkers of bronchial inflammation and could be a potential candidate for treating allergic asthma.

Keywords: Asthma; Ovalbumin; Bronchoalveolar lavage fluid; Interleukin 6; Akt Serine/Threonine Kinase 1; Proto-oncogene tyrosine-protein kinase Src; Network pharmacology; Tumor necrosis factor-alpha; *Zephyranthes rosea*

Table S1. Effect of different doses of *Z. rosea* on hematological parameters in blood (mean \pm S.D.).

Parameter	Unit	Normal Control	Diseased Control	Positive Control	<i>Z. rosea</i> 200	<i>Z. rosea</i> 400	<i>Z. rosea</i> 600
Hb	g/dL	14.9 \pm 0.058	13.21 \pm 0.152	13.95 \pm 0.08	12.85 \pm 0.088	14.21 \pm 0.12	14.97 \pm 0.088
RBCs	$\times 10^6/\mu\text{L}$	8.95 \pm 0.117	7.21 \pm 0.09	7.91 \pm 0.216	7.29 \pm 0.35	8.26 \pm 0.05	8.81 \pm 0.03
Platelets	$\times 10^3/\mu\text{L}$	571.0 \pm 18.77	1071 \pm 26.72	821.31 \pm 17.05	989.12 \pm 12.71	923.31 \pm 4.98	891.62 \pm 12.72
Lymphocytes	%	42.24 \pm 1.45	79.12 \pm 4.16	51.0 \pm 0.578	62.32 \pm 1.76	61.21 \pm 1.2	58.51 \pm 1.45
Monocytes	%	3.27 \pm 0.88	6.0 \pm 0.58	3.91 \pm 0.167	4.12 \pm 0.58	3.82 \pm 0.33	3.59 \pm 0.58