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Original Article

Allium hookeri Inhibits Inflammation in OVA-induced Allergic Rhinitis in Mice via NF-κB pathway

Yan Jing Fan[1,†,‡], Juan Jin[1,†], Chun Hua Piao[1,&], Thi Van Nguyen[1], You-Suk Kim[3], Ok Hee Chai[1,2], Chang Ho Song[1,2]

[1]*Department of Anatomy, Jeonbuk National University Medical School*
[2]*Institute for Medical Sciences, Jeonbuk National University*
[3]*Goldtree Co., Ltd.*
[‡]*Current address: School of Medicine, Liaocheng University, Liaocheng 252000, China*
[&]*Current address: Department of Pulmonary and Critical Care Medicine, Yantai Yuhuangding Hospital, Yantai 264000, China.*

Abstract : *Allium hookeri* (*A. hookeri*) has been used as a traditional medicine for many years in the Southeast Asian region. Multiple investigations have demonstrated its role in preventing of microbial infection, immune regulation, coronary thrombosis, and atherosclerosis has been demonstrated by several studies. Therefore, this study aims to investigate the suppressing effect of *A. hookeri* on the ovalbumin (OVA)-induced allergic rhinitis (AR) murine model. In this study, AR was induced in BALB/c mice by using OVA emulsified in aluminum on days 1, 8 and 15. Then, nasally challenged with OVA on days 22 to 28. On days 16 to 28, OVA-induced mice were treated either with *A. hookeri* (50, 100, 200 mg/kg) or Dexamethasone (Dex, 2.5 mg/kg) by oral gavage. On days 22 to 28, mice received either *A. hookeri* (50, 100, 200 mg/kg) or Dexamethasone (Dex, 2.5 mg/kg) an hour before the OVA challenge. The mice in the control group were not sensitized, treated, or tested. In this study, *A. hookeri* ameliorated nasal symptoms include rubbing and sneezing; reduced thickness of nasal mucosa; alleviated goblet cell hyperplasia and eosinophil infiltration in the nasal mucosa; decreased levels of OVA-specific IgE and OVA-specific IgG1 in serum; and suppressed Th2 (IL-4, IL-5, and IL-13) cytokines level in nasal lavage fluid (NALF). In addition, *A. hookeri* recovered the production of Th1 (IL-12 and IFN-γ) cytokines level in NALF, inhibited the inflammation through NF-κB pathway. Collectively, our results suggest that *A. hookeri* effectively protects against AR by regulating Th1/Th2 cytokines imbalance and NF-κB mechanisms. These observations suggest *A. hookeri* as a potential therapeutic option for AR.

Keywords : *Allium hookeri*, Allergic rhinitis, Th1 cytokines, Th2 cytokines, NF-κB

[†]These authors contributed equally to this work
The author(s) agree to abide by the good publication practice guideline for medical journals.
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Co-correspondence to: Ok Hee Chai (Department of Anatomy and Institute for Medical Sciences, Jeonbuk National University Medical School, Jeonju, Jeonbuk 54896, Republic of Korea), Chang Ho Song (Department of Anatomy and Institute for Medical Sciences, Jeonbuk National University Medical School, Jeonju, Jeonbuk 54896, Republic of Korea)
E-mail: okchai1004@jbnu.ac.kr (O.H. Chai), asch@jbnu.ac.kr (C.H. Song)

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INTRODUCTION

Allergic rhinitis (AR) is an immunoglobulin E (IgE) mediated allergic disease that can occur after exposure to causative allergens with characteristic symptoms, including sneezing, nasal itching, nasal congestion, and rhinorrhea [1]. AR and other allergic diseases are characterized by T helper 2 (Th2) mediated allergic inflammation [2], and an imbalance between Th1/Th2 cells plays a pivotal role in AR pathogenesis [3]. In the pathological process of AR, Th2 cells tend to increase and induce cytokines such as interleukin 4 (IL-4), IL-5, and IL-13, which are implicated in AR response [2]. These changes could modulate the response of innate effector cells, which include eosinophil, basophils, and mast cells. Further, T helper cells are recruited by the effectors cells, and promote the secretion of IgE [4]. The high affinity IgE receptor (FcεRI) on the surface of these effector cells could bind with allergen-specific IgE, then release allergic mediators, including histamine and cytokines, and give rise to the development of type 1 hypersensitivity [5,6]. Hence, the regulation of Th2 cells could be the target of AR therapy [7]. The nuclear factor-kappa B (NF-κB) is a multicellular transcription factor that plays an important role in immune response and inflammation. NF-κB regulates many cellular pathways and processes, including the immune response, inflammation, proliferation, and apoptosis [8]. After being activated, the inactive form of NF-κB transfers into the nucleus and regulates the inflammatory cytokine secretion and the expression of inflammatory genes [9,10]. Moreover, the suppression of NF-κB contributes to the inhibition of lung neutrophilia and cytokine production in LPS-induced acute lung injury [11] and asthma [12,13]. Inhibitory kappa B alpha (IκBa) and p65 are key regulators of activation and inhibition in the NF-κB pathway, respectively [10]. Therefore, NF-κB has emerged as a vital therapeutic molecular target, and inhibition of NF-κB activity would be a welcome method for managing AR [8]. The common treatment options for AR involve pharmacotherapy, allergen avoidance, surgery, and immunotherapy [14]. The pharmacotherapy treatment consists of antihistamines, nasal glucocorticoids, decongestants, leukotriene receptor antagonists, and more [15]. However, long-term use of these drugs can lead to side effects such as headaches, nasal dryness, and throat irritation [16-18]. Thus, it is desirable to develop safer and more effective therapeutic agents [19], especially natural products, for the treatment of AR.

Allium hookeri (AH, *A. hookeri*) belongs to the *Amaryllidaceae* family. It is a traditional herb and vegetable in Asia and has been used as a food additive in China [20]. *A. hookeri* has been used to treat the common cold and cough, heal burns and wounds as well as to recover immunity [21]. Recently, several biological effects of *A. hookeri* have been reported, including antimicrobial [22], anti-obesity [23], antioxidant [24], and anti-inflammatory [25] effects. Kim. et al. have investigated whether *A. hookeri* might protect against AR via regulation of the vascular endothelial growth factor [26]. Interestingly, an investigation by Lee. et al. demonstrated that *C. longa* and *A. hookeri* co-treatment synergistically inhibit inflammation by regulating the NF-κB/COX-2/iNOS pathway in an air pouch mouse model [27]. Despite this progress, further studies are needed to understand the mechanism of *A. hookeri* for AR. In this study, we investigated the anti-AR effect of *A. hookeri* through immunological modulation of Th1/Th2-related factors and NF-κB pathway in an ovalbumin (OVA) induced AR mouse model.

MATERIALS AND METHODS

1. Animals

Six-week-old male BALB/c mice were purchased from Damool Science (Daejeon, Korea). The animals were housed in an air-conditioned room with a 12 h light/dark cycle. All animal experiments were performed in accordance with the guidelines for animals care and use of the Jeonbuk national university laboratory animal center (CBN 2019-071).

2. Plant materials and sample preparation

Fresh *A. hookeri* root (100 kg) was purchased from the Hanam farm in Sunchang-gun (Jeonbuk, Republic of Korea). *A. hookeri* was harvested in November 2014 and identified by the Agricultural Technology Research Institute. A voucher specimen (RDAAH15) of *A. hookeri* has been maintained at the Rural Development Administration [28]. Root of *A. hookeri* were hot air-dried for 12 h at 60°C. The dried roots (12 kg) were extracted with distilled water at 100°C for 4 h. *A. hookeri* root extract (AHE) was concentrated under reduced pressure at 60°C for 12 h to obtain concentrated one (7.2 kg). AHE was freeze-dried at −66°C for 72 h under approximately 5~10 mmHg in a vacuum freeze-drying apparatus (4.5 kg).

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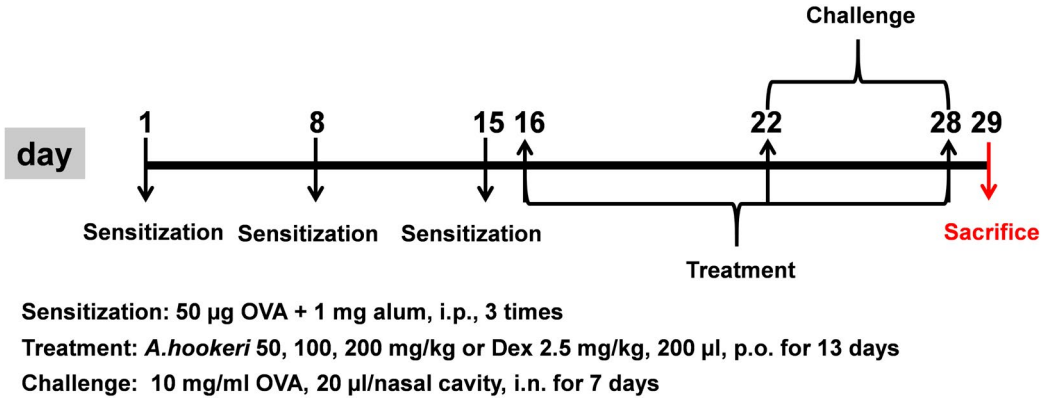


Fig. 1. Schematic representation of the experimental design. In this investigation, mice were intraperitoneally injected with 50 μg OVA and 1 mg alum on days 1, 8, and 15 to induce systemic sensitization. On days 22 to 28, the mice in the OVA group, *A. hookeri* groups, and Dex group received an intranasal challenge in each nasal cavity with 20 μL of 10 mg/mL OVA solution. From days 16 to 28, *A. hookeri* group received 200 μL *A. hookeri* (50, 100, 200 mg/kg), while the Dex group received 200 μL of Dex (2.5 mg/kg). All the test products are administered through oral gavage. OVA-induced sham group mice received the same volume of saline. Mice of the control group were without sensitization, treatment, and challenge. Mice were sacrificed 24h after the last OVA challenge, on day 29.

3. AR murine model establishment and *A. hookeri* treatment

Mice were divided into 6 groups (n = 6) as follows: Control group; OVA group; *A. hookeri* 50 mg/kg treatment group; *A. hookeri* 100 mg/kg treatment group; *A. hookeri* 200 mg/kg treatment group; and dexamethasone (Dex) 2.5 mg/kg treatment group. Briefly, mice were intraperitoneally injected with 50 μg OVA and 1 mg alum on days 1, 8, and 15 to induce systemic sensitization. On days 22 to 28, the mice in the OVA group, *A. hookeri* groups, and Dex group received an intranasal challenge in each nasal cavity with 20 μL of 10 mg/mL OVA solution. From days 16 to 28, *A. hookeri* group received 200 μL *A. hookeri* (50, 100, 200 mg/kg) while the Dex group received 200 μL of Dex (2.5 mg/kg). All the test products are administered through oral gavage. OVA-induced sham group mice received the same volume of saline. Mice of the control group were without sensitization, treatment, and challenge. Mice were sacrificed 24 h after the last OVA challenge, on day 29 (Fig. 1) [29].

4. Observation of nasal symptoms

Mice were intranasally challenged with 10 mg/mL OVA using 20 μL per nasal cavity for 7 consecutive days from day 22 to 28. After the last challenge, the sneezing and nasal rubbing behaviors were video recorded during a 15 min period. All observations and counts were recorded by researchers who were blind to the study objectives.

5. Collection of serum, nasal lavage fluid (NALF) and cell count in NALF

Twenty four hours after the last challenge, the mice were anesthetized with diethyl ether and sacrificed. Blood samples were harvested by an orbital puncture and centrifuged to obtain serum. NALF was collected via an 18-gauge catheter. The trachea was partially resected, a catheter was inserted from the trachea into the nasopharynx, and the nasal passages were gently perfused with 1 mL saline. The NALF were centrifuged at 10,000 RPM/min for 10 min at 4°C. The supernatants were kept at −80°C for enzyme-linked immunosorbent assay (ELISA) analysis. One hundred fifty microliter NALF was centrifuged onto slides at 1,000 RPM/min for 10 min to determine the differential cell counts, using a cytospin device (Centrifuge 5403, Eppendorf, Hamburg, Germany). Then the slides were stained with Diff-Quik staining reagent (1-5-1 Wakinohama-Kaigandori, Chou-Ku, Kobe, Japan) for cell staining, according to the manufacturer's instruction. The numbers of each cell type, including eosinophils, macrophages, neutrophils, and lymphocytes, were identified using a light microscope.

6. Histopathological analysis of lung tissue

The histopathologic observation of lungs was performed according to routine procedures. Briefly, the middle lobe of the lung was removed and fixed in 10% formalin. The specimen was dehydrated with ethyl alcohol and xylene.

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Tissues were embedded in paraffin and cut into 4 μm-thick sections. The sections were stained with hematoxylin and eosin (H&E) to identify the inflammatory cell infiltration and lung architecture. The mucus secretion level was detected by Periodic acid-Schiff (PAS) staining.

7. Histopathological analysis of nasal cavity tissue

Nasal tissue for histopathological analysis was fixed in 10% formalin for 3 days at room temperature. Then, the heads were decalcified in a histological decalcifying agent containing hydrochloric acid and EDTA (National Diagnostics, Atlanta, GA) for 6 days and embedded in paraffin. Samples were cut into 4 μm-thick sections, and the sections were stained with H&E for overall inflammation, PAS for goblet cells hyperplasia, and Giemsa for eosinophil infiltration. Moreover, we used Giemsa stain to investigate the level of eosinophil infiltration in the nasal mucosa, and the number of eosinophils infiltrated into the nasal mucosa were counted.

8. Measurement of OVA-specific immunoglobulins levels in serum

After the mice were sacrificed, blood samples were collected and centrifuged at 10,000 RPM/min for 10 min at 4°C to collect the serum. The levels of OVA-specific IgE (# 439807, BioLegend, USA), OVA-specific IgG1 (#500830, Cayman, USA) and OVA-specific IgG2a (#3015, Chondrex, Redmond, WA, USA) in the serum were quantified using ELISA kits following the manufacturer's instruction.

9. Enzyme-linked Immunosorbent assay

After the last challenge, the mice were sacrificed, and the NALF samples were harvested for further analysis. The levels of Th1 cytokines interferon-gamma (#MIF00, IFN-γ, R&D Systems, USA), interleukin (IL)-12 (# M1270, R&D Systems, USA); Th2 cytokines IL-4 (#M40000B, R&D Systems, USA), IL-5 (#M5000, R&D Systems, USA) and IL-13 (#CSB-E04602m, CUSABIO, USA), total NF-κB p65 (#7174C, Cell Signaling, USA), Phospho -NF-κB p65 (#7173C, Cell Signaling, USA), IκBa (Inhibitor of kB kinase alpha, # MBS728866, MyBioSource, USA), Phospho-IκB (# MBS2600002, MyBioSource, USA) in NALF were measured using the cytokine assay kits according to the manufacturer's instruction.

10. Immunohistochemistry (IHC) Assay

The paraffined nasal mucosa tissue was sectioned into 4 μm, and IHC stain was used to investigate NF-kB p65 expression. IHC was performed using an IHC kit (ab 7481, Abcam Inc., Cambridge, MA, USA). Briefly, after the sections were deparaffinized and dehydrated in gradient ethanol, they were heated by boiling water in 1× antigen retrieval buffer for 10 min. The slides with buffer were cooled at 4°C for 30 min. The sections were then washed in distilled water and PBS twice, 5 min each time. 3% hydrogen peroxide was applied for 10 min to block the endogenous peroxidase. Nonspecific binding was blocked using protein block for 30 min. Sections were incubated with primary antibody (NF-κB p65, 1 : 400 dilutions, #8242, Cell Signaling, USA) overnight at 4°C. Finally, the sections were stained with 3, 3′-diaminobenzidine tetrahydrochloride and counterstained with hematoxylin, dehydrated in ethanol and xylene, and mounted and observed under an optical microscope (×400 magnification).

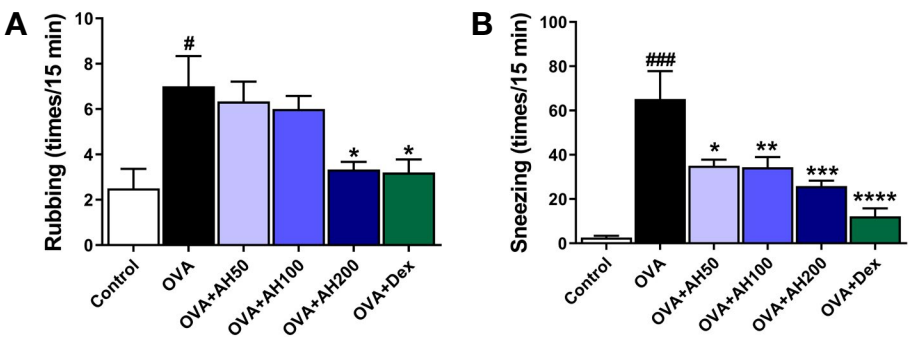


Fig. 2. The nasal symptoms in the murine allergic rhinitis model. (A) Rubbing and (B) Sneezing scores. The values represent the mean ± SEM (n = 6/group). Significant differences at $^{\#}p < 0.05$, $^{\#\#\#}p < 0.001$ compared with the control group; ; ****$p < 0.0001$, ***$p < 0.001$, **$p < 0.01$ and *$p < 0.05$ compared with the OVA group.

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11. Statistical analysis

The results were calculated using Graph Pad Prism software (v 5.0, La Jolla, CA, USA). Data were expressed as means ± SEM and we confirmed their normal distribution with the Shapiro-Wilk normality test. The differences among the groups were analyzed by one-way ANOVA, followed by Tukey's test. Statistical significance was considered at $P < 0.05$.

RESULTS

1. *A. hookeri* ameliorated nasal symptoms in OVA-induced AR mice

The murine model of AR was established via sensitization and challenge by OVA in the present study. Allergic symptoms, including nose rubbing and sneezing, were assessed based on behavioral observations of the mice. The current

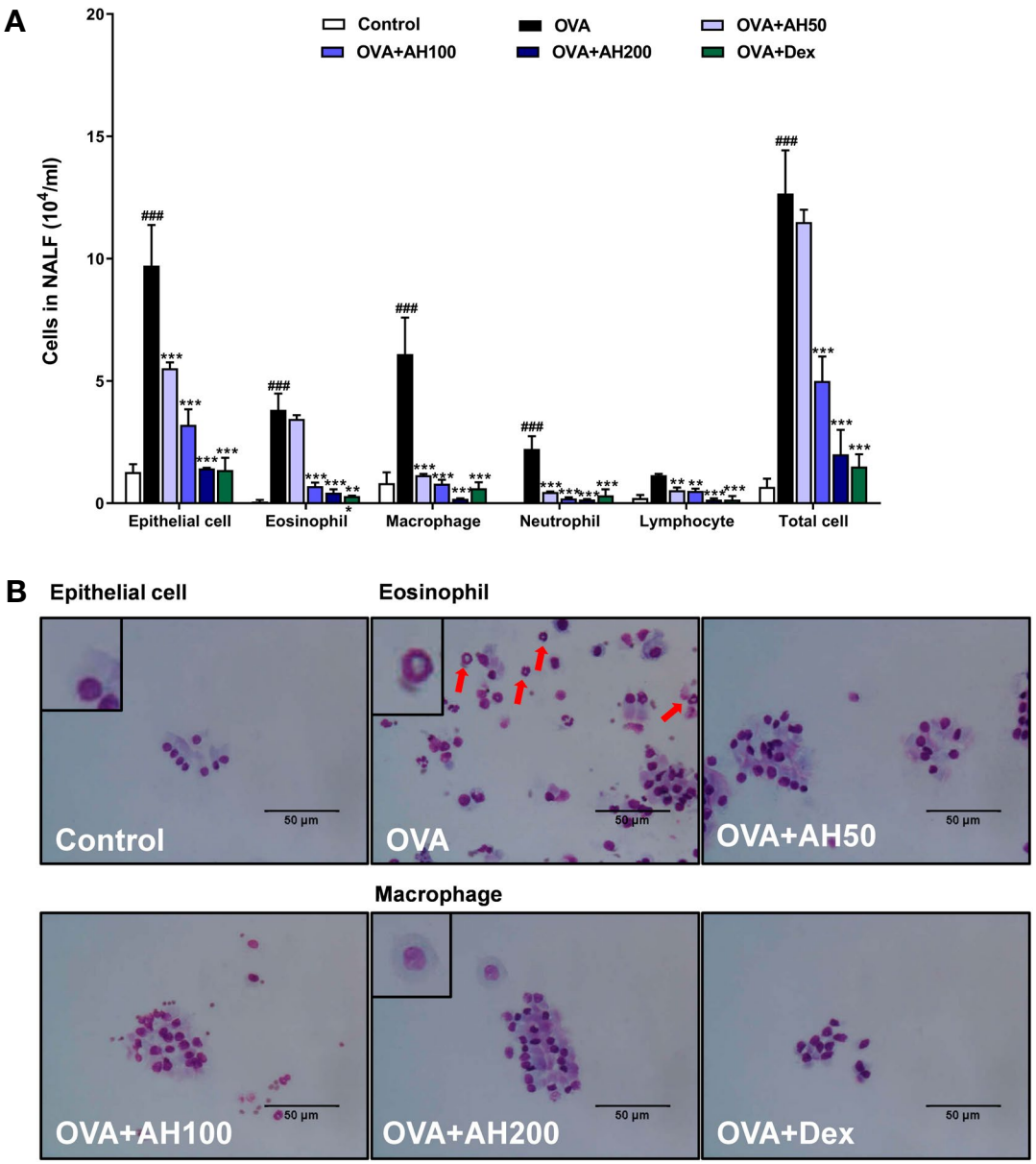


Fig. 3. The number of differential and total cells (A) and the photograph of cytospin cells (B) in nasal lavage fluid (NALF). Red arrows indicated eosinophils. The values represent the mean ± SEM (n = 6/group). Significant differences at $^{###}p < 0.001$ compared with the control group; $^{***}p < 0.001$ and $^{**}p < 0.01$ compared with the OVA group.

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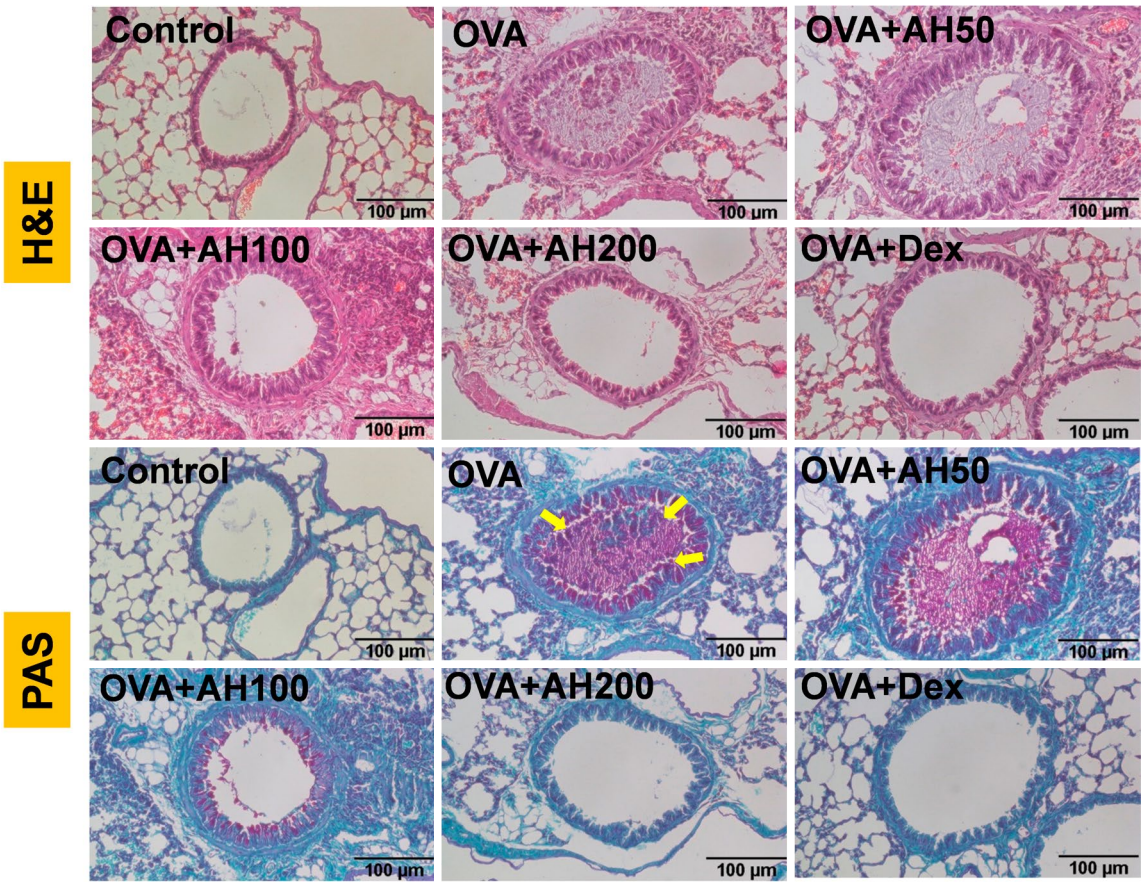


Fig. 4. Histopathological changes of the lung tissues. (A) Hematoxylin and eosin (H&E) stain. (B) Periodic acid-schiff (PAS) stain. Yellow arrows indicated goblet cells. The sections show the inflammation in the lung tissue, and the treatment of *A. hookeri* significantly alleviated inflammation.

study examined the effects of *A. hookeri* on allergic symptoms. As shown in Figure 2, 15 min after the last OVA challenge, the frequency of sneezing was significantly higher in the AR group compared to the control group. In the AR + *A. hookeri* treatment groups, the frequencies of sneezing decreased significantly compared to the AR group. For the nose itching, the number was lower after *A. hookeri* treatment, especially in the high concentration condition. These results indicate that *A. hookeri* could alleviate nasal allergic symptoms in OVA-challenged mice.

2. *A. hookeri* reduced inflammatory cells number in NALF in OVA-induced AR mice

After the last OVA challenge, the NALF samples were collected, and the number of total cells were counted. Total cell and inflammatory cells were notably elevated in the NALF of the OVA group compared with that of the control group. *A. hookeri* and Dex treatment groups had suppressed cell numbers (Fig. 3A). The preparation of NALF cytospin showed inflammatory cells, especially the eosinophils indicated by red arrows, were obviously increased in the OVA group compared with that in the control group. However, when treated with *A. hookeri* and Dex, the number of eosinophils considerably reduced (Fig. 3B).

3. *A. hookeri* alleviated airway inflammation in OVA-induced AR mice

To confirm the potential effect of *A. hookeri* on AR mice, histopathological analysis was performed using H&E and PAS (Fig. 4). The epithelial hyperplasia and mucous hypersecretion were observed in the OVA treated group compared to the lungs of the control group. A high dose (200 mg/kg) of *A. hookeri* treatment ameliorated the OVA-induced morphological changes. Similar observations are

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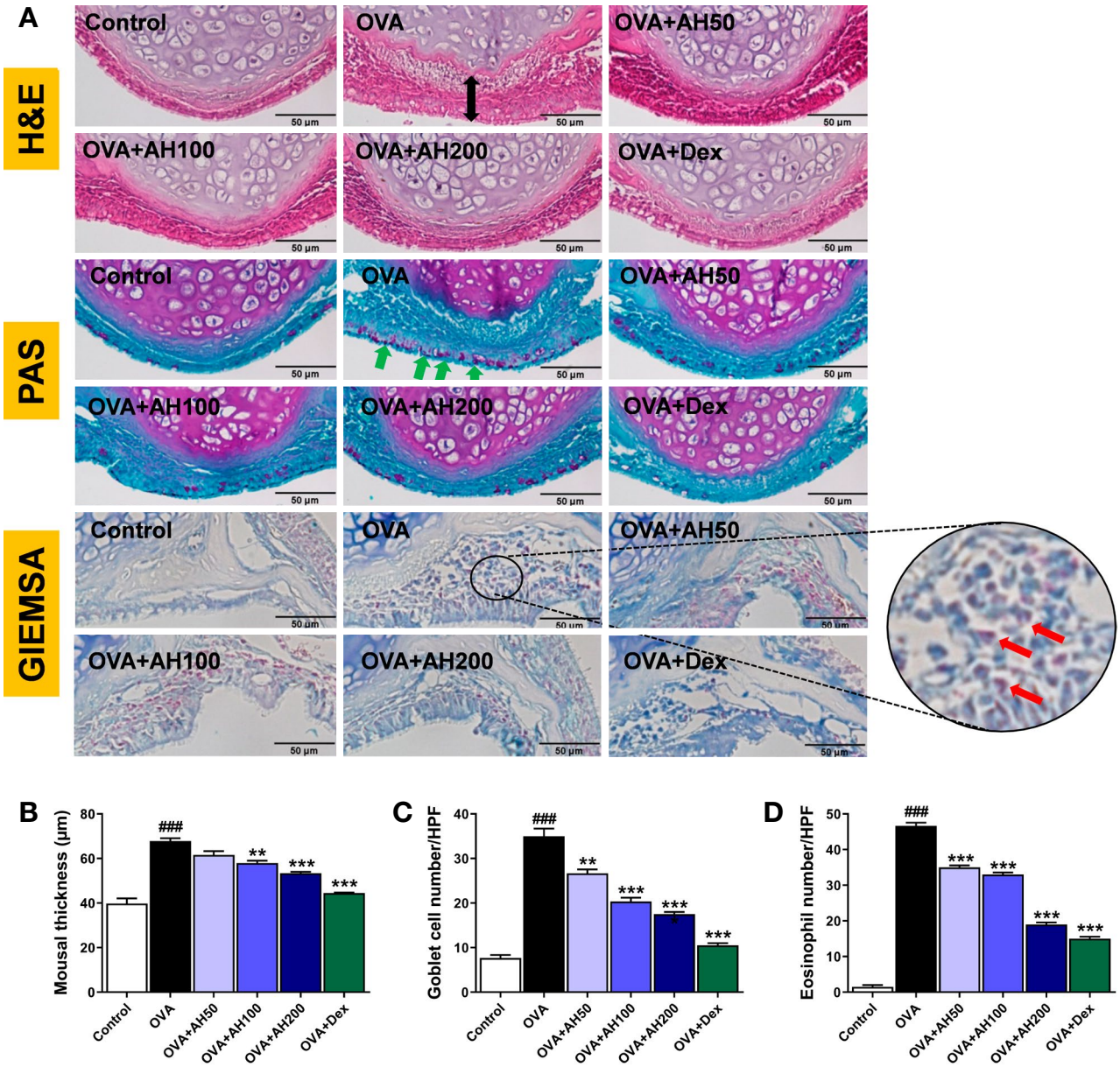


Fig. 5. Histopathological changes of the nasal cavity tissues. H&E staining showing thickness of nasal mucosa (double-headed arrow), goblet cell hyperplasia (green arrows) as shown in PAS staining, and eosinophil infiltration (red arrows) is shown with Giemsa staining. The values represent the mean ± SEM (n = 5/group). Significant differences at $^{###}p < 0.001$ compared with the control group; $^{***}p < 0.001$ and $^{**}p < 0.01$ compared with the OVA group.

found with Dex.

4. *A. hookeri* alleviated nasal cavity inflammation in OVA-induced AR mice

To determine whether *A. hookeri* could suppress the allergic responses in AR, the pathological changes in nasal mucosa were analyzed. As shown in the Figure 5, the nasal mucosa in the control mice showed a normal structure, slight goblet cell hyperplasia, and eosinophil infiltration. The AR group demonstrated a thicker epithelial layer, increased goblet cell hyperplasia, and severe eosinophil infiltration but *A. hookeri* administration significantly decreased the expression of nasal pathological changes in the nasal mucosa.

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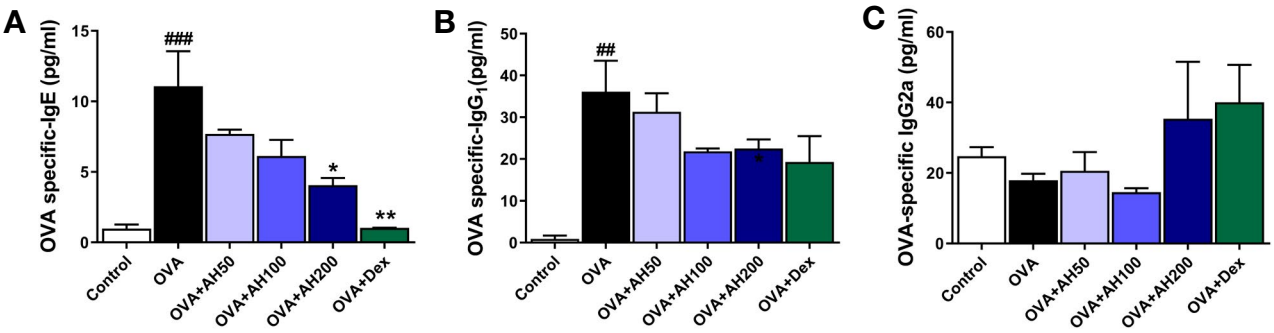


Fig. 6. The levels of ovalbumin (OVA)-specific antibodies level in serum. (A) The level of OVA-specific IgE. (B) The level of OVA-specific IgG1. (C) The level of OVA-specific IgG2a. The values represent the mean ± SEM (n = 6/group). Significant differences at $^{###}p < 0.001$ and $^{##}p < 0.01$ compared with the control group; $^{**}p < 0.01$ and $^{*}p < 0.05$ compared with the OVA group.

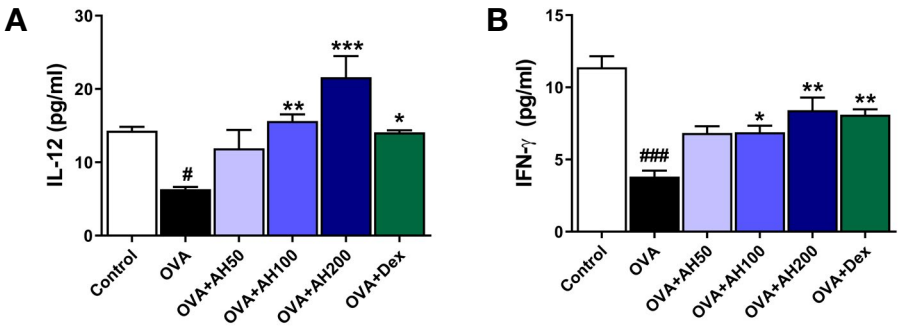


Fig. 7. The levels of Th1 cytokines in NALF. (A) The level of IL-12. (B) The level of IFN-γ. The values represent the mean ± SEM (n = 6/group). Significant differences at $^{###}p < 0.001$ and $^{#}p < 0.05$ compared with the control group; $^{***}p < 0.001$, $^{**}p < 0.01$ and $^{*}p < 0.05$ compared with the OVA group.

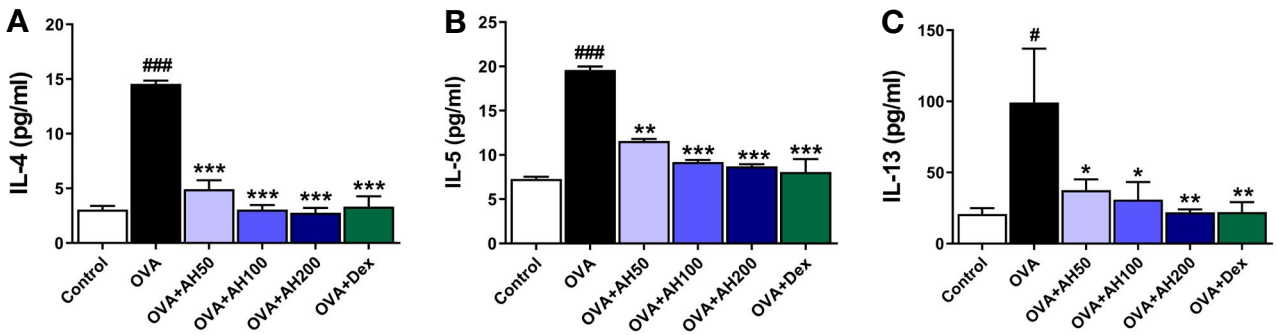


Fig. 8. The levels of Th2 cytokines in NALF. (A) The level of IL-4. (B) The level of IL-5. (C) The level of IL-13. The values represent the mean ± SEM (n = 6/group). Significant differences at $^{###}p < 0.001$ and $^{#}p < 0.05$ compared with the control group; $^{***}p < 0.001$, $^{**}p < 0.01$ and $p < 0.05$ compared with the OVA group.

5. *A. hookeri* modulated OVA-specific Igs in serum in OVA-induced AR mice

The changes in the levels of OVA-specific antibodies, including OVA-specific IgE, OVA-specific IgG1, and OVA-specific IgG2a in the serum, were measured by ELISA kit. The OVA-specific IgE and OVA-specific IgG1 production in the OVA-treated group was significantly increased compared with the control group (Fig. 6A & B). However, in the mice treated with *A. hookeri* and Dex, all levels were decreased compared with the OVA group. The level of OVA-specific IgG2a was decreased in the OVA group, and it re-

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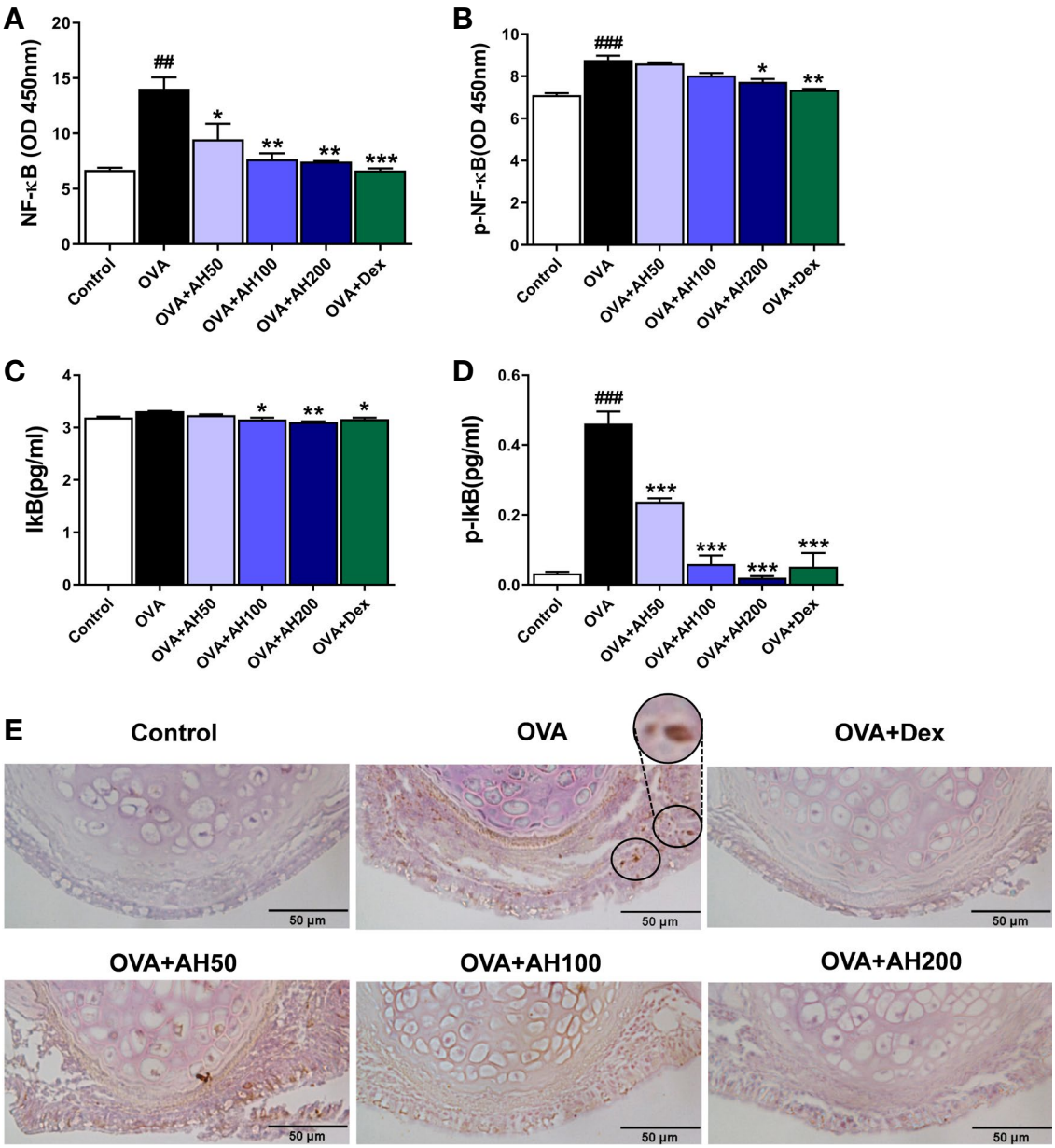


Fig. 9. The levels of NF-κB and IκBα in NALF and IHC stain of NF-κB in nasal mucosa tissue. (A) The level of NF-κB p65. (B) The level of p-NF-κB p65. (C) The level of IκBα. (D) The level of p-IκBα. (E) Positive expression of NF-κB in nasal mucosa tissue by immunohistochemistry (400×). The values represent the mean±SEM (n=6/group). Significant differences at $^{###}p<0.001$ compared with the control group; $^{***}p<0.001$, $^{**}p<0.01$ and $^{*}p<0.05$ compared with the OVA group.

covered after *A. hookeri* and Dex treatment (Fig. 6C).

6. *A. hookeri* upregulated Th1 cytokines in the NALF in OVA-induced AR mice

AR is accompanied by a decrease in Th1 inflammatory factors. The secretion of Th1-related cytokines IL-12 and IFN-γ were assayed in the NALF. As showed in Figure 7A and B, the levels of the cytokines were significantly decreased in the OVA-treated group, whereas the expressions were increased in the *A. hookeri* and Dex treatment.

7. *A. hookeri* downregulated Th2 cytokines in NALF in OVA-induced AR mice

Th2 cells are involved in allergic inflammation responses.

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The changes in the expression of Th2-related cytokines IL-4 (Fig. 8A), IL-5 (Fig. 8B) and IL-13 (Fig. 8C) were assayed in the NALF by ELISA kit. The cytokines were increased during the inflammation conditions in the OVA group. Notably, the levels of these cytokines decreased following the treatment with *A. hookeri* and Dex.

8. *A. hookeri* modulated inflammation response via the NF-κB pathway in OVA-induced AR mice

The NF-κB signaling pathway plays an important role in AR. ELISA and IHC assay were performed to investigate whether *A. hookeri* could alleviate AR by intervening in the NF-κB signaling pathway. As shown in Figure 9A~D, OVA caused the up-regulation of the NF-κB/IκBα signaling-related protein expressions in mice, suggesting OVA could induce AR through active NF-κB/IκBα. In contrast, *A. hookeri* showed inhibition of the expression levels of the NF-κB/IκBα pathway in the OVA-induced AR mice. To further verify our results, IHC staining was performed. The expression of NF-κB was significantly elevated in the AR mice compared with the normal control group. In the *A. hookeri* treated groups, the NF-κB expression was diminished (Fig. 9E).

DISCUSSION

AR is a chronic inflammation disorder of the nasal cavity and an increasing challenge to public health worldwide, affecting nearly 30% of the world population [30]. People with AR may experience headaches, daytime fatigue, sleep disturbance, irritability, and more [31]. Current AR-alleviating medications have low clinical efficacy and undesirable side effects, which motivates us to look for more effective and safer alternatives [32]. In recent years, the traditional medicines that could improve multifactorial diseases, including AR, have been considered a good therapeutic option [33]. *A. hookeri* is widely consumed in China and Korea and has been reported to have an anti-inflammation effect. In the present study, we assessed the effect of *A. hookeri* on nasal allergic inflammation and investigated the latent mechanism. We found that *A. hookeri* could alleviate the inflammation in the OVA-induced AR mouse via regulation of the Th1/Th2 imbalance.

In the development of AR, the release and infiltration of inflammatory mediators secreted by immune cells play an important role [34]. Particularly in allergic response, T lymphocytes are key immune cells, especially Th1 and Th2 cells [35]. The imbalance between Th2 and Th1 polarization results in allergic phenotype features such as asthma, AR, and atopic dermatitis [36-38]. IFN-γ is a Th1 cytokine produced by Th1 lymphocytes that performs cell-mediated immunity in auto-immune diseases [39]. Here, we measured the levels of IFN-γ and IL-12 in the NALF, and the results indicated that the production of Th1 cytokines was decreased in the OVA-treated group, but *A. hookeri* treatment increased the levels significantly. Recently accumulated evidence indicates that Th2 cells play a triggering role in the activation and/or recruitment of IgE antibody producing B cells, which are involved in allergic inflammation [40]. Moreover, Th2 cytokines such as IL-4, IL-5, and IL-13 are important mediators in the pathogenesis of AR [2]. Our present study showed that the NALF in the OVA-treated group had significantly increased Th2 cytokines, whereas *A. hookeri* could suppress the secretion to mediate anti-inflammation. In the AR response, various inflammation cells infiltrate into the nasal mucosa and airway, and goblet cell hyperplasia occurs. The indicators that *A. hookeri* can attenuate inflammation in the AR mouse model were that *A. hookeri* alleviated the histopathological changes in the tissue. The results showed that *A. hookeri* effectivity improved the pathological changes of tissues and inhibited goblet cell proliferation and infiltration of eosinophils.

NF-κB is a transcription factor and has a function in immune response. It can stimulate the expression of pro-inflammation cytokines. Inhibitor-κBα (IκBα), a kind of inhibitory protein called inhibitor of NF-κB, keeps NF-κB inactive in the cytoplasm in non-stimulated cells [41]. To evaluate the mechanisms of inhibition of *A. hookeri* on the expression of pro-inflammation cytokines, we examined the effect of *A. hookeri* on the NF-κB pathway. In the present study, *A. hookeri* decreased the production of NF-κB p65 and phospho-NF-κB, and also the degradation of phosphorylated IκBα. *A. hookeri* treatment blocked NF-κB translocation into the nucleus, and prevented IκBα degradation, both of which are related to inflammatory response. Consequently, the production of several cytokines, including IFN-γ, IL-12, IL-4, IL-5 and IL-13, is reduced. These results indicate that inhibiting the expression of NF-κB could alleviate nasal inflammation in the AR mouse model.

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CONCLUSION

In conclusion, the study has investigated the anti-allergic rhinitis effect of *A. hookeri* in an OVA-induced AR mouse model. Our results have shown that *A. hookeri* could inhibit inflammation in AR through the NF-κB pathway. Moreover, *A. hookeri* attenuated the inflammation in the nasal cavity and lung tissue, and suppressed Th2 related cytokines, but it activated Th1 cytokines in the NALF and modulated the expression the OVA-specific antibodies in the serum, suggesting that *A. hookeri* could be an effective therapeutic candidate for AR.

CONFLICTS OF INTEREST

The authors declare no conflict of interests.

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Chang Ho Song and Ok Hee Chai designed this study and proofread the article. Yan Jing Fan, Juan Jin, Chun Hua Piao, and Thi Van Nguyen contribute to the animal experiments. Yan Jing Fan contributed to the initial draft of the manuscript and interpretation of experimental results.

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