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Anti-inflammatory effects of linalool on ovalbumin-induced pulmonary inflammation

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ABSTRACT

Linalool is a natural product present in fruits and aromatic plants with biological activities. Researchers have reported that the inhalation of linalool exerts anti-inflammatory activities. In this study, we examined the therapeutic effects of linalool on airway inflammation and mucus overproduction in mice with allergic asthma. Oral administration of linalool significantly inhibited the levels of eosinophil numbers, Th2 cytokines and immunoglobulin E (IgE) caused by ovalbumin (OVA) exposure. Linalool exerted preventive effects against the influx of inflammatory cells and mucus hypersecretion in the lung tissues. Linalool also dose-dependently decreased the levels of inducible nitric oxide synthase (iNOS) expression and protein kinase B (AKT) activation in the lung tissues. Linalool effectively downregulated the activation of mitogen-activated protein kinases (MAPKs) and nuclear factor-κB (NF-κB) caused by OVA exposure. Furthermore, linalool exerted inhibitory effect on OVA-induced airway hyperresponsiveness (AHR). In the in vitro study, the increased secretion of MCP-1 was attenuated with linalool treatment in lipopolysaccharide (LPS)-stimulated H292 airway epithelial cells. In conclusion, linalool effectively exerts a protective role in OVA-induced airway inflammation and mucus hypersecretion, and its protective effects are closely related to the downregulation of inflammatory mediators and MAPKs/NF-κB signaling.

1. Introduction

Allergic asthma is an important cause of morbidity and mortality around the world [1], and its prevalence is continuously increasing [2]. Airway inflammation and mucus hypersecretion are the main etiological causes of allergic asthma [3]. The elevated levels of Th2 cytokines, such as IL-4, IL-5 and IL-13 are closely linked with allergic airway inflammation by the upregulation of eosinophil recruitment, IgE overproduction and airway hyper-responsiveness (AHR) [4,5]. It is believed that mucus hypersecretion contributes to airway obstruction and hyperresponsiveness [6,7]. The increased level of monocyte chemoattractant protein-1 (MCP-1) leads to inflammatory cells recruitment [8]. The increase of inducible nitric oxide synthase (iNOS) expression and AKT activation was upregulated in the lungs of OVA-induced asthmatic mouse [9,10]. Mitogen-activated protein kinase (MAPK) and nuclear factor-NF-κB signaling are the key mediators of inflammatory diseases, and the extended activation of MAPK and NF-κB has been confirmed in allergic asthma [11,12].

Natural fruits and herbs and their components can prevent the spread of inflammatory diseases [13]. Linalool is a natural monoterpene commonly present in fruits, aromatic plants, spices and tea such as oranges, tomatoes, peaches, lemon balm, basil, and green tea

Abbreviations: BALF, bronchoalveolar lavage fluid; IgE, immunoglobulin E; IκB, inhibitor of NF-κB; IL-4, *interleukin-4*; IL-5, *interleukin-5*; IL-13, *interleukin-13*; MCP-1, monocyte chemoattractant protein-1; NF-κB, nuclear factor-κB; OVA, ovalbumin

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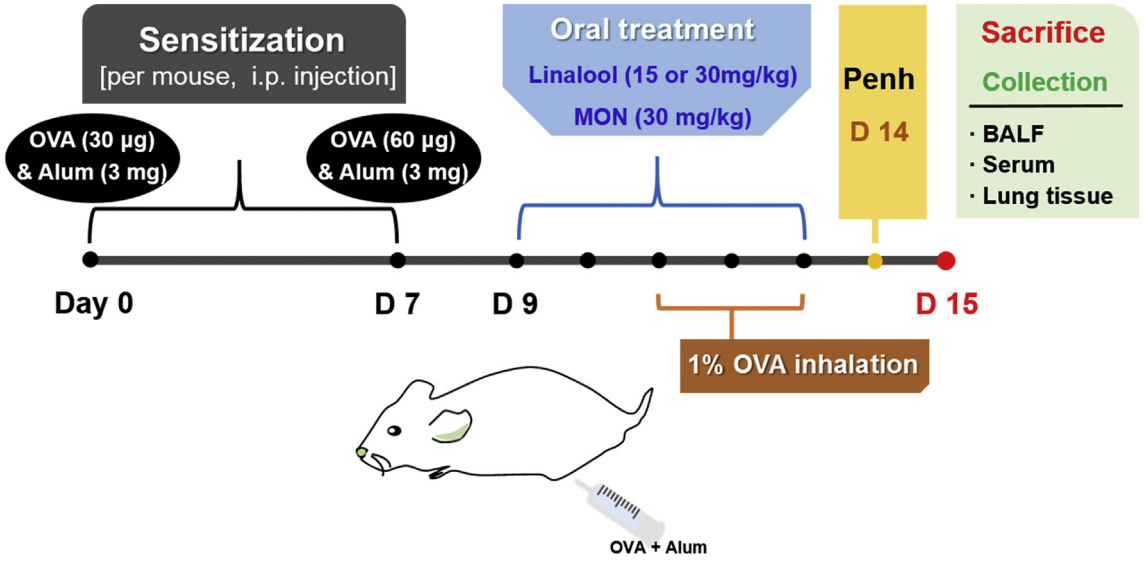

Fig. 1. Experimental procedure. BALB/c mice were divided into four groups ($n = 6$ in each group) and sensitized by OVA and Alum injection (i.p.) on days 0 and 7 and challenged by a 1% OVA aerosol on days 11 to 13. The mice were orally administered with linalool (15 or 30 mg/kg) or MON (30 mg/kg) from days 9 to 13. On day 15, the mice were sacrificed, and the BALF, serum and lung tissues were acquired.

[14–18]. The essential oils including linalool in several plant species have been used in traditional medicine and cooking and have pharmacological activities, such as antinociception and anticonvulsant effects [19–21]. Recently, the anti-inflammatory effect of linalool from the leaf essential oil of *C. osmophloeum* Kanehira was identified in an endotoxin-injected animal model [22]. Simultaneously, other recent studies reported that linalool ameliorates cigarette smoke (CS)-induced pulmonary inflammation [23]. Linalool also exerts anti-inflammatory effects in lipopolysaccharide (LPS)-stimulated inflammatory response in BV2 microglial cells [24]. However, the protective effect of linalool has not been revealed yet in allergic asthma. Therefore, we evaluated the effects of linalool on ovalbumin (OVA)-induced pulmonary inflammation in mice.

2. Materials and methods

2.1. Reagents

Ovalbumin (A5503) and linalool (L2602, PubChem 24896318) were purchased from Sigma Aldrich (St. Louis, MO, USA). Alum was purchased from ThermoFisher Scientific (#77161, Rockford, IL).

2.2. Induction of ovalbumin (OVA) and alum-induced pulmonary inflammation in murine models

Female BALB/c mice (SPF, six weeks old) were purchased from the Koatech Laboratory Animal Center (Pyeongtaek-si, Korea). The experimental procedure was modified by the method of Park et al. [25]. The mice randomly were divided into five groups: (A) the normal control (NC), (B) the ovalbumin sensitization and challenge group (OVA), (C) the OVA + monteleukast (30 mg/kg, per oral) group (MON), (D) the OVA + linalool (15 mg/kg, per oral) group (LINA 15), and (E) the OVA + linalool (30 mg/kg, per oral) group (LINA 30). Monteleukast and lilanool were dissolved with 2% DMSO + 2% Tween-20 in phosphate-buffered saline (PBS), and were administered orally on days 11–13. On day 0 and 7, the mice were sensitized twice intraperitoneally with 0.2 ml of ovalbumin (OVA) (day 0, 30 μg; day 7, 60 μg) and 3 mg Alum. On days 11–13, OVA inhalation was performed using a nebulizer (1% OVA: alum-free saline solution, 60 min/day). The oral administration of linalool or montelukast (MON) was given for five consecutive days (from day 9 to 13). On day 14, the level of enhanced pause (Penh) value was evaluated using whole body plethysmograph (OCP3000, Allmedicus, Korea). The experimental protocol is presented in Fig. 1. All experimental procedures involving animals were approved by the Korea Research Institute of Bioscience and Biotechnology (KRIBB) IACUC and were performed in compliance with the National Institutes of Health Guidelines for the Care and Use of Laboratory Animals and Korean National Laws for Animal Welfare.

2.3. BALF collection and cell counting

To collect the BALF, the cannula was inserted into the trachea, and 0.5 ml of PBS was inserted into the airway two times [26]. 0.1 ml BALF sample of each group was centrifuged on a glass slide with a Cytospin centrifuge (1500 rpm for 5 min.) and stained with Diff-Quik stain kit to distinguish different cell types. Using microscopy, the different cells were distinguished, and cell counts were conducted.

2.4. Quantitative analyses of IL-4, IL-5, IL-13 and IgE production

The levels of IL-4, IL-5 and IL-13 in the BALF were determined by commercial ELISA kit (R&D Systems Inc., Minneapolis, MN, USA, cat. no M4000B, M5000 and M1300CB). The blood was collected 48 h after the final administration of linalool and montelukast (MON), and the serum was prepared. Total IgE and OVA-specific IgE measurement was conducted using each ELISA kit (Total IgE ELISA kit, BioLegend, Inc., cat. no. 432404; OVA-specific IgE, R&D Systems Inc., cat no. 439807).

2.5. Western blot analysis

Lung tissues were homogenized using a homogenizer in a cell lysis reagent containing a phosphatase and protease inhibitor (Roche, Basel, Switzerland). Western blot analysis was subjected to detect MCP-1, ERK, phospho (p)-JNK, JNK, p38, NF-κB p65 (Santa Cruz, TX, USA), p-ERK, p-p38, p-NF-κB p65, anti-IκBα or anti-β-actin (Cell Signaling, MA, USA). Each blot was developed with ECL detection system (Thermo Fisher Scientific, Inc., Rockford, IL).

2.6. Histopathological studies

After the collection of the BALF and serum, the lung tissues from mice were fixed in a 10% formalin solution and embedded in paraffin.

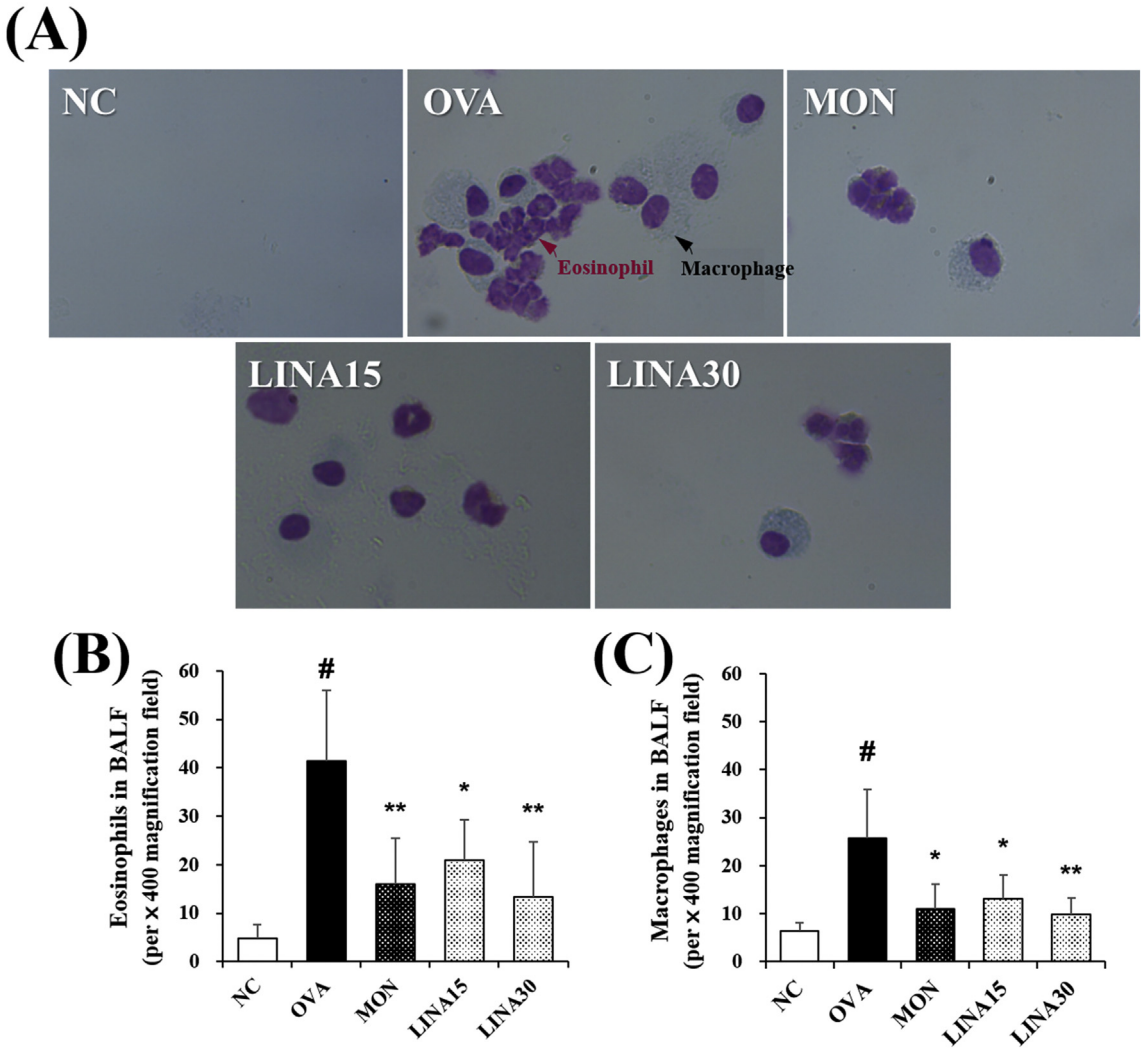

Fig. 2. Effect of linalool on the numbers of inflammatory cell count in the BALF. (A) The microscope images of inflammatory cells (magnification, x400). (B) The count of eosinophils and macrophages. Data are expressed as the means ± standard deviation (n = 6). NC, normal control mice; OVA, mice administered ovalbumin (OVA); MON, mice administered montelukast (30 mg/kg) + OVA; LI15 or LI30, mice administered linalool (15 or 30 mg/kg) + OVA. $^{\#}P < 0.01$, vs. NC group; $^{*}P < 0.05$ and $^{**}P < 0.01$, vs. OVA-induced group. NC, normal control group; OVA, ovalbumin; MON, montelukast; LI or LINA, linalool.

For histopathological evaluation, paraffin-embedded blocks were sectioned at a 4 μm thickness and were stained with H&E and PAS stain solutions. The degree of inflammatory cell influx and mucus hypersecretion in each group were examined by two independent observers using a scoring standard [H&E staining evaluation: (0, no influx of inflammatory cells; 0.5, a few influx; 1, moderate influx; 2, large influx; PAS staining evaluation: (0, no mucus; 1, mild mucus; 2, moderate mucus; 3, distinct mucus; 4, severe mucus).

2.7. Cell culture

H292 airway epithelial cells were purchased from the ATCC (Manassas, VA, USA) and cultured in RPMI 1640 supplemented with 10% fetal bovine serum (FBS) in the presence of 100 U/ml penicillin, and 100 μg/ml streptomycin in a humidified chamber in 5% CO_2 at 37 °C. Dimethyl sulfoxide (DMSO) was used to dilute linalool (linalool stock solution, 80 mg/ml). Final DMSO concentration did not exceed 1%. To examine the effect of linalool on MCP-1 secretion, the cells were pretreated with linalool (2.5, 5, 10, 20, 40 and 80 μg/ml) 1 h before the incubation with lipopolysaccharide LPS (10 μg/ml) for 24 h. MCP-1 ELISA assay was performed using the cell culture supernatant.

2.8. Statistical analysis

The values are presented as the means ± standard deviation (S.D.). The statistical significance was determined using ANOVA followed by a multiple comparison test with Dunnett's adjustment. Data with values of $p < 0.05$ were accepted as statistically significant.

3. Results

3.1. Effect of linalool on the numbers of eosinophils and macrophages in OVA-challenged mice

The infiltration of eosinophils and macrophages has been characterized in allergic asthma and observed in OVA-induced airway inflammation in mice [27,28]. To examine the regulatory effect of linalool on eosinophil/macrophage recruitment, we counted the number of these cells in the BALF using Diff-Quik® staining and a microscope. As shown in Fig. 2, a remarkable increase of eosinophils and macrophages was detected in the OVA-exposed mice. However, the numbers of these cells were effectively decreased with linalool treatment. The inhibitory effects of linalool were similar to those of MON (Fig. 2). These findings imply that the linalool treatment reduced eosinophil/macrophage

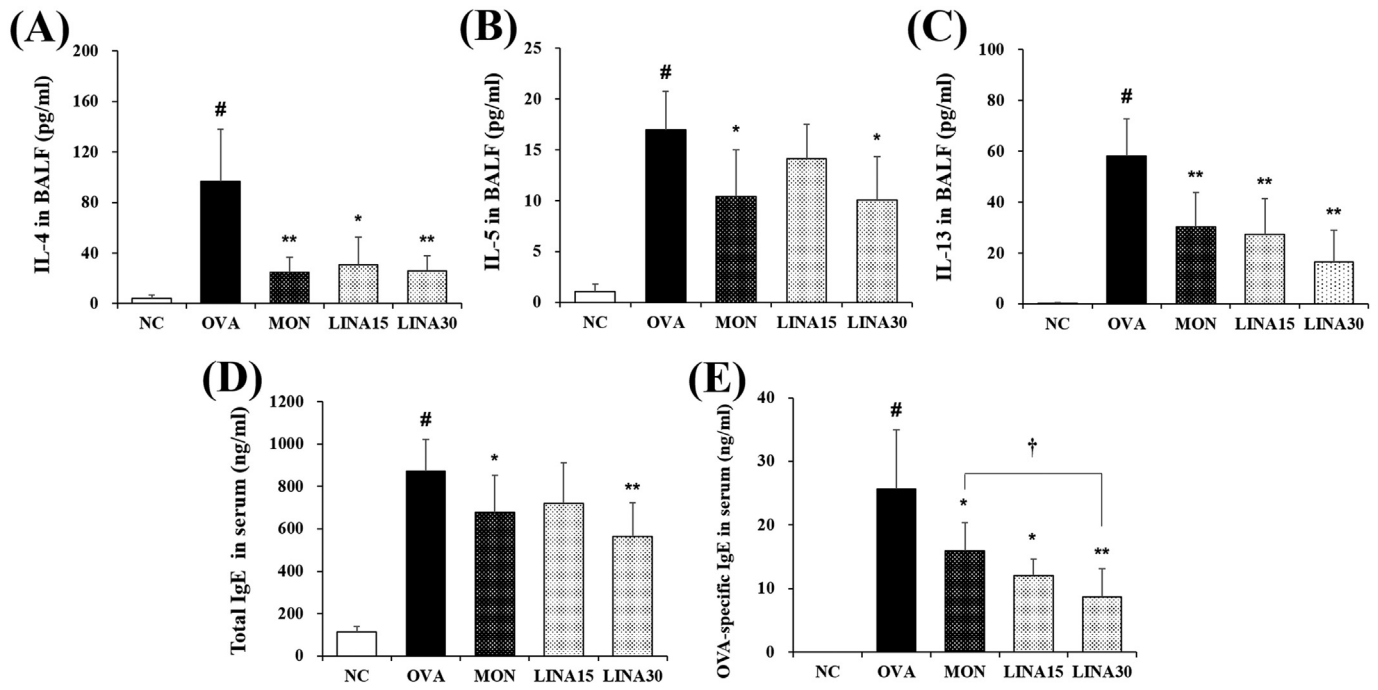

Fig. 3. Effect of linalool on the levels of Th2 cytokines and IgE in the BALF and serum. (A-C) BALF cytokines IL-4, IL-5 and IL-13 were determined by ELISA kits. (D–E) Serum total IgE and OVA-specific IgE were measured by ELISA kits. The absorbance was measured at 450 nm with a microplate reader. $^{\#}P < 0.05$ vs. NC group; $^{*}P < 0.05$ and $^{**}P < 0.01$ vs. OVA-induced group; $^{\dagger}P < 0.05$ vs. MON group. IL-4, interleukin-4; IL-5, interleukin-5; IL-13, interleukin-13; IgE, immunoglobulin E.

recruitment induced by OVA.

3.2. Effect of linalool on the production of Th2 cytokines and IgE in OVA-challenged mice

The high levels of Th2 cytokines and IgE are closely correlated to increased airway inflammation in asthma. Thus, we first evaluated the effect of linalool on OVA-induced IL-4, IL-5 and IL-13 using ELISA. The remarkable increase of these cytokines was shown in the BALF of OVA-exposed mice compared with the NC mice (Fig. 3A-C). However, linalool treatment significantly reduced this increase. We next determined the effectiveness of linalool against IgE secretion. As shown in Fig. 3, the levels of total IgE and OVA-specific IgE were markedly higher in the OVA-exposed mice. However, these levels were reduced with linalool treatment (Fig. 3D and E). Interestingly, the inhibitory effect of linalool (30 mg/kg) on OVA-specific IgE production was better than that of the 30 mg/kg MON ($^{\dagger}p < 0.05$, vs. MON group).

3.3. Effect of linalool on inflammatory cell influx and MCP-1 production in OVA-challenged mice

We examined the effect of linalool on inflammatory cell influx using H&E staining. The degree of cell influx near the peribronchial lesions in the lungs was notably elevated in the OVA-exposed mice compared to that observed for the NC mice (Fig. 4A). However, the linalool-treated mice showed an effectively reduced infiltration. MCP-1, a key chemokine, is released from various cell types, such as epithelial cells and macrophages, and its upregulation is associated with airway inflammation by eosinophils, lymphocyte and monocyte chemotactic activities [29–31]. Therefore, the regulation of MCP-1 exerts a protective effect in allergic airway inflammation. As shown in Fig. 4C, a significant increase of MCP-1 production was shown in the BALF of OVA-exposed mice compared to that of the NC group, whereas this trend was effectively suppressed with linalool treatment (Fig. 4C). Similar to the results of Fig. 4C, the increased expression of MCP-1 in the lung tissues of OVA-exposed mice was notably reduced and minimal in the linalool-treated group (Fig. 4D). To further evaluate the regulatory effect of linalool on MCP-1 in activated-airway epithelial cells, an in vitro study with linalool was done. Lipopolysaccharide (LPS) was used to activate H292 cells. The level of MCP-1 secretion was highly elevated in the LPS-stimulated H292 cells (Fig. 4F). However, pretreatment with linalool significantly reduced the level of MCP-1. We confirmed the regulatory effect of linalool on the recruitment of inflammatory cell and on the production of MCP-1. These findings suggest that linalool may be helpful in regulating inflammatory cells influx in inflammatory diseases including allergic asthma.

3.4. Effect of linalool on mucus hypersecretion in OVA-challenged mice

The respiratory tract mucus overproduction leads to airway inflammatory response and airway obstruction, and is one of the important cause of death from asthma [32]. In the present study, we investigated the regulatory effect of linalool on the mucus production in the lung tissues of the OVA-challenged mice. The PAS staining results show that the mucus overproduction of the OVA-challenged group was remarkably upregulated. However, treatment with linalool notably downregulated the mucus production (Fig. 5).

3.5. Effect of linalool on iNOS expression in OVA-challenged mice

The upregulation of iNOS is significantly related to inflammatory response in the lungs of OVA-exposed mice [12]. Based on these results, we examined the regulatory effect of linalool on OVA-induced iNOS expression using western blot analysis. The results show that the level of iNOS was significantly upregulated in the lung lysates of OVA-exposure mice (Fig. 6). However, the increased expression of iNOS was decreased with linalool treatment in a concentration-dependent manner.

3.6. Effect of linalool on AKT activation in OVA-challenged mice

The AKT activation is well established in allergic asthma [33], and

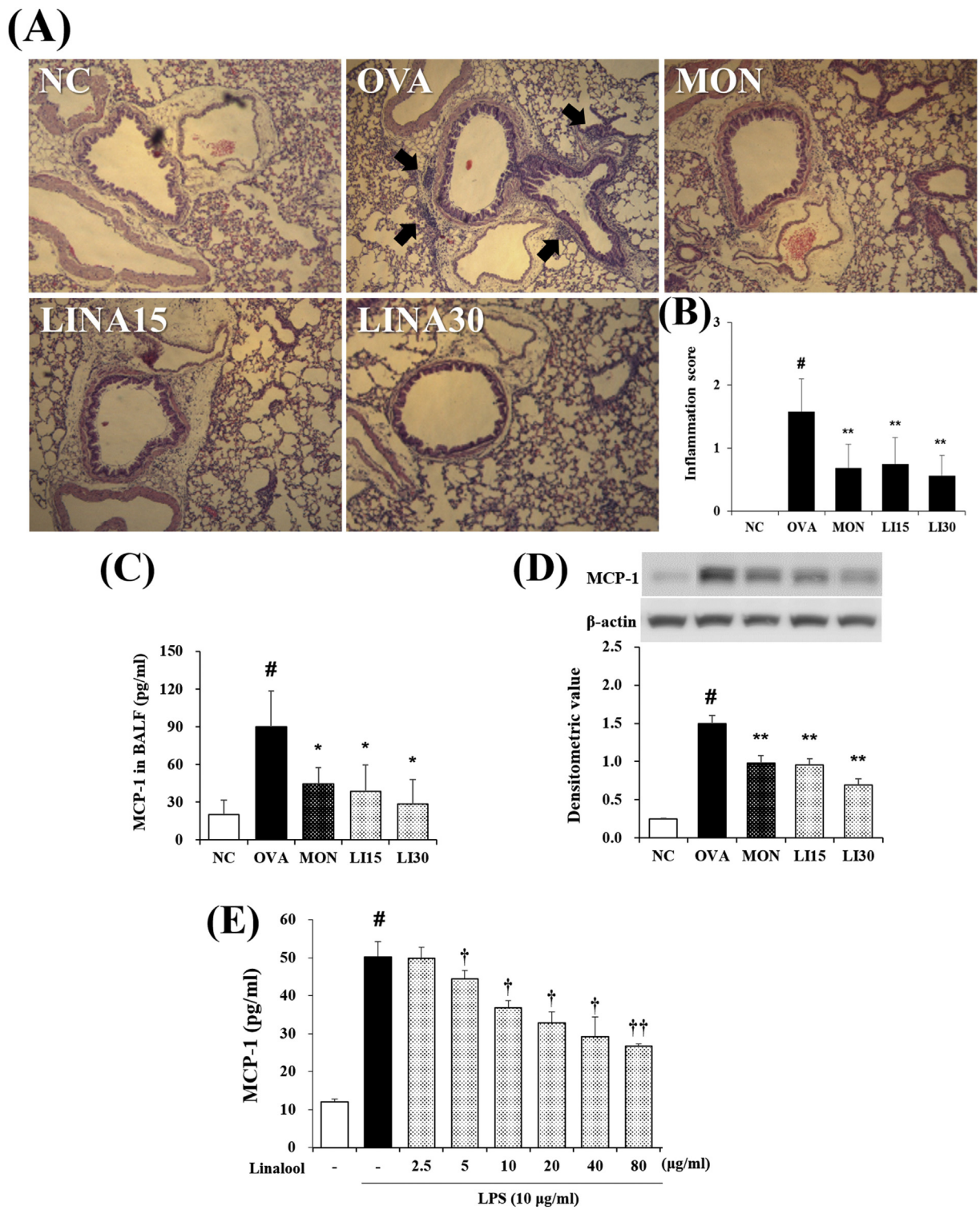

Fig. 4. Effect of linalool on the influx of inflammatory cells and the production of MCP-1 in the lungs. (A) H&E staining was used to assess the inflammatory cell influx (Peribronchial lesion, magnification x100). (B) Quantitative analysis of airway inflammation. (C) The production of MCP-1 was determined with ELISA. (D) The expression level of MCP-1 was determined with western blot analysis and quantitative analysis of MCP-1 expression was performed by densitometric analysis. (E) Effect of linalool on the production of MCP-1 in LPS-stimulated H292 airway epithelial cells. $^{\#}P < 0.05$ vs. NC group; $^{*}P < 0.05$ and $^{**}P < 0.01$ vs. OVA group; $^{\dagger}P < 0.05$ and $^{\dagger\dagger}P < 0.01$ vs. LPS group. MCP-1, monocyte chemoattractant protein-1.

an AKT inhibitor exerts a protective role in OVA-induced inflammatory cells infiltration [34]. Therefore, we examined whether linalool affects the level of OVA-induced AKT activation. In the present study, the upregulation of AKT activation was detected in the lungs of OVA-exposure mice. However, this increased level was effectively reduced by linalool treatment (Fig. 7). The inhibitory activity of linalool on AKT activation was higher than that of MON.

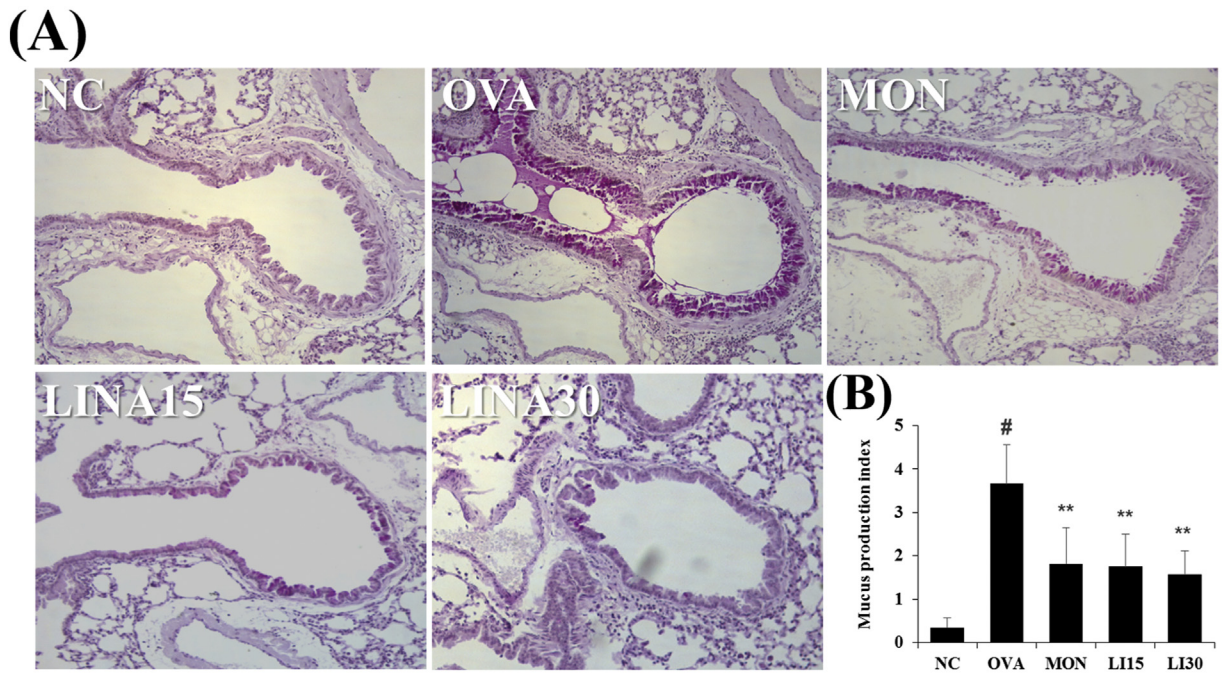

Fig. 5. Effect of linalool on the production of mucus in the lungs. (A) The levels of mucus production were confirmed with PAS staining (magnification x200). (B) Quantitative analysis of mucus production. $^{\#}P < 0.05$ vs. NC group; $^{**}P < 0.01$ vs. OVA group.

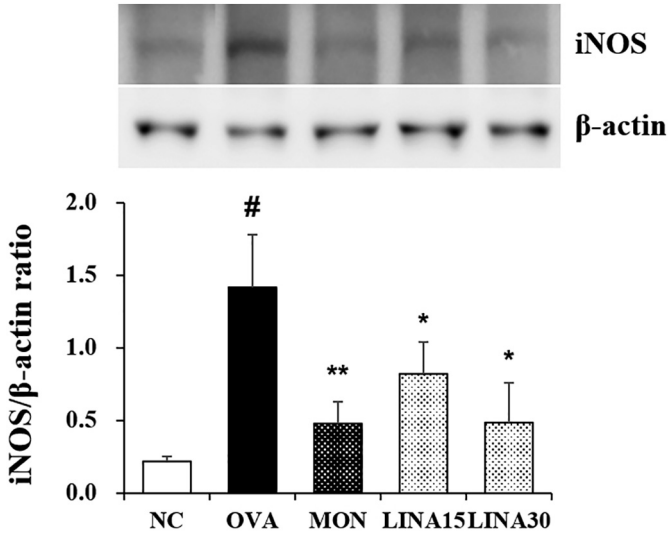

Fig. 6. Effect of linalool on the expression of iNOS in the lungs. The expression level of iNOS was determined with western blot analysis, and quantitative analysis of iNOS was performed by densitometric analysis. $^{\#}P < 0.05$ vs. NC group; $^{*}P < 0.05$ and $^{**}P < 0.01$vs. OVA group. β-actin was used as an internal control. iNOS, inducible nitric oxide.

3.7. Effect of linalool on MAPKs and NF-κB activation in OVA-induced pulmonary inflammation

The MAPKs and NF-κB signaling pathways are important in allergic airway inflammation and, therefore we evaluated the regulatory effects of linalool on the activation of these molecules. The increased level of ERK, JNK and p38 activation was observed in the lungs of OVA-exposure mice group (Fig. 8A). Notably, treatment with linalool effectively blocked the activation of these molecules. Next, we examined the effect of linalool in OVA-induced NF-κB activation. NF-κB activation is upregulated by IκBα and NF-κB p65 phosphorylation [35–37]. Thus, we explored whether linalool leads to downregulation of phosphorylation of those molecules. The results show that increase of IκBα and NF-κB p65 phosphorylation was detected in the lungs of OVA-exposure mice group compared with that of the NC group (Fig. 8C). Treatment with linalool effectively suppressed these levels.

3.8. Effect of linalool on Penh value in OVA-challenged mice

To evaluate the effect of linalool on OVA-induced airway hyperresponsiveness (AHR), we assessed the level of enhanced pause (Penh) value on day 14. Mice were exposed to methacholine (MCH) aerosols for 3 min with an ultrasonic nebulizer (MCH dose; 12.5 mg/ml and 25 mg/ml). As shown in Fig. 9, the levels of Penh value were upregulated with the exposure of MCH in a concentration-dependent manners. However, linalool administration led to a decrease. These results suggest that the protective effect of linalool on airway inflammatory and excessive mucus production may affect amelioration of AHR.

4. Discussion

The severity of asthmatic symptoms is closely related to increased Th2 cytokines, eosinophil influx and IgE overproduction. IL-4 has a critical role in the inhibition of Th1 cell generation, the promotion of Th2 cell development and the stimulation of activated B-cell proliferation [38]. IL-5 leads to eosinophil survival and differentiation [27]. IL-13 causes the secretion of IgE by B cells [27]. IgE triggers inflammatory cells including eosinophils and mast cells to produce inflammatory molecules, such as IL-4, IL-5 and IL-13 [39]. Therefore, the modulation of Th2 cytokines, eosinophils influx and IgE has important medicinal values. In this study, linalool effectively inhibited the levels of eosinophil recruitment, Th2 cytokines and IgE in mice with allergic asthma (Figs. 2 and 3). Our results indicate that linalool has the potential to ameliorate pulmonary inflammatory response in allergic asthma.

Targeting MCP-1 may be useful in the amelioration of airway inflammation [30]. In this study, we confirmed that linalool exerts the regulatory effect on MCP-1 overproduction (Fig. 4A - D). Linalool also exerted inhibitory effects on MCP-1 secretion in activated macrophage cells (Fig. 4E). The regulatory effect of linalool on MCP-1 production is

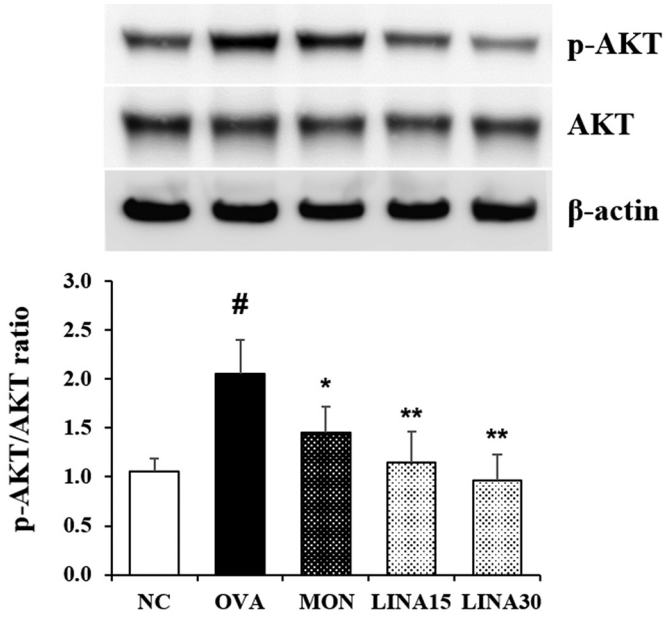

Fig. 7. Effect of linalool on the activation of AKT in the lungs. The phosphorylation level of AKT was determined with western blot analysis, and quantitative analysis of AKT phosphorylation was performed by densitometric analysis. $^{\#}P < 0.05$ vs. NC group; $^{*}P < 0.05$ and $^{**}P < 0.01$vs. OVA group. β-actin was used as an internal control.

closely related to the suppression of inflammatory cell influx, and therefore, linalool can be a useful inhibitor of MCP-1 in airway inflammatory diseases.

The excess production of mucus is a cause of airway obstruction and leads to death in patients with allergic asthma [40]. Thus, it is very important to suppress the overproduction of mucus in asthma. To our knowledge, the regulatory property of linalool on mucus hypersecretion has not been examined before unlike its various anti-inflammatory effects. Here, we showed the regulatory effect of linalool against mucus hypersecretion in OVA-exposed mice (Fig. 6). The inhibitory effects of linalool were similar to those of positive control, MON.

The increased iNOS expression results in the overproduction of NO, which leads to mucus overproduction and airway damage [12,41]. We have already confirmed the protective effect of linalool on mucus overproduction and therefore, focused on the regulatory role of linalool in the expression of iNOS. Interestingly, the OVA-induced increased levels of iNOS were significantly downregulated in the linalool group, suggesting that linalool could act as an inhibitor of iNOS.

The AKT signaling pathway plays an important role in promoting airway inflammation, and specific AKT inhibitors have shown therapeutic potential for protecting against airway inflammation in asthma [33,34,42]. In this study, we confirmed that AKT activation was downregulated by linalool (Fig. 7). This result implies that AKT signaling may be associated with the amelioration of airway inflammatory response, and linalool could be useful as an inhibitor of AKT activation.

The activation of MAPKs/NF-κB signaling pathways is well-known to be associated with allergic inflammation. The extracellular signal-regulated kinase (ERK) pathway was confirmed to activate transcription factors including NF-κB [43]. p38 is related to the regulation of cytokines including IL-5 [44]. JNK is known to be related to the accumulation of nitric oxide (NO) [43]. The recent report suggests that the downregulation of MAPKs activity exerts protective effects in an allergic inflammatory response [45,46]. Interestingly, the inhibitory effects of linalool on MAPKs (p-38, ERK and JNK) activation are well known in in vitro [47] and in vivo studies [48]. Therefore, we examined the inhibitory activity of linalool on MAPKs signaling. In this study, linalool inhibited JNK, p38 and ERK activation (Fig. 8). This result

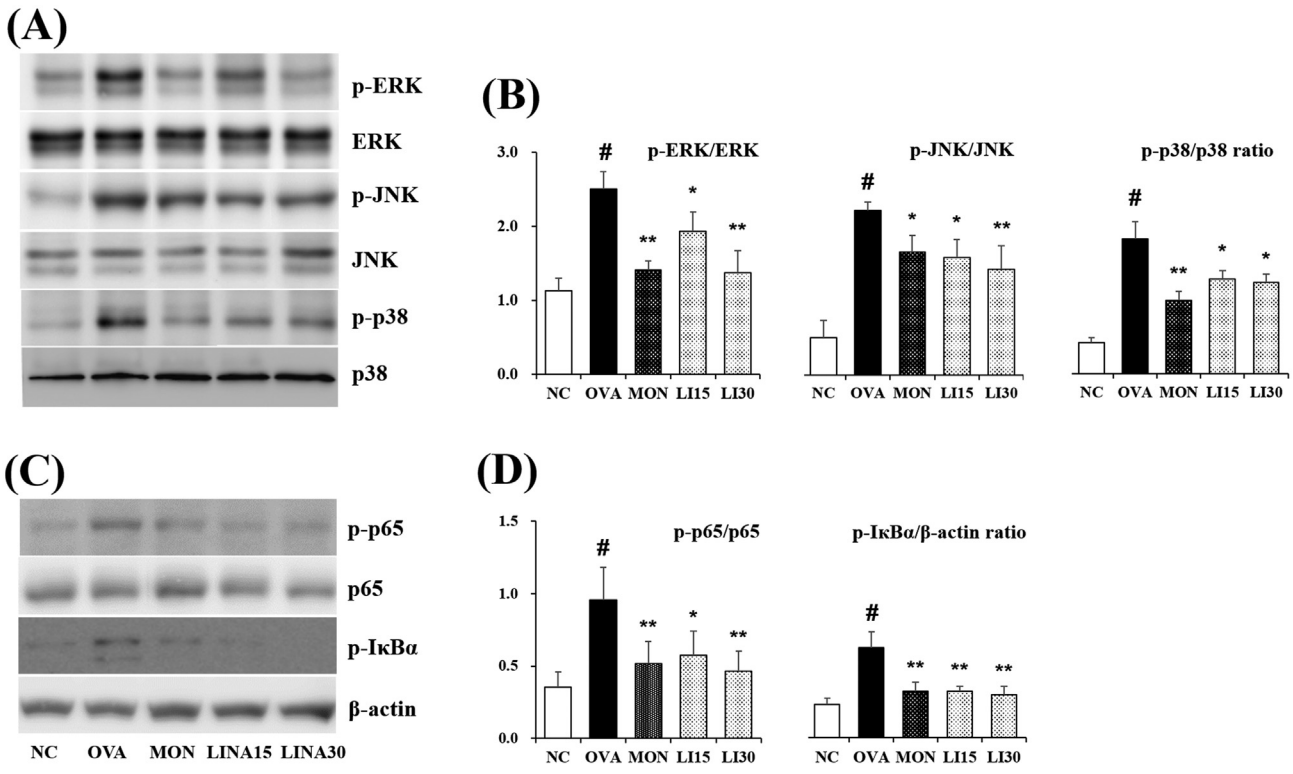

Fig. 8. Effect of linalool on the activation of MAPKs and NF-κB in the lungs (A and C). The levels of MAPKs and NF-κB activation were determined by western blot analysis and (B and D) quantitative analysis of MAPKs and NF-κB was performed by densitometric analysis. $^{\#}P < 0.05$ vs. NC group; $^{*}P < 0.05$ and $^{**}P < 0.01$ vs. OVA group. MAPKs, mitogen-activated protein kinases; NF-κB, nuclear factor kappa beta; JNK, c-Jun N-terminal kinase; ERK, extracellular signal-regulated kinase; IκB, inhibitor of NF-κB.

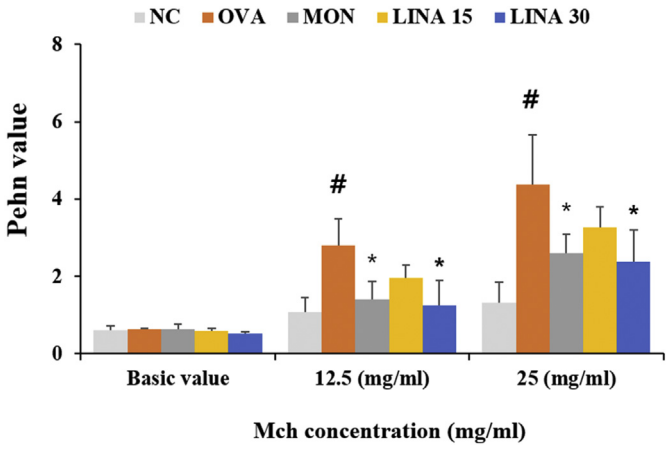

Fig. 9. Effect of linalool on the levels of AHR. The levels of Penh value represent the degree of AHR. $^{\#}P < 0.05$ vs. control group; $^{*}P < 0.05$ vs. OVA group. AHR, airway hyperresponsiveness; Penh, enhanced pause; MCH, methacholine.

suggests that linalool exerts a beneficial effect on pulmonary inflammation by suppressing MAPKs. The activation of NF-κB leads to Th2 cell differentiation and upregulation of inflammatory cytokines and chemokines in allergic asthma [40,46,49], and thus, the suppression of NF-κB activation considerably promotes the allergic inflammatory response [50]. In an in vivo study, linalool treatment effectively ameliorated airway inflammatory response by reducing NF-κB activation in cigarette smoke (CS)-exposed mice [23]. In an in vivo study, linalool treatment effectively ameliorated airway inflammatory response by reducing NF-κB activation in cigarette smoke (CS)-exposed mice. Linalool also exerted a protective effect in acute hepatic injury animal model by inhibiting NF-κB activation [51]. In LPS-stimulated BV2 Microglia, NF-κB activation was effectively downregulated with linalool treatment [24]. Similar to those previous studies, in this study, linalool downregulated NF-κB activation in the lung of OVA-exposed mice. Therefore, it can be suggested that linalool exerts anti-inflammatory action by regulating the MAPKs/NF-κB activation.

There is growing evidence indicating that essential oils have anti-inflammatory and antioxidant properties [52,53]. Linalool shows up in large amounts in many fruits and plants [54–58] and exerts protective effects in inflammatory diseases by the downregulation of inflammatory mediators production and MAPK/NF-κB activation shown in in vitro and in vivo studies [23,24,48,59]. In our study, linalool showed protective effects in airway inflammation and mucus hypersecretion. The levels of phosphorylated ERK, JNK, p38, Iκb and p65 were significantly downregulated in the linalool administration group (Figs. 7 and 8). These results indicate that the regulatory ability of linalool on MAPK and NF-κB activation has contributed to the amelioration of inflammatory response.

In conclusion, linalool ameliorated pulmonary inflammation and mucus overproduction in OVA-exposed mice. The anti-inflammatory effects of linalool were accompanied by a suppression of inflammatory cell recruitment and inflammatory molecules. Linalool inhibited key inflammatory molecular targets including AKT, MAPKs and NF-κB, which are closely related with the progression of pulmonary inflammation. In addition, linalool reduced AHR. It is therefore expected that linalool exerts protective effects in asthma symptoms.

Declaration of Competing Interest

All authors declare no conflict of interest.

Acknowledgments

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