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Exacerbation of Experimental Allergic Asthma by Augmented Th2 Responses in WSX-1-Deficient Mice1

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WSX-1 (IL-27R) is a class I cytokine receptor with homology to gp130 and IL-12 receptors and is typically expressed on CD4+ T lymphocytes. Although previous reports have clarified that IL-27/WSX-1 signaling plays critical roles in both Th1 differentiation and attenuation of cell activation and proinflammatory cytokine production during some bacterial or protozoan infections, little is known about the importance of WSX-1 in cytokine-mediated diseases of allergic origin. To this aim, we took advantage of WSX-1-deficient (WSX-1−/−) mice and induced experimental asthma, in which Th2 cytokines are central modulators of the pathology. OVA-challenged WSX-1−/− mice showed marked enhancement of airway responsiveness with goblet cell hyperplasia, pulmonary eosinophil infiltration, and increased serum IgE levels compared with wild-type mice. Production of Th2 cytokines, which are largely responsible for the pathogenesis of asthma, was augmented in the lung or in the culture supernatants of peripheral lymph node CD4+ T cells from WSX-1−/− mice compared with those from wild-type mice. Surprisingly, IFN-γ production was also enhanced in WSX-1−/− mice, albeit at a low concentration. The cytokine overproduction, thus, seems independent from the Th1-promoting property of WSX-1. These results demonstrated that IL-27/WSX-1 also plays an important role in the down-regulation of airway hyper-reactivity and lung inflammation during the development of allergic asthma through its suppressive effect on cytokine production. The Journal of Immunology, 2005, 175: 2401–2407.

Allergic asthma is a chronic airway disease characterized by local and systemic allergic inflammation and reversible airway obstruction. The hallmark features of allergic asthma are airway hyper-responsiveness (AHR),3 chronic pulmonary inflammation with eosinophilic infiltration, mucus hypersecretion, and elevated serum Ag-specific IgE (1). Th2-type cytokines play a critical role in the inflammation seen in asthma (2), although IFN-γ, a Th1-type cytokine, has also been implicated in the pathogenesis (3). Among Th2 cytokines, IL-4 participates in Th2 cell differentiation (4), IgE isotype class switching (5), and IL-6 promotes differentiation and migration of eosinophils (7–9). IL-13 is highly expressed in Th2 cells from asthma patients (10) and plays a role in the development of allergic asthma (11, 12) by acting on epithelial cells to produce AHR and mucus production (13). These results indicate that inappropriate development of Th2 responses plays a central role in the development of asthma. Although Th1 cells have been shown to prevent allergic disease by turning down the activity of Th2 cells (14–16), a contradictory report has shown that Ag-specific Th1 cells induced severe airway inflammation and acute lung pathology (3), raising controversy about the role of Th1 cytokines in AHR.

WSX-1 is a class I cytokine receptor with homology to gp130 of IL-6R and to IL-12R families (17). WSX-1 is highly expressed in spleen, thymus, and lymph nodes, particularly in the CD4+ T cell and NK cell compartments (17–19). IL-27, a heterodimeric cytokine consisting of p28 and EBV-induced gene 3 (EBI3), has been identified as a ligand for WSX-1, inducing the proliferation of and IFN-γ production by naive CD4+ T cells in synergy with IL-12 (20). Takeda et al. (21) demonstrated that IL-27/WSX-1 interaction induces T-bet and IL-12Rγ2 expression through STAT1 activation. Experiments using WSX-1-deficient (WSX-1−/−) mice revealed that WSX-1 plays an important role in Th1 differentiation during infection with some intracellular pathogens. Namely, WSX-1−/− mice showed increased susceptibility to infection with Listeria monocytogenes and Leishmania major due to impaired IFN-γ production (18, 19). In addition, Artis et al. (22) revealed that WSX-1 is required for suppression of the early IL-4 responses and the resultant Th1 promotion during L. major infection. These results clearly indicated that WSX-1 signaling acts before IL-12 to provide IL-12 responsiveness in naive CD4+ cells during the Th1 commitment of the cells. More recently, Villarino et al. (23) reported an interesting phenomena that WSX-1−/− mice infected with Toxoplasma gondii exhibited lethal inflammation mediated by hyperactivated CD4+ T cells. Infection with Trypanosoma cruzi also induced severe liver injury accompanied by overproduction of proinflammatory cytokines in WSX-1−/− mice (24). A similar anti-inflammatory property of WSX-1 has been observed in

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3 Abbreviations used in this paper: AHR, airway hyper-responsiveness; BAL, bronchoalveolar lavage; DC, dendritic cell; EB13, EBV-induced gene 3; LN, lymph node; PAS, periodic acid-Schiff; PC150, provocative concentration of acetylcholine aerosol; SOCS, suppressor of cytokine signaling; TPBS, Tween-PBS; WT, wild type.
mice infected with *Mycobacterium tuberculosis* (25). These findings have revealed novel functions of WSX-1 as a down-regulator of inflammatory cytokine production.

Although roles and mechanisms of WSX-1 in promotion or inhibition of immune responses against pathogenic infection has been clear, little is known about the importance of WSX-1 in diseases other than infection. In the current study we investigated the influence of WSX-1 deficiency in a murine model of asthma. We found that WSX-1−/− mice exhibited enhanced asthmatic phenotypes, including AHR, pulmonary inflammation with mucus overproduction and eosinophilic infiltration, and increased Ag-specific IgE in response to Ag challenge. Moreover, Th2 cytokine productions were enhanced in challenged WSX-1−/− mice compared with wild-type (WT) mice. These results indicated that WSX-1 plays an important role in the regulation of allergic inflammation by cytokine production.

### Materials and Methods

**Animals**

WSX-1−/− mice were generated as described previously by Yoshida et al. (19) and were backcrossed more than nine times to C57BL/6 mice (containing backcrossing). Mice were housed in microisolator cages and were used between 8 and 14 wk of age. Age- and sex-matched WT C57BL/6 mice (Seac Yoshitomi) were used as controls. All experiments were approved by the institutional animal research committee of Kyushu University and conformed to the animal care guidelines of the American Physiologic Society.

**Immunization and airway challenge with OVA**

WT or WSX-1−/− female mice were immunized i.p. twice with 20 μg of OVA (0.1 ml of 200 μg/ml in saline) adsorbed to aluminum potassium sulfate (Inject Alum; Pierce) at a 2-wk interval. On days 28, 29, and 30, the sensitized mice were challenged with a 1% (w/v) OVA/saline aerosol for 30 min in a chamber (24 × 17 × 17 cm). Thirty-six hours after the last aerosol challenge, measurements of airway responsiveness and other parameters were performed.

**Measurement of airway responsiveness**

Mice were anesthetized with a mixture of ketamine and sodium pentobarbital i.p., and their tracheas were cannulated via tracheotomy. The animals were ventilated mechanically (model 687; Harvard Apparatus), with a tidal volume of 0.01 ml/kg body weight (−13 ml) and a frequency of 120 breaths/min. Airway opening pressure was measured with a differential pressure transducer (model TP-6037; Nihon Kohden) and was recorded continuously with a pen recorder (Nihon Kohden RG-4124). Stepwise increases in the acetylcholine (diluted in 0.9% saline) dose (5–20 mg/ml/120 breaths) were given with an ultrasonic nebulizer (NE-U07; Omron Healthcare). The data were expressed as a percentage of the baseline airway opening pressure (BOP) at 0 breaths/min. Airway opening pressure was measured with a 1205 Betaplate (Wallac/PerkinElmer).

**Histological assessment and bronchoalveolar lavage**

Thirty-six hours after the last aerosol challenge, the lungs were removed and fixed with 4% paraformaldehyde-PBS and embedded in paraffin. The tissues were sectioned and stained with periodic acid–Schiff (PAS) according to a standard protocol. Cellular infiltration and goblet cells hyperplasia in the airway and lung tissues were evaluated by light microscopy. For bronchoalveolar lavage (BAL), mice were given a lethal dose of pentobarbital, and the lungs were gently lavaged with 1 ml of saline via the tracheal cannula. The lavage fluid was centrifuged at 800 rpm for 5 min at 4°C, and the supernatants were collected for determination of cytokines with ELISA kits (R&D Systems). The cell pellet was resuspended in saline, and cytoplasm preparations (Cytopsin 3; Shandon) were made. Total cell counts and differential cell counts on 200 cells were performed.

**Determination of Ag-specific Abs in serum**

The amounts of total or Ag-specific IgG1, IgG2a, and IgE in serum were measured by ELISA. For total Ig determination, ELISA plates were coated with anti-mouse IgG1 (AM21311; BioSource International), anti-mouse IgG2a (A90107A; Bethyl), or anti-mouse IgE (MCA419, Serotec) after 1/500, 1/1000, and 1/500 dilutions, respectively. Plates were washed three times with PBS containing 0.05% Tween 20 (TPBS), and serum samples were added to the wells after serial dilution in 3% BSA/TPBS. After 1-h incubation at room temperature, wells were washed with TPBS; incubated with biotinylated anti-mouse IgG1 (MCA336B; Serotec), anti-mouse IgG2a (61-0240; Zymed Laboratories), or anti-mouse IgE (MCA420B; Serotec) after 1/4000, 1/5000, and 1/2000 dilutions, respectively, for 1 h at room temperature; washed with TPBS; incubated with streptavidin-conjugated HRP (1/5000 dilution; 43-8332; Zymed) for 20 min. The reaction was developed with OPD (Wako Pure Chemical). For OVA-specific Ig determination, ELISA plates were coated with OVA (50 μg/ml) and blocked with blocking buffer (PBS containing 3% BSA). Detection of each Ig subclass was performed similarly.

**RT-PCR analysis**

Thirty-six hours after the last OVA challenge, total RNA was isolated from lung tissue or purified draining lymph node (LN) CD4+ T lymphocytes using TRIzol reagent (Invirotech Life Technologies) and reverse transcribed to cDNA with random primers. RT-PCR were performed using following appropriate pairs of primers (sense and antisense): IL-5, 5′-CTC TAG TAA GCC CCC TCA TT-3′ and 5′-TGA TAC CTG AAT AAC ATC CC-3′; IL-10, 5′-TAC CTG GTA GAA GTG ATC CC-3′ and 5′-CCT TCT GTA GCC TGG ATC GC-3′; IL-13, 5′-CTC CTT CCT ACG AAG GAG-3′ and 5′-GAA GGG TTC GGG ATG AAA CAG-3′; Muc5ac, 5′-CAG CCG AGA GGA GGG TTT GAT CT-3′ and 5′-AGT CTC TCT CCG CCT CTC TCA AT-3′; suppressor of cytokine signaling 1 (SOCS1), 5′-CAC TCA CCT CCG CAC CCT CC-3′ and 5′-CAG CCG GTG AGA TCT GGA AG-3′; SOCS3, 5′-GGT GAG CCG CAC GAC CCA GT-3′ and 5′-GGG TGG CAA AGA AAA GGA G-3′; WXS-1, 5′-GCC ATG AGG TAA GTA GTG GGT GGA GTG CTG-3′ and 5′-CTC CTT GGT AGA GGG TTG CCC AGA-3′; p28, 5′-CTG GTC CTA GCA GGT GCC GTC TCT TTG TG-3′ and 5′-CTC CAG CCG AGT AAG GAG ACG CTG-3′; and EBI3, 5′-CAG AGT GCA ATG CCA TGC TTC TC-3′ and 5′-CTG TGG CTC CAG CTG AC-3′. Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26). Primer sequences for other cytokines were described previously (26).

### Results

Enhanced airway responsiveness in WSX-1−/− mice

AHR is one of the main features of bronchial asthma (1). Therefore, we assessed the airway reactivity of WSX-1−/− mice to acetylcholine after immunization and aerosol challenge with OVA. Airway responsiveness to acetylcholine was slightly increased in OVA-challenged WT mice compared with untreated naive WT mice (Fig. 1A), showing that these mice were successfully sensitized to OVA by the immunization protocol used in this study. There were no significant differences in baseline airway responsiveness between naive WT and WSX-1−/− mice (Fig. 1A). In OVA-challenged WSX-1−/− mice, airway responsiveness to acetylcholine was significantly enhanced over the OVA-challenged...
WT mice (Fig. 1A). Accordingly, the PC\textsubscript{150} was significantly lower in challenged WSX-1\textsuperscript{-/-} mice than in challenged WT mice. These results clearly demonstrated that the WSX-1 deficiency enhanced AHR after Ag sensitization. In Fig. 1B, the expressions of WSX-1, p28 of IL-27, and EB1-3 of IL-27 in the lung during the time course of asthma development are shown. Although the expression of WSX-1 and p28 appeared constant, expression levels of EB1-3 were up-regulated after OVA challenge, indicating possible involvement of IL-27/WSX-1 in asthma development.

WSX-1 deficiency gave rise to exacerbated lung inflammation with augmented infiltration of eosinophils

Lung-localized inflammation is also a feature of bronchial asthma (1). As shown in Fig. 2A, PAS staining of lung tissues revealed that histological asthmatic features were also exacerbated in the lungs of OVA-challenged WSX-1\textsuperscript{-/-} mice. In naive WT or WSX-1\textsuperscript{-/-} mice, there were no apparent pathological changes in the lung (Fig. 2, A-i and A-iii). However, after OVA challenge, cellular infiltration and goblet cell hyperplasia at bronchiole were apparent in WSX-1\textsuperscript{-/-} mice, whereas only mild pathological changes were observed in WT mice (compare Fig. 2, A-ii and A-iv). As shown in Fig. 2A-viii, massive PAS-positive regions were detected under the epithelium and in the lumen of bronchioles of OVA-challenged WSX-1\textsuperscript{-/-} mice, whereas minimum PAS-positive regions were observed in the lungs of WT mice (compare Fig. 2, A-vi and A-viii). Furthermore, enhanced expression of MucSac mRNA, another marker of goblet cell hyperplasia (27), was detected especially in the lungs of OVA-challenged WSX-1\textsuperscript{-/-} mice (Fig. 2B). Additionally, cell counts from BAL fluid revealed that infiltration of inflammatory cells, such as lymphocytes and eosinophils, into the bronchoalveolar space was significantly augmented in OVA-challenged WSX-1\textsuperscript{-/-} mice compared with that in WT mice (Fig. 2C). There was no significant difference in the number of macrophages and neutrophils between OVA-challenged wild-type and WSX-1\textsuperscript{-/-} mice (Fig. 2C). Therefore, WSX-1 deficiency resulted in severe lung inflammatory changes with eosinophilic infiltration into bronchioles.

WSX-1 deficiency augmented Ag-specific IgE production

The concentration of serum IgE Abs is closely associated with the severity of asthma, especially in children (28). The production of subclasses of IgEs is regulated by cytokines derived mainly from Th cells. The Th2 cytokine IL-4 induces IgG1 and IgE production, whereas the Th1 cytokine IFN-\gamma induces IgG2a in mice (29, 30). First, we measured total Ig levels in sera from OVA-sensitized (but not challenged) mice as shown in Fig. 3A. In sensitized WSX-1\textsuperscript{-/-} mice, total IgE levels were higher than those in sensitized WT mice, and inversely, total IgG2a levels were lower (Fig. 3A). Similar serum Ig profiles were observed in naive WSX-1\textsuperscript{-/-} mice before sensitization (Fig. 3B, upper panels). Hence, WSX-1\textsuperscript{-/-} mice were likely to be somewhat Th2 prone even in the baseline state before OVA airway challenge. Next, we assessed serum Ig levels from OVA-sensitized mice after challenge. As shown in Fig. 3B (upper panels), total IgG1 and IgE levels were significantly higher in OVA-challenged WSX-1\textsuperscript{-/-} mice than in challenged WT mice. In response to OVA challenge, all OVA-specific IgG1, IgG2a, and IgE in sera increased in both WT and WSX-1\textsuperscript{-/-} mice, as shown in Fig. 3B (lower panels). Especially, OVA-specific IgE levels in
sera of OVA-sensitized (but not challenge) WT (circles) and OVA-sensitized (but not challenge) WSX-1−/− mice were measured by ELISA as described in Materials and Methods. B, Sera were collected from OVA-sensitized WT (●) and OVA-sensitized WSX-1−/− (▲) mice 36 h after the last OVA inhalation. Similarly, sera were collected from naive WT (○) and WSX-1−/− (+) mice. Total (upper panels) and OVA-specific (lower panels) Ig amounts were determined by ELISA. Data are the mean ± SEM (n = 5–6 mice/group). *, p < 0.05; **, p < 0.01 (compared with OVA-challenged WT samples). Experiments were repeated three times with similar results.

FIGURE 3. Augmented Ag-specific IgE production in OVA-challenged WSX-1−/− mice. A, Total IgG1, IgG2a, and IgE levels in sera from OVA-sensitized WT and naive WSX-1−/− mice were measured by ELISA as described in Materials and Methods. B, Sera were collected from OVA-challenged WT (●) and OVA-challenged WSX-1−/− (▲) mice 36 h after the last OVA inhalation. Similarly, sera were taken from naive WT (○) and naive WSX-1−/− (+) mice. The levels of IL-4, IL-13, and IFN-γ were determined by ELISA. Data are the mean ± SEM (n = 6–9 mice/group). *, p < 0.05; **, p < 0.01. Experiments were repeated three times with similar results.

FIGURE 4. Augmented accumulation of Th2 cytokines in the bronchiole of OVA-challenged WSX-1−/− mice. Thirty-six hours after the last OVA inhalation, BAL fluids were collected from OVA-challenged WT (●) and OVA-challenged WSX-1−/− (▲) mice. Similarly, BAL fluids were taken from naive WT (○) and naive WSX-1−/− (+) mice. The levels of IL-4, IL-13, and IFN-γ were determined by ELISA. Data are the mean ± SEM (n = 6–9 mice/group). *, p < 0.05; **, p < 0.01. Experiments were repeated three times with similar results.

higher amounts of IL-4 and IL-13 were produced in both mouse groups. The levels of IL-4 and IL-13 in BAL samples from OVA-challenged WSX-1−/− mice were significantly higher than those in samples from challenged WT mice. In contrast, the level of IFN-γ was barely detectable in both WSX-1−/− and WT mice before OVA challenge and were not increased even after challenge (Fig. 4). These data clearly demonstrated augmented Th2 cytokine production in the lungs of WSX-1−/− mice, especially IL-13, which is likely to participate in severe asthmatic phenotypes in the mice.

Augmented cytokine production by peribronchial LN CD4+ T lymphocytes from WSX-1−/− mice

WSX-1 is highly expressed on CD4+ T lymphocytes (17–19), which produce various immunoregulatory cytokines. Therefore, we investigated cytokine expression in peribronchial LN CD4+ T lymphocytes purified from OVA-challenged mice by RT-PCR (Fig. 5A). As expected, the expressions of IL-4, IL-5, and IL-13 were enhanced after OVA challenge in both WT and WSX-1−/− mice. In challenged WSX-1−/− mice, however, these cytokines were overexpressed compared with those in challenged WT mice. Interestingly, the expression of IFN-γ was also augmented in WSX-1−/− mice after the OVA challenge, whereas it was down-regulated in WT mice after challenge. The expressions of IL-10 and TGF-β, two well-known immunosuppressive cytokines (31, 32), were not significantly altered.

For additional confirmation of cytokine overproduction by WSX-1-deficient CD4+ T cells, we assessed the Ag-induced proliferation and cytokine production of CD4+ T cells purified from peribronchial LN of OVA-challenged mice. As shown in Fig. 5B, cellular proliferation was augmented by OVA adjunction in a dose-dependent manner, and the intensity of the response to Ag was significantly higher in WSX-1−/− T cells compared with WT T cells. At the same time, WSX-1−/− T cells from OVA-challenged mice produced more IL-4 and IL-13 than the WT T cells in response to OVA in vitro. Although not significant, the production of IL-5 in response to OVA also tended to be higher in WSX-1−/− T cells compared with WT T cells. As expected from the RT-PCR results (Fig. 5A), OVA-induced IFN-γ production by WSX-1−/− T cells was significantly higher than that by wild-type T cells. However, the amounts of IFN-γ produced were much lower than the levels of Th2 cytokines produced. Taken together with the results in Fig. 4, it was demonstrated that WSX-1 deficiency resulted in augmented production of Th2 cytokines, including IL-4 and IL-13, and also production of IFN-γ. Given the very low amounts of IFN-γ produced by WSX-1−/− CD4+ T cells or accumulated in

WSX-1 deficiency augmented accumulation of IL-13 in the bronchiole

Next we determined whether the enhanced AHR and lung inflammation in WSX-1−/− mice reflected altered cytokine profiles in OVA-challenged mice. There were barely detectable levels of IL-4, IL-13, and IFN-γ in BAL samples from both naive WT and naive WSX-1−/− mice (Fig. 4). By OVA challenge, significantly
FIGURE 5. Augmented cytokine production by WSX-1-deficient peribronchial LN CD4+ T lymphocytes. A. Expression of cytokine mRNA evaluated by RT-PCR. Thirty-six hours after the last OVA inhalation, total RNAs were isolated from peribronchial LN CD4+ T lymphocytes of WT or WSX-1−/− mice. RNAs were similarly prepared from naive WT and WSX-1−/− mice. RT-PCR was performed as described in Materials and Methods. cDNAs for β-actin were amplified as a control. Shown are representative data from three independent experiments. B. Ag-specific proliferation and cytokine production of peribronchial LN CD4+ T cells. CD4+ T cells were prepared from OVA-challenged WT and OVA-challenged WSX-1−/− mice. Four days after culture in the presence of 0–500 µg/ml OVA, proliferation and cytokine levels in the supernatants were determined by thymidine incorporation or ELISA, respectively. Data are the mean ± SEM (n = 4–5 mice/group). Experiments were repeated three times with similar results. *, p < 0.05; **, p < 0.01; ***, p < 0.005.

BAL after challenge, however, the overproduction of mainly Th2 cytokines caused by WSX-1 deficiency should predominantly affect the exacerbation of AHR and asthmatic pathology in the knockout mice.

Enhanced IFN-γ production in WSX-1−/− CD4+ T cells was independent of T-bet expression

Among transcription factors, GATA-3 and T-bet function as master regulators for Th2 and Th1 development, respectively (33, 34). The expression of GATA-3 is reportedly increased in patients with atopic asthma (35). Inconsistent with this, the expression of GATA-3 in peribronchial LN CD4+ T cells from OVA-challenged mice was augmented over that in naive mice of both groups (Fig. 6). The expression levels of GATA-3, however, were comparable between WT and WSX-1−/− mice. In contrast, the expression of T-bet, which contributes to the induction of IFN-γ production, was greatly reduced in WSX-1−/− mice after OVA challenge, which was significantly lower than that in OVA-challenged WT mice (Fig. 6). This was apparently contradictory to the overproduction of IFN-γ by WSX-1−/− T cells (Fig. 5). Therefore, in the current experimental circumstance, WSX-1 is likely to participate in the down-regulation of IFN-γ production by CD4+ T cells through a T-bet-independent pathway.

SOCS did not mediate WSX-1 cytokine suppression

Most cytokine signaling pathways leading to cellular activation, including gene expression, are negatively regulated by suitable SOCS (36). Although SOCS are mainly involved in the down-regulation of immune responses (36), some SOCS molecules are implicated in Th1/Th2 responses (37, 38). In this context, we assessed the expression of SOCS1 and SOCS3 in peribronchial LN CD4+ T lymphocytes isolated 36 h after the last OVA challenge. Although SOCS1 was induced after OVA challenge in both WT and WSX-1−/− mice, there was no significant difference in the expression levels between WSX-1−/− and WT T cells (Fig. 7). Although SOCS3 was also induced by OVA challenge, the expression levels were again comparable between the two groups (Fig. 7). Therefore, SOCS1 and SOCS3 were not implicated in the cytokine overproduction by WSX-1-deficient T cells in the current experimental system.

Discussion

AHR and lung-localized inflammatory responses participate in the development of asthmatic symptoms (1). Various studies have
shown that cytokines, such as IL-4, IL-5, and IL-13, play crucial roles in the initiation of Ag-induced airway responses and lung inflammation (2). In this study we demonstrated that WSX-1 deficiency gave rise to augmentation of allergen-induced AHR and lung inflammation as follows. WSX-1−/− mice did develop enhanced AHR induced by intranasal challenge with OVA compared with WT mice (Fig. 1). Ag-induced lung inflammation involving goblet cell hyperplasia and eosinophilic infiltration was significantly augmented in OVA-challenged WSX-1−/− mice (Fig. 2). Furthermore, an increased level of Ag-specific serum IgE, another typical feature in atopic bronchial asthma, was observed in WSX-1−/− mice (Fig. 3). These results provide the first evidence that WSX-1 plays an inhibitory role in allergic asthma development.

Given the Th1-promoting property of IL-27/WSX-1 (18–21), we initially hypothesized that asthma phenotypes in WSX-1−/− mice should be worse than those in WT mice simply due to the lack of Th1-mediated suppression of Th2 cytokine production. Although WSX-1−/− mice challenged with OVA indeed produced more Th2 cytokines than WT mice, the enhanced production of these Th2 cytokines in WSX-1−/− mice is not likely due to the lack of WSX-1-mediated suppression of Th2 cytokine production. However, augme...