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Wheezing and itching
The requirement for STAT proteins in allergic inflammation

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Abbreviations: AAD, allergic airway disease; AD, atopic dermatitis; ASM, airway smooth muscle; CCL, C-C chemokine ligand; IL, interleukin; LPS, lipopolysaccharide; OVA, ovalbumin; SNP, single nucleotide polymorphism; Th, CD4+ T helper cell subset; WT, wild-type

The development of allergic inflammation requires the orchestration of gene expression from the inflamed tissue and from the infiltrating immune cells. Since many of the cytokines that promote allergic inflammation signal through hematopoietin family receptors, the Signal Transducer and Activator of Transcription (STAT) family have obligate roles in pro-allergic cytokine-induced gene regulation in multiple cell types. In this review, we summarize work defining the contribution of each of the STAT family members to the development of allergic inflammation, using data from mouse models of allergic inflammation, studies on patient samples and correlations with single nucleotide polymorphisms in STAT genes.

Inappropriate immune responses to otherwise innocuous antigens result in the development of inflammatory disease.[1,2] Hypersensitivity responses to allergens lead to a variety of allergic diseases, including asthma, rhinitis, atopic dermatitis, intestinal anaphylaxis and eosinophilic esophagitis. The incidence of allergic disease is growing in Western society, creating considerable health care costs. Following initial allergen exposure, epithelial cells at the environmental interface such as keratinocytes and airway epithelial cells, secrete cytokines that recruit innate and adaptive immune cells to the site of exposure, initiating the induction of an allergen-specific immune response. Dendritic cells prime the development of effector T cells that produce cytokines including IL-4, IL-5, IL-9, IL-13 and IL-17 to promote inflammation.[3,4] Although Th2 cells are thought to be the major cells involved in allergic inflammation, roles for Th1, Th9 and Th17 cells have been demonstrated. Cytokines produced by T helper cells stimulate allergen-specific antibody production, particularly IgE that sensitizes mast cells for degranulation upon subsequent exposure to allergen. Allergen challenge results in the recruitment of a variety of cells to the site including T cells, B cells, dendritic cells, eosinophils and neutrophils. Inflammation may be transient, but chronic exposure to allergens results in disease such as asthma characterized by pulmonary inflammation, mucus production, tissue remodeling and airway hyperreactivity (AHR). At various stages in the development of allergic inflammation, the generation of T helper subsets and the responses of resident cells in the inflamed target organs rely upon cytokine stimulation and the subsequent activation of Signal Transducer and Activator of Transcription (STAT) proteins.

In this review we focus on the role of STAT proteins in the development of allergic inflammation. We review data both from biological samples from patients and single nucleotide polymorphisms supporting a role for STATs in human disease (summarized in **Table 1**). We also summarize the evidence from a number of models of asthma that have been developed in mice, collectively referred to here as allergic airway disease (AAD), and models of other allergic diseases that demonstrate a requirement for STAT family members in the pathogenesis of allergic inflammation.

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STAT1 and STAT2

STAT1 is primarily activated by type I and type II interferons, although stimulation with a number of other cytokines can result in STAT1 activation. In contrast, STAT2 activation is restricted to type I interferons. Interferons are often considered antagonists of Th2 development and allergic inflammation, though there is evidence that active STAT1 may also contribute to inflammation. Levels of activated STAT1, but not other transcription factors such as STAT3, NFκB or AP-1, are increased in airway epithelial cells of asthmatic patients compared with non-asthmatic subjects or patients with chronic bronchitis.[5] Similarly, levels of phospho-STAT1, but not phospho-STAT3 or STAT5, are increased in peripheral CD4+CD161+ T cells isolated from asthmatic patients compared with healthy subjects.[6] Consistent with an anti-allergic function of STAT1, the presence of the *STAT1* SNP rs3771300 is inversely related to total serum IgE levels in a population of German children and may be protective for atopic sensitization,[7] and there is a significant association between *STAT2* SNP rs2066807 and asthma in Chinese and Taiwanese children.[8]

IFN-γ has been shown to suppress murine AAD in a STAT1-dependent manner. Upon continuous exposure to allergen, IFN-γ

Table 1. Correlations of allergic disease with SNPs in STAT genes

Gene	Polymorphism	Disease/Phenotype	Population	References
STAT1	rs3771300	Protective for atopic sensitization	German	7
STAT2	rs2066807	Associated with asthma	Chinese and Taiwanese	8
STAT3	rs2306581, rs957971, rs1026916	Associated with decreased lung function	Caucasian	23
STAT4	T90089C	Associated with risk of mite allergen IgE production in asthmatics	Korean	37
STAT6	rs324015	Inconsistent findings with asthma correlation	Japanese, Swedish, Finnish	38, 39, 65, 67–69
STAT6	rs324015	No association with bronchial asthma, atopic dermatitis, food-related anaphylaxis, serum IgE	Japanese	65, 67
STAT6	rs324015	Associated with susceptibility to nut allergy	British Caucasian	66
STAT6	rs324011, rs4559	Associated with increased IgE levels	German and Swedish	69, 73, 74
STAT6	rs324011	Association with recurrent wheezing but not asthma susceptibility	Slovene	72
STAT6	rs1059513	Associated with increased IgE levels and bronchial responsiveness to methacholine	German and Swedish	69, 74
STAT6	rs3024974	Associated with improved lung function	Chinese	76
STAT6	rs3024975, rs841718, rs167769, rs703817	Associated with AD patients with viral skin infections (eczema herpeticum)	Caucasian	77
STAT6	GT repeat in exon 1 (A1 allele, GT_{13} repeat)	Increased frequency in patients with bronchial asthma, atopic dermatitis, and/or food-related anaphylaxis	Japanese	67
STAT6	GT repeat A1-A5 (GT_{13}-GT_{17})	No correlation with asthma, serum IgE or bronchial responsiveness	German and Swedish	69
STAT6	GT repeat A4 (GT_{16})	Associated with increased numbers of peripheral eosinophils	German and Swedish	69
STAT6	GT repeat A1 allele (GT_{13})	Associated with atopic asthma and elevated serum IgE	American and British Caucasians	71
STAT6	GT repeat A3 allele (GT_{15})	Associated with asthma but not elevated serum IgE	North Indian	70

induces apoptosis of airway epithelial cells, leading to the eventual resolution of Ag-induced goblet cell hyperplasia.[9,10] IFN-γ induces STAT1-dependent induction of the chemokines CXCL9 and CXCL10, which can negatively regulate Ag-induced eosinophil recruitment into the airways of allergic mice.[11,12] Studies using mouse embryonic fibroblasts have shown that IFN-γ/STAT1 signaling can impair IL-4-induced STAT6 phosphorylation and production of eotaxin, an eosinophil chemoattractant, through the induction of SOCS-1.[13]

In contrast, STAT1 can also promote allergen-induced AAD in mice when Th1 cells contribute to the inflammation. Administration of STAT1-specific decoy oligonucleotides to the airways of allergic mice results in reduced lung expression of the co-stimulatory molecule, CD40 and adhesion molecule, VCAM-1, which correlates with reduced pulmonary lymphocytic and eosinophilic infiltration along with reduced AHR.[14] STAT1-deficient mice display limited recruitment of adoptively transferred Ag-specific Th1 cells to their lungs and airways after local allergen challenge. STAT1 induces the expression of the CXCR3 ligands CXCL9, CXCL10, and CXCL11 in the lung whose expression is important for the Ag-induced recruitment of CXCR3-expressing Th1 cells.[15] Upon exposure to lipopolysaccharide (LPS) and ovalbumin (OVA), mice that have received adoptively transferred Ag-specific Th1 cells display increased IFN-γ- and STAT1-dependent expression of KC and MIP-2, two CXCR2 ligands, which correlates with enhanced pulmonary recruitment of CXCR2-expressing neutrophils.[16] Taken together, STAT1 may inhibit or promote the development of AAD depending on the cells involved in a particular state of inflammation.

STAT3

STAT3 is activated by a large number of cytokines present in the pro-allergic milieu. Moreover, it is expressed in multiple cell types including epithelial cells, airway smooth muscle cells and immune cells. Thus, it is a critical component in multiple aspects of allergic disease.

Although not associated with classical atopic disease, heterozygous mutations in the DNA binding, SH2, linker and transactivation domains of STAT3 have been identified as the primary molecular cause of autosomal-dominant Hyper-IgE syndrome (HIES), and this has been extensively reviewed elsewhere.[17-20] An examination of B cells isolated from control and HIES patients demonstrated that STAT3 is required for IL-21-stimulated IgE production and thus, IgE production in response to IL-21 was decreased in B cells from HIES patients.[21] However, a later report found that the STAT3 mutations identified in HIES were not responsible for elevated serum IgE levels in asthmatic patients.[22] It is not clear whether SNPs in the STAT3 gene are associated with allergic phenotypes in patients. Three STAT3 polymorphisms (rs2306581, rs957971, rs1026916) are strongly associated with decreased lung function in asthmatic adults and children.[23] In contrast, an analysis of 25 *STAT3* SNPs demonstrated no association of any of the STAT3 polymorphisms with asthma, lung

function, high levels of total or specific serum IgE, or elevated eosinophil counts.[22]

In human airway smooth muscle (ASM) cells, STAT3 is required for the expression of eotaxin-1/CCL11 following IL-9,[24] IL-17A[25] and Oncostatin M[26] stimulation, VEGF expression following Oncostatin M stimulation[27] and IL-6 and IL-8/CXCL8 induction following TSLP stimulation.[28] PDGF-stimulated proliferation of ASM cells requires STAT3 to regulate cyclin D3 and p27 expression.[29]

STAT3 expression in epithelial cells and CD4+ T cells is essential for the development of allergic inflammation in mice. In a chronic model of murine AAD, continuous local exposure to allergen induces STAT3 activation in the epithelium, smooth muscle, and surrounding cells of the airway in wild type (WT) mice. STAT3 expression in lung epithelial cells is necessary for the induction of the chemokines TARC and KC, and mice that lack STAT3 expression in their lung epithelial cells display impaired recruitment of eosinophils and Th2 cells into their lungs, which correlates with reduced pulmonary inflammation and AHR.[30] In addition, it has recently been shown that STAT3 cooperates with STAT6 to promote the development of Th2 cells and Th2-mediated allergic inflammation. In an OVA-induced model of AAD, mice with a T cell specific deletion of STAT3 ($Stat3^{Cd4-/-}$) display decreased pulmonary inflammation and eosinophilia along with reduced levels of Th2 cytokines and IL-17A in their airways.[31] Since STAT3 is important for the development of both Th2 and Th17 cells, these observations cannot be solely attributed to Th2 cells, and the role of STAT3 in Th2-mediated allergic inflammation was further assessed through crossing $Stat3^{Cd4-/-}$ mice to mice that express a constitutively active STAT6 (STAT6VT) in T cells. STAT3-sufficient STAT6VT mice are characterized by IL-4-dependent spontaneous pulmonary and skin inflammation and blepharitis.[32,33] STAT6VT mice with STAT3-deficient T cells do not develop spontaneous pulmonary and skin inflammation nor do they develop blepharitis, demonstrating that STAT3 expression in T cells is important for the development of Th2-mediated allergic inflammation.[31] Thus, STAT3 expression in multiple cell types is critical for the development of allergic inflammation.

STAT4

Among the STAT proteins, STAT4 is the only one that shows a tissue-restricted pattern of expression, primarily in lymphoid and myeloid cells. STAT4 is required for all known IL-12 biological responses, including IFN-γ production and the differentiation of Th1 cells. STAT4 also mediates some responses to type I IFNs and to IL-23.[34-36]

Considerably more work has been done to define a role for STAT4 in mouse models than has been reported for disease in patients. In humans, a *STAT4* SNP (T90089C) is positively associated with risk of production of mite allergen-specific IgE in asthmatic Korean patients, although no association to asthma is found in Korean or Finnish populations.[37,38] A later study on the same *STAT4* polymorphism identified a significant positive association with asthma in a Chinese population.[39]

As was noted for STAT1, STAT4 can have both positive and negative roles in the development of allergic inflammation. The obligate requirement for STAT4 in IFN-γ-producing Th1 cells, which can inhibit Th2-mediated inflammation, provides a mechanism for negative regulation. IFN-γ-induced negative regulation of AAD is dependent on IL-12 and STAT4 signals, as STAT4-deficient mice, or WT mice exposed to IL-12 neutralizing antibodies, display attenuated pulmonary inflammation, cytokine responses and AHR.[40] In addition, the combination of IL-2 and IL-18, which increases IL-12 and IFN-γ in the airways, can reduce pulmonary inflammation and AHR in OVA-sensitized mice and these effects are dependent on local IL-12 and STAT4-induced IFN-γ production from NK cells.[41]

Based on its role in Th1 cells, the demonstration that STAT4 is required for allergic inflammation is somewhat counter-intuitive. Several mechanisms have been proposed to explain this phenomenon. In a cockroach allergen model, STAT4-deficient mice, and WT mice treated with anti-IL-12, display reduced inflammation that correlates with decreased pulmonary chemokine production. These defects cannot be rescued by adoptive transfer of WT sensitized splenocytes, suggesting that there may be dominant repressive factors present in the STAT4-deficient environment.[42] In an OVA-induced model, two possible, and not mutually exclusive, mechanisms have been defined for decreased allergic inflammation in the absence of STAT4. The first is based on the observation that IL-17 is required for allergic inflammation in this model, and that IL-17 production from lymphocytes isolated from sensitized and challenged STAT4-deficient mice is decreased compared with control mice. It is possible that IL-17 plays an important role in the development of STAT4-dependent AAD, perhaps by recruiting neutrophils to the airways of challenged mice.[43] Parallel studies also implicate IL-12 and STAT4 in limiting the development of inducible Tregs during the development of allergic inflammation.[44] Mechanistically, STAT4 may mediate this effect by inducing repressive chromatin modifications at the *Foxp3* locus and impairing STAT5 binding to *Foxp3*.

STAT4 also contributes to AAD when LPS is used as an adjuvant. Mice exposed to high or low doses of LPS during airway allergen sensitization develop allergic inflammation associated with Th1 or Th2 responses, respectively, after local Ag-challenge.[45] When administered during the induction of AAD, high-dose LPS results in an increase in serum IgE and IgG2a, pulmonary IL-12 production, and enhanced AHR that is associated with pulmonary infiltration of macrophages, lymphocytes and neutrophils but not eosinophils. Importantly, the responses induced by high-dose LPS are dependent on STAT4.[46] Thus, the pro-inflammatory role of STAT4 in many autoimmune diseases extends to the development of allergic inflammation in the lung.

STAT5

STAT5 is activated by a number of γc receptor cytokines on various cells that contribute to the development of AAD, including IL-2 and IL-9. STAT5 is activated by thymic stromal lymphopoietin, a cytokine critical for the development of allergic

inflammation, in T cells and dendritic cells.[47-49] STAT5 plays an important role in mast cell and eosinophil responses, both of which are important for the development of inflammatory diseases, such as AAD. Studies using STAT5-deficient mice have shown that STAT5 is critical for mast cell proliferation, survival, and function.[50,51] STAT5 is rapidly activated in response to IgE and Ag-mediated FcεRI cross-linkage in WT bone marrow mast cells and STAT5-deficient mast cells display impaired IgE-mediated degranulation and decreased leukotriene and cytokine secretion.[51] IL-5-induced STAT5A and STAT5B signaling is important for murine eosinophilopoiesis and eosinophil chemotaxis and can also induce the development of IL-4-producing eosinophils.[52,53] Upon induction of AAD, STAT5A-deficient and STAT5B-deficient mice display reduced pulmonary lymphocytic and eosinophilic infiltration as well as lower levels of IL-5 in their airways in comparison to WT allergic mice. In addition, splenocytes from STAT5A-deficient and STAT5B-deficient allergic mice proliferate less than those from WT mice when stimulated with relevant antigen, suggesting that STAT5 may be important for the priming of Ag-specific T cells during allergic inflammation.[52] Furthermore, while STAT6 is critical for the development of Th2 cells and allergic inflammation, STAT6-independent Th2 differentiation can occur in a STAT5A-dependent manner. Allergic mice deficient in STAT5A or STAT6 display reduced pulmonary lymphocytic and eosinophilic infiltration in comparison to WT mice, whereas in the absence of both STAT5A and STAT6, pulmonary inflammation is nearly abolished.[54] Together, these data show that STAT5-induced signals are important for the normal development and function of mast cells and eosinophils and allergen-induced responses in murine AAD.

STAT6

STAT6 has been the focus of extensive research in allergic inflammation and atopic disease. It is activated by IL-4 and IL-13, cytokines that are critical for the differentiation of Th2 cells, and responses to Th2 cytokines by resident tissue cells.[55-58] Considerable evidence supports a central role for STAT6 in human and murine allergic inflammation.

Patient samples. An examination of patients with allergic rhinitis revealed that STAT6 expression (nuclear and cytoplasmic) in the nasal mucosa increases following allergen challenge, but steroid treatment prevented the allergen-induced increase in STAT6.[59] Similarly, baseline levels of phospho-STAT6 in CD4+CD161+ peripheral T cells are higher in atopic asthmatics, but treatment with oral corticosteroids decreased levels of phospho-STAT6. Interestingly, the corticosteroids were specifically targeting STAT6 since baseline levels of phospho-STAT1 (which are also increased in asthmatics) were not decreased following steroid treatment.[6] Increased numbers of STAT6+ cells are present in bronchial epithelial cells from atopic asthmatic patients compared with non-atopic asthmatics and control patients,[60] and similarly, the number of cells expressing STAT6 as well as c-Maf and GATA-3, Th2-associated transcription factors, in the sputum of asthmatic patients is significantly higher than that of healthy subjects.[61] However, neither protein levels nor DNA-binding activity of STAT6 in peripheral blood monocytes is different between patients with asthma or elevated serum IgE and nonatopic subjects.[62] An examination of bronchial epithelium from patients with mild asthma, severe asthma or non-asthmatic controls demonstrated that STAT6 expression is significantly higher in bronchial epithelial cells from patients with severe asthma, but not in patients with mild asthma, compared with non-asthmatic subjects,[63] and following allergen challenge, an increased number of phospho-STAT6+ epithelial cells and fibroblasts are observed in the bronchial tissue of atopic asthmatics as compared with control subjects.[64]

SNPs. Polymorphisms of the *STAT6* gene are associated with various aspects of asthma and atopy (**Table 1**). In an examination of British and Japanese populations, a single nucleotide polymorphism (*STAT6* G2964A; rs324015) is associated with mild atopic asthma, but not atopy or elevated serum IgE levels, in a Japanese population, but not in a British population.[65] While this SNP is associated with the susceptibility and severity of nut allergy in an atopic British population,[66] other studies found no association of the polymorphism with asthma in Japanese,[67,68] German, Swedish,[69] and Chinese[39] populations nor with serum IgE levels in a Finnish population.[38] A dinucleotide polymorphism (GT 13–17 repeats) in exon 1 of *STAT6* has also been studied for its potential link to atopic disorders.[67] The A1 allele (GT 13 repeat) is found more frequently in Japanese children with allergic diseases (bronchial asthma, atopic dermatitis, and/or food-related anaphylaxis) than controls and there is a strong association between allergic disease and the A1/A3 (GT 15 repeat) heterozygote. However, no variants of the dinucleotide polymorphism are associated with elevated serum levels of IgE in the allergic patients. Further examination of the GT polymorphism alleles A1–A5 (13–17 repeats, respectively) demonstrated no association with asthma, total serum IgE levels or bronchial responsiveness to methacholine challenge. However, GT repeat A4 has a significant association with increased numbers of peripheral eosinophils.[69] Conversely, an examination of an Indian population found an association of GT repeat A3 (GT_{15}) with asthma but not elevated serum IgE.[70] A later study of the GT polymorphisms identified two novel repeats GT_{12} and GT_{18} in American and British populations, respectively, although the previously identified A1 (13 repeat) and A3 (15 repeat) alleles were more common.[71] Importantly, Gao et al. demonstrated a significant association of the A1 allele with atopic asthma and elevated serum IgE levels in American and British Caucasians and also revealed the ability of the GT repeat alleles to differentially regulate *STAT6* promoter activity.[71]

Examination of additional *STAT6* SNPs in a German and Swedish sibling-pair study revealed no association with asthma or blood eosinophil count, but four SNPs (6613C/T; rs324011, 1309A/G, 1507C/T, and 4671A/G; rs4559) are associated with increased levels of serum IgE and an additional SNP (4610A/G; rs1059513) shows a strong correlation with increased bronchial responsiveness to methacholine challenge.[69] Further studies examining *STAT6* SNP rs324011 demonstrated no association with asthma[72] but revealed a strong association with total serum

IgE levels[73] and in other similar studies both rs324011 and rs1059513 were significantly associated with elevated levels of total serum IgE.[74,75] Importantly, Kabesch et al. demonstrated that *STAT6* SNP rs324011 in combination with IL-13 and IL-4 polymorphisms (C1112T and C589T, respectively) increase the risk of asthma and elevated serum IgE levels significantly above the maximum effect of any individual SNP or combination of only two SNPs.[75] Interestingly, an examination of *STAT6* SNP rs3024974 in a population of Chinese children revealed a significant association of this SNP with improved lung function assessed by forced expiratory volume.[76] Lastly, in a Caucasian population, four *STAT6* SNPs (rs3024975, rs841718, rs167769, rs703817) are significantly associated with atopic dermatitis eczema herpeticum, a disseminated herpesvirus skin infection in AD patients.[77] This observation is consistent with decreased antiviral immunity in the skin of patients with AD and mice that express a constitutively active STAT6, and may be linked to a requirement for STAT6 in multiple aspects of antiviral immunity.[77-79] Taken together, these findings provide evidence for genetic association of *STAT6* with allergic disease but also identify population-specific roles of *STAT6* polymorphisms in asthma and atopy.

Mouse models. STAT6 activation is essential for IL-4- and IL-13-induced responses and development of inflammation in several models of allergen-induced AAD. Upon induction of acute (OVA)-induced AAD, mice deficient in STAT6 display impaired Ag-induced pulmonary inflammation including reduced Th2 cytokines and eosinophilic infiltration into their airways, coincident with reduced goblet cell hyperplasia, serum IgE and AHR, in comparison to WT mice.[80-82] The degree to which eosinophil infiltration depends upon STAT6 might vary with the number of allergen challenges, with chronic challenges resulting in inflammation that is only partially STAT6-dependent.[83] Administration of IL-13 to WT mice, or transgenic mice that express a mutant IL-4Rα that has a glutamine to arginine substitution at position 576 (R576), display STAT6-dependent pulmonary inflammation associated with pulmonary eosinophilc inflammation, AHR and goblet cell hyperplasia.[84,85] Similarly, transgenic mice expressing IL-13 in airway epithelial cells develop STAT6-dependent pulmonary eosinophilia, goblet cell hyperplasia, fibrosis and AHR.[86] STAT6 is also required for the increased OVA-induced AAD observed in T-bet-deficient mice that display enhanced pulmonary eosinophilia, goblet cell hyperplasia, and Th2 cytokines in their airways in comparison to WT mice.[87] Pulmonary viral infection results in AAD similar to allergen challenge, and a similar requirement for STAT6 was observed in the AAD resulting from respiratory syncytial virus infection, or from OVA sensitization using the viral mimic poly-[I:C].[88,89]

In contrast to acute models of AAD that have shown a role for STAT6 in the development of allergen-induced pulmonary eosinophilia, goblet cell hyperplasia and AHR, a chronic model in which sensitized mice are challenged with OVA allergen over a 6 week period, results in the development of eosinophilia and AHR independent of STAT6, although goblet cell hyperplasia is STAT6-dependent in both models. Interestingly, mice doubly deficient in IL-4 and IL-13 do not develop pulmonary eosinophilia or AHR in OVA-induced chronic AAD, suggesting that these cytokines might have STAT6-independent function in some disease models.[90] In support of this, when chronic AAD is induced after mice are exposed to *Aspergillus fumigatus*, mice develop AHR in an IL-13-dependent and STAT6-independent manner.[91] Together, these data suggest that while IL-4 and IL-13 are critical for the development of pulmonary eosinophilia and AHR in chronic models of AAD, STAT6-independent mechanisms must occur to facilitate the development of these responses.

The requirement for STAT6 in the development of AAD represents function in multiple cell types. An approach that used a combination of bone marrow chimeras and adoptive transfer demonstrated that STAT6 is required both in bone marrow-derived cells and tissue resident cells.[92] Compared with WT recipients, STAT6-deficient mice that receive adoptively transferred WT Ag-specific Th2 cells are impaired in their ability to recruit transferred Th2 cells to their lungs which results in reduced Th2-induced pulmonary eosinophilia, goblet cell hyperplasia and AHR upon airway antigen challenge. Furthermore, expression of chemokines associated with the recruitment of Th2 cells and eosinophils are significantly reduced in the lungs of STAT6-deficient recipients in comparison to WT recipients.[93] Similar conclusions were made from studies using adoptive transfer of splenic T cells from WT sensitized mice to STAT6-sufficient and STAT6-deficient mice followed by allergen challenge. Transfer results in an increase in AHR, although to a lesser extent in the STAT6-deficient recipients, and occurs independently of pulmonary eosinophilia. AHR does not develop in allergen challenged STAT6-deficient recipients after adoptive transfer of splenic T cells from sensitized STAT6-deficient mice.[94] Moreover, while STAT6 is required for transgenic IL-13-induced AAD, reconstitution of hSTAT6 in the airway epithelial cells of STAT6-deficient IL-13 transgenic mice reconstitutes goblet cell hyperplasia and AHR, which suggests that STAT6 in individual cell types is sufficient to generate some aspects of AAD.[86] AAD can also be recovered in the absence of STAT6 by administration of cytokines downstream of STAT6. Injection of recombinant mIL-5 prior to Ag-challenge can reconstitute pulmonary eosinophilia and AHR in STAT6-deficient mice.[82] Similarly, intranasal administration of eotaxin reconstitutes pulmonary eosinophilia, but not goblet cell hyperplasia and AHR in Th2 cell-transferred STAT6-deficient recipients after airway challenge.[95] Together, these data suggest that STAT6 plays an obligate role in the development of many features of AAD.

STAT6VT. Although no coding mutations of STAT6 have been identified in patients, as reviewed above, atopy is often associated with increased expression or activation of STAT6. As a model of this phenomenon, transgenic mice were generated that express a mutant STAT6 that has a valine and threonine in the SH2 domain altered to alanines (STAT6VT) and is constitutively phosphorylated and transcriptionally active in the absence of IL-4 stimulation.[96,97] Transgenic mice expressing STAT6VT under control of the CD2 locus have a predisposition toward the development of allergic inflammation in lungs, skin and in periocular mucosal tissues.[32,33,97] Transgenic mice have

increased production of IgE, and STAT6VT transgenic T cells have an increased ability to differentiate into Th2 cells.[97] IL-4-deficient STAT6VT mice are protected from the spontaneous development of AAD, allergic skin inflammation, and blepharitis.[32,33] Allergic skin inflammation correlates with a decrease in epidermal differentiation complex (EDC) genes by IL-4, and decreased EDC gene expression, and barrier function, correlates with disease onset.[33,98,99] Interestingly, IL-4-deficient STAT6VT mice display an increase in EDC genes and are protected from allergic skin inflammation, demonstrating a role for IL-4 in the homeostasis of epidermal barrier function.[33] This phenotype is distinct from skin lesion-prone NC/Nga mice that develop skin lesions even in the absence of STAT6, suggesting that STAT6VT transgenic mice provide a better model for Th2-restricted inflammation.[100] STAT6VT transgenic mice are also more susceptible to vaccinia virus infection. Upon vaccinia virus skin inoculation, STAT6VT mice that are free of skin irritation at the time of exposure, display increased mortality in comparison to WT mice, correlating with enhanced viral replication in primary skin lesions and increased numbers of satellite lesions, the latter of which indicates a possible fatal systemic infection.[77] Together, these data indicate that activated STAT6 expression in T cells is sufficient to promote allergic inflammation.

STAT6 in mast cells. Although STAT6 is not required for mast cell development,[101] Th2 cytokines can limit mast cell function and inflammatory responses. STAT6-dependent IL-4 signals downregulate FcεR1 expression in mouse bone marrow-derived mast cells (BMMC), which reduces the inflammatory response associated with IgE stimulation.[102] In addition, STAT6-deficent BMMCs transduced with a retrovirus expressing constitutively active STAT6 (STAT6VT) display significantly decreased FcεR1 expression in comparison to control cells.[103] Furthermore, IL-4 induces STAT6-dependent apoptosis in WT mouse bone marrow cultures through the mitochondrial pathway. Overexpression of STAT6VT in mouse bone marrow cells cultured in the absence of IL-4 results in decreased cell survival.[104] Together, these data indicate that IL-4 induced STAT6 activation inhibits mouse bone marrow mast cell development and effector function, suggesting a homeostatic role for IL-4 and STAT6 in attenuating immune responses which are associated with mast cell function, such as allergic inflammation.

Allergic intestinal inflammation. In a mouse model of intestinal allergic inflammation, sensitized mice that receive oral administration of OVA develop severe diarrhea associated with increased serum IgE levels as well infiltration of eosinophils, mast cells and Th2 cells in the large intestine but not the small intestine. Mice deficient in STAT6 are protected from OVA-induced diarrhea and display undetected eosinophilia in the large intestine along with decreased serum IgE levels in comparison to WT mice.[105] In another study, it was shown that transgenic mice that have mIL-9 overexpressed in the enterocytes of the small intestine develop both spontaneous and oral-Ag-induced IL-4Rα- and STAT6-dependent intestinal anaphylaxis, which is associated with intestinal mastocytosis, permeability and intravascular leakage.[106] Similarly, STAT6 is required for cytokine-induced eosinophilic esophagitis.[107] Thus, IL-9-mediated mast cell responses and Th2 cytokine responses in the intestine elicit inflammation in a STAT6-dependent manner.

Inhibitors. As STAT6 is central to the development of allergic inflammation, targeting it might be an effective therapy. A cell penetrating peptide that binds to STAT6 has been shown to effectively inhibit several aspects of OVA-induced AAD. When administered to the airway prior to each allergen challenge, the STAT6 inhibitor localizes to airway epithelial cells and impairs Ag-induced pulmonary eosinophilia, goblet cell hyperplasia, IL-13 production in the airways, AHR, and induction of eotaxin-1 and mucus genes in the lung.[108] Similar effects on AAD have been shown with small molecule inhibitors of STAT6.[109] Moreover, a topical STAT6 oligodeoxynucleotide ointment reduced the severity of the inflammatory skin lesions present in patients with atopic dermatitis.[110] Although specificity and targeting continue to be challenges, STAT6 remains an attractive target for treatment of allergic inflammation.

Direct targets of STAT6 in inflamed tissue. STAT6 functions as a regulator of gene expression.[58] Although much of the published work on direct effects of STAT6 has focused on T and

Table 2. STAT target genes in airway and skin cells

STAT protein	Target gene	Cell type	Stimulation	References
STAT3	Eotaxin-1/CCL11	HASMC	IL-9	24
STAT3	Eotaxin-1/CCL11	HASMC	IL-17A	25
STAT3	Eotaxin-1/CCL11	HASMC	Oncostatin M	26
STAT3	IL-6	HASMC	PDGF	29
STAT3	IL-8/CXCL8	HASMC	PDGF	29
STAT3	VEGF	HASMC	Oncostatin M	27
STAT6	Eotaxin-1/CCL11	BEAS-2B	IL-4 and IL-13	113, 114
STAT6	IL-19	NHBE	IL-13	116
STAT6	Eotaxin-1/CCL11	Human dermal fibroblasts	IL-4	115
STAT6	Eotaxin-3/CCL26	Human dermal fibroblasts; Esophageal epithelial cells	IL-4 and IL-13	118, 119
STAT6	P-selectin	Human dermal microvascular endothelial cells	IL-4	120
STAT6	12/15-lipoxygenase	BEAS-2B	IL-4	121

B cells, and microarrays of tissues with allergic inflammation define genes that may be directly or indirectly affected by STAT6, some studies have defined roles for STAT6 in airway epithelial cells and smooth muscle cells (**Table 2**). In ASM, dermal fibroblasts, and in the human airway epithelial cell line BEAS-2B, STAT6 is required for the IL-4-stimulated expression of eotaxin-1/CCL-11.[111-115] In differentiated primary normal human bronchial epithelial (NHBE) cells, IL-13-stimulated expression of IL-19 and the goblet cell metaplasia transcription factor Sam-Pointed Domain Ets Factor (SPDEF) is STAT6-dependent.[116,117] In human dermal fibroblasts and esophageal epithelial cells, IL-4- and IL-13-stimulated eotaxin-3/CCL26 expression was found to be STAT6-dependent,[118,119] and STAT6 is required for the IL-4-induced expression of P-selectin by human dermal microvascular endothelial cells, and of 12/15-lipoxygenase in BEAS-2B cells.[120,121]

Conclusions

As required factors for cytokine responses, STATs provide a critical link between the pro-allergic microenvironment and both immune and target-organ cells. A number of the STAT proteins impact the development of T helper subsets that are important components of the inflammatory response. However, several STATs also function in resident tissue cells such as airway epithelial cells and keratinocytes, allowing response to pro-allergic cytokines that potentiate inflammation and alter cellular function, thus contributing to disease pathogenesis. Further work will define how each STAT functions at the molecular level to promote the allergic phenotype. The association of STAT gene SNPs with atopic phenotypes further supports a role for these factors in allergic inflammation and suggests specific genotypes or haplotypes that may be useful in identifying children at risk for the development of atopic disease. Targeting STATs with small molecules or biological inhibitors remains a potentially useful approach to treating allergic disease.

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References

1. Barnes PJ. Pathophysiology of allergic inflammation. Immunol Rev 2011; 242:31-50; PMID:21682737; http://dx.doi.org/10.1111/j.1600-065X.2011.01020.x
2. Galli SJ, Tsai M, Piliponsky AM. The development of allergic inflammation. Nature 2008; 454:445-54; PMID:18650915; http://dx.doi.org/10.1038/nature07204
3. Finkelman FD, Hogan SP, Hershey GK, Rothenberg ME, Wills-Karp M. Importance of cytokines in murine allergic airway disease and human asthma. J Immunol 2010; 184:1663-74; PMID:20130218; http://dx.doi.org/10.4049/jimmunol.0902185
4. Goswami R, Kaplan MH. A brief history of IL-9. J Immunol 2011; 186:3283-8; PMID:21368237; http://dx.doi.org/10.4049/jimmunol.1003049
5. Sampath D, Castro M, Look DC, Holtzman MJ. Constitutive activation of an epithelial signal transducer and activator of transcription (STAT) pathway in asthma. J Clin Invest 1999; 103:1353-61; PMID:10225979; http://dx.doi.org/10.1172/JCI6130
6. Gernez Y, Tirouvanziam R, Nguyen KD, Herzenberg LA, Krensky AM, Nadeau KC. Altered phosphorylated signal transducer and activator of transcription profile of CD4+CD161+ T cells in asthma: modulation by allergic status and oral corticosteroids. J Allergy Clin Immunol 2007; 120:1441-8; PMID:17919711; http://dx.doi.org/10.1016/j.jaci.2007.08.012
7. Pinto LA, Steudemann L, Depner M, Klopp N, Illig T, Weiland SK, et al. STAT1 gene variations, IgE regulation and atopy. Allergy 2007; 62:1456-61; PMID:17983380; http://dx.doi.org/10.1111/j.1398-9995.2007.01479.x
8. Hsieh YY, Wan L, Chang CC, Tsai CH, Tsai FJ. STAT2*C related genotypes and allele but not TLR4 and CD40 gene polymorphisms are associated with higher susceptibility for asthma. Int J Biol Sci 2009; 5:74-81; PMID:19159017
9. Shi ZO, Fischer MJ, De Sanctis GT, Schuyler MR, Tesfaigzi Y. IFN-gamma, but not Fas, mediates reduction of allergen-induced mucous cell metaplasia by inducing apoptosis. J Immunol 2002; 168:4764-71; PMID:11971027
10. Stout BA, Melendez K, Seagrave J, Holtzman MJ, Wilson B, Xiang J, et al. STAT1 activation causes translocation of Bax to the endoplasmic reticulum during the resolution of airway mucous cell hyperplasia by IFN-gamma. J Immunol 2007; 178:8107-16; PMID:17548649
11. Fulkerson PC, Zimmermann N, Hassman LM, Finkelman FD, Rothenberg ME. Pulmonary chemokine expression is coordinately regulated by STAT1, STAT6, and IFN-gamma. J Immunol 2004; 173:7565-74; PMID:15585884
12. Fulkerson PC, Zimmermann N, Brandt EB, Muntel EE, Doepker MP, Kavanaugh JL, et al. Negative regulation of eosinophil recruitment to the lung by the chemokine monokine induced by IFN-gamma (Mig, CXCL9). Proc Natl Acad Sci USA 2004; 101:1987-92; PMID:14769916; http://dx.doi.org/10.1073/pnas.0308544100
13. Sato T, Saito R, Jinushi T, Tsuji T, Matsuzaki J, Koda T, et al. IFN-gamma-induced SOCS-1 regulates STAT6-dependent eotaxin production triggered by IL-4 and TNF-alpha. Biochem Biophys Res Commun 2004; 314:468-75; PMID:14733929; http://dx.doi.org/10.1016/j.bbrc.2003.12.124
14. Quarcoo D, Weixler S, Groneberg D, Joachim R, Ahrens B, Wagner AH, et al. Inhibition of signal transducer and activator of transcription 1 attenuates allergen-induced airway inflammation and hyperreactivity. J Allergy Clin Immunol 2004; 114:288-95; PMID:15316505; http://dx.doi.org/10.1016/j.jaci.2004.03.055
15. Mikhak Z, Fleming CM, Medoff BD, Thomas SY, Tager AM, Campanella GS, et al. STAT1 in peripheral tissue differentially regulates homing of antigen-specific Th1 and Th2 cells. J Immunol 2006; 176:4959-67; PMID:16585592
16. Mikhak Z, Farsidjani A, Luster AD. Endotoxin augmented antigen-induced Th1 cell trafficking amplifies airway neutrophilic inflammation. J Immunol 2009; 182:7946-56; PMID:19494319; http://dx.doi.org/10.4049/jimmunol.0803522
17. Minegishi Y, Saito M, Tsuchiya S, Tsuge I, Takada H, Hara T, et al. Dominant-negative mutations in the DNA-binding domain of STAT3 cause hyper-IgE syndrome. Nature 2007; 448:1058-62; PMID:17676033; http://dx.doi.org/10.1038/nature06096
18. Holland SM, DeLeo FR, Elloumi HZ, Hsu AP, Uzel G, Brodsky N, et al. STAT3 mutations in the hyper-IgE syndrome. N Engl J Med 2007; 357:1608-19; PMID:17881745; http://dx.doi.org/10.1056/NEJMoa073687
19. Renner ED, Torgerson TR, Rylaarsdam S, Anover-Sombke S, Golob K, LaFlam T, et al. STAT3 mutation in the original patient with Job's syndrome. N Engl J Med 2007; 357:1667-8; PMID:17942886; http://dx.doi.org/10.1056/NEJMc076367
20. Tangye SG, Cook MC, Fulcher DA. Insights into the role of STAT3 in human lymphocyte differentiation as revealed by the hyper-IgE syndrome. J Immunol 2009; 182:21-8; PMID:19109129
21. Avery DT, Ma CS, Bryant VL, Santner-Nanan B, Nanan R, Wong M, et al. STAT3 is required for IL-21-induced secretion of IgE from human naive B cells. Blood 2008; 112:1784-93; PMID:18579794; http://dx.doi.org/10.1182/blood-2008-02-142745
22. Wjst M, Lichtner P, Meitinger T, Grimbacher B. STAT3 single-nucleotide polymorphisms and STAT3 mutations associated with hyper-IgE syndrome are not responsible for increased serum IgE serum levels in asthma families. Eur J Hum Genet 2009; 17:352-6; PMID:18841165; http://dx.doi.org/10.1038/ejhg.2008.169
23. Litonjua AA, Tantisira KG, Lake S, Lazarus R, Richter BG, Gabriel S, et al. Polymorphisms in signal transducer and activator of transcription 3 and lung function in asthma. Respir Res 2005; 6:52; PMID:15935090; http://dx.doi.org/10.1186/1465-9921-6-52
24. Yamasaki A, Saleh A, Koussih L, Muro S, Halayko AJ, Gounni AS. IL-9 induces CCL11 expression via STAT3 signalling in human airway smooth muscle cells. PLoS ONE 2010; 5:e9178; PMID:20169197; http://dx.doi.org/10.1371/journal.pone.0009178

25. Saleh A, Shan L, Halayko AJ, Kung S, Gounni AS. Critical role for STAT3 in IL-17A-mediated CCL11 expression in human airway smooth muscle cells. J Immunol 2009; 182:3357-65; PMID:19265112; http://dx.doi.org/10.4049/jimmunol.0801882
26. Faffe DS, Flynt L, Mellema M, Moore PE, Silverman ES, Subramaniam V, et al. Oncostatin M causes eotaxin-1 release from airway smooth muscle: synergy with IL-4 and IL-13. J Allergy Clin Immunol 2005; 115:514-20; PMID:15753898; http://dx.doi.org/10.1016/j.jaci.2004.11.033
27. Faffe DS, Flynt L, Mellema M, Whitehead TR, Bourgeois K, Panettieri RA, Jr., et al. Oncostatin M causes VEGF release from human airway smooth muscle: synergy with IL-1beta. Am J Physiol Lung Cell Mol Physiol 2005; 288:L1040-8; PMID:15665043; http://dx.doi.org/10.1152/ajplung.00333.2004
28. Shan L, Redhu NS, Saleh A, Halayko AJ, Chakir J, Gounni AS. Thymic stromal lymphopoietin receptor-mediated IL-6 and CC/CXC chemokines expression in human airway smooth muscle cells: role of MAPKs (ERK1/2, p38, and JNK) and STAT3 pathways. J Immunol 2010; 184:7134-43; PMID:20483734; http://dx.doi.org/10.4049/jimmunol.0902515
29. Simeone-Penney MC, Severgnini M, Rozo L, Takahashi S, Cochran BH, Simon AR. PDGF-induced human airway smooth muscle cell proliferation requires STAT3 and the small GTPase Rac1. Am J Physiol Lung Cell Mol Physiol 2008; 294:L698-704; PMID:18310224; http://dx.doi.org/10.1152/ajplung.00529.2007
30. Simeone-Penney MC, Severgnini M, Tu P, Homer RJ, Mariani TJ, Cohn L, et al. Airway epithelial STAT3 is required for allergic inflammation in a murine model of asthma. J Immunol 2007; 178:6191-9; PMID:17475846
31. Stritesky GL, Muthukrishnan R, Sehra S, Goswami R, Pham D, Travers J, et al. The transcription factor STAT3 is required for T helper 2 cell development. Immunity 2011; 34:39-49; PMID:21215659; http://dx.doi.org/10.1016/j.immuni.2010.12.013
32. Sehra S, Bruns HA, Ahyi AN, Nguyen ET, Schmidt NW, Michels EG, et al. IL-4 is a critical determinant in the generation of allergic inflammation initiated by a constitutively active Stat6. J Immunol 2008; 180:3551-9; PMID:18292582
33. Sehra S, Yao Y, Howell MD, Nguyen ET, Kansas GS, Leung DY, et al. IL-4 regulates skin homeostasis and the predisposition toward allergic skin inflammation. J Immunol 2010; 184:3186-90; PMID:20147633; http://dx.doi.org/10.4049/jimmunol.0901860
34. Kaplan MH. STAT4: a critical regulator of inflammation in vivo. Immunol Res 2005; 31:231-42; PMID:15888914; http://dx.doi.org/10.1385/IR:31:3:231
35. Mathur AN, Chang HC, Zisoulis DG, Stritesky GL, Yu Q, O'Malley JT, et al. Stat3 and Stat4 direct development of IL-17-secreting Th cells. J Immunol 2007; 178:4901-7; PMID:17404271
36. Nguyen KB, Watford WT, Salomon R, Hofmann SR, Pien GC, Morinobu A, et al. Critical role for STAT4 activation by type 1 interferons in the interferon-gamma response to viral infection. Science 2002; 297:2063-6; PMID:12242445; http://dx.doi.org/10.1126/science.1074900
37. Park BL, Cheong HS, Kim LH, Choi YH, Namgoong S, Park HS, et al. Association analysis of signal transducer and activator of transcription 4 (STAT4) polymorphisms with asthma. J Hum Genet 2005; 50:133-8; PMID:15744455; http://dx.doi.org/10.1007/s10038-005-0232-1
38. Pykäläinen M, Kinos R, Valkonen S, Rydman P, Kilpelainen M, Laitinen LA, et al. Association analysis of common variants of STAT6, GATA3, and STAT4 to asthma and high serum IgE phenotypes. J Allergy Clin Immunol 2005; 115:80-7; PMID:15637551; http://dx.doi.org/10.1016/j.jaci.2004.10.006
39. Li Y, Wu B, Xiong H, Zhu C, Zhang L. Polymorphisms of STAT-6, STAT-4 and IFN-gamma genes and the risk of asthma in Chinese population. Respir Med 2007; 101:1977-81; PMID:17532201; http://dx.doi.org/10.1016/j.rmed.2007.04.006
40. Behera AK, Kumar M, Lockey RF, Mohapatra SS. Adenovirus-mediated interferon gamma gene therapy for allergic asthma: involvement of interleukin 12 and STAT4 signaling. Hum Gene Ther 2002; 13:1697-709; PMID:12396623; http://dx.doi.org/10.1089/10430340276029354 7
41. Matsubara S, Takeda K, Kodama T, Joetham A, Miyahara N, Koya T, et al. IL-2 and IL-18 attenuation of airway hyperresponsiveness requires STAT4, IFN-gamma, and natural killer cells. Am J Respir Cell Mol Biol 2007; 36:324-32; PMID:17038663; http://dx.doi.org/10.1165/rcmb.2006-0231OC
42. Raman K, Kaplan MH, Hogaboam CM, Berlin A, Lukacs NW. STAT4 signal pathways regulate inflammation and airway physiology changes in allergic airway inflammation locally via alteration of chemokines. J Immunol 2003; 170:3859-65; PMID:12646654
43. Furuta S, Kagami S, Tamachi T, Ikeda K, Fujiwara M, Suto A, et al. Overlapping and distinct roles of STAT4 and T-bet in the regulation of T cell differentiation and allergic airway inflammation. J Immunol 2008; 180:6656-62; PMID:18453585
44. O'Malley JT, Sehra S, Thieu VT, Yu Q, Chang HC, Stritesky GL, et al. Signal transducer and activator of transcription 4 limits the development of adaptive regulatory T cells. Immunology 2009; 127:587-95; PMID:19604309; http://dx.doi.org/10.1111/j.1365-2567.2008.03037.x
45. Eisenbarth SC, Piggott DA, Huleatt JW, Visintin I, Herrick CA, Bottomly K. Lipopolysaccharide-enhanced, toll-like receptor 4-dependent T helper cell type 2 responses to inhaled antigen. J Exp Med 2002; 196:1645-51; PMID:12486107; http://dx.doi.org/10.1084/jem.20021340
46. Kim YK, Oh SY, Jeon SG, Park HW, Lee SY, Chun EY, et al. Airway exposure levels of lipopolysaccharide determine type 1 versus type 2 experimental asthma. J Immunol 2007; 178:5375-82; PMID:17404323
47. Isaksen DE, Baumann H, Zhou B, Nivollet S, Farr AG, Levin SD, et al. Uncoupling of proliferation and Stat5 activation in thymic stromal lymphopoietin-mediated signal transduction. J Immunol 2002; 168:3288-94; PMID:11907084
48. Rochman Y, Kashyap M, Robinson GW, Sakamoto K, Gomez-Rodriguez J, Wagner KU, et al. Thymic stromal lymphopoietin-mediated STAT5 phosphorylation via kinases JAK1 and JAK2 reveals a key difference from IL-7-induced signaling. Proc Natl Acad Sci USA 2010; 107:19455-60; PMID:20974963; http://dx.doi.org/10.1073/pnas.1008271107
49. Lu N, Wang YH, Wang YH, Arima K, Hanabuchi S, Liu YJ. TSLP and IL-7 use two different mechanisms to regulate human CD4+ T cell homeostasis. J Exp Med 2009; 206:2111-9; PMID:19770269; http://dx.doi.org/10.1084/jem.20090153
50. Shelburne CP, McCoy ME, Piekorz R, Sexl V, Roh KH, Jacobs-Helber SM, et al. Stat5 expression is critical for mast cell development and survival. Blood 2003; 102:1290-7; PMID:12714518; http://dx.doi.org/10.1182/blood-2002-11-3490
51. Barnstein BO, Li G, Wang Z, Kennedy S, Chalfant C, Nakajima H, et al. Stat5 expression is required for IgE-mediated mast cell function. J Immunol 2006; 177:3421-6; PMID:16920984
52. Kagami S, Nakajima H, Kumano K, Suzuki K, Suto A, Imada K, et al. Both stat5a and stat5b are required for antigen-induced eosinophil and T-cell recruitment into the tissue. Blood 2000; 95:1370-7; PMID:10666213
53. Zhu Y, Chen L, Huang Z, Alkan S, Bunting KD, Wen R, et al. Cutting edge: IL-5 primes Th2 cytokine-producing capacity in eosinophils through a STAT5-dependent mechanism. J Immunol 2004; 173:2918-22; PMID:15322148
54. Takatori H, Nakajima H, Hirose K, Kagami S, Tamachi T, Suto A, et al. Indispensable role of Stat5a in Stat6-independent Th2 cell differentiation and allergic airway inflammation. J Immunol 2005; 174:3734-40; PMID:15749913
55. Kaplan MH, Schindler U, Smiley ST, Grusby MJ. Stat6 is required for mediating responses to IL-4 and for development of Th2 cells. Immunity 1996; 4:313-9; PMID:8624821; http://dx.doi.org/10.1016/S1074-7613(00)80439-2
56. Shimoda K, van Deursen J, Sangster MY, Sarawar SR, Carson RT, Tripp RA, et al. Lack of IL-4-induced Th2 response and IgE class switching in mice with disrupted Stat6 gene. Nature 1996; 380:630-3; PMID:8602264; http://dx.doi.org/10.1038/380630a0
57. Takeda K, Tanaka T, Shi W, Matsumoto M, Minami M, Kashiwamura S, et al. Essential role of Stat6 in IL-4 signalling. Nature 1996; 380:627-30; PMID:8602263; http://dx.doi.org/10.1038/380627a0
58. Goenka S, Kaplan MH. Transcriptional regulation by STAT6. Immunol Res 2011; 50:87-96; PMID:21442426; http://dx.doi.org/10.1007/s12026-011-8205-2
59. Ghaffar O, Christodoulopoulos P, Lamkhioued B, Wright E, Ihaku D, Nakamura Y, et al. In vivo expression of signal transducer and activator of transcription factor 6 (STAT6) in nasal mucosa from atopic allergic rhinitis: effect of topical corticosteroids. Clin Exp Allergy 2000; 30:86-93; PMID:10606935; http://dx.doi.org/10.1046/j.1365-2222.2000.00781.x
60. Christodoulopoulos P, Cameron L, Nakamura Y, Lemiere C, Muro S, Dugas M, et al. TH2 cytokine-associated transcription factors in atopic and nonatopic asthma: evidence for differential signal transducer and activator of transcription 6 expression. J Allergy Clin Immunol 2001; 107:586-91; PMID:11295643; http://dx.doi.org/10.1067/mai.2001.114883
61. Taha R, Hamid Q, Cameron L, Olivenstein R. T helper type 2 cytokine receptors and associated transcription factors GATA-3, c-MAF, and signal transducer and activator of transcription factor-6 in induced sputum of atopic asthmatic patients. Chest 2003; 123:2074-82; PMID:12796191; http://dx.doi.org/10.1378/chest.123.6.2074
62. Miller RL, Eppinger TM, McConnell D, Cunningham-Rundles C, Rothman P. Analysis of cytokine signaling in patients with extrinsic asthma and hyperimmunoglobulin E. J Allergy Clin Immunol 1998; 102:503-11; PMID:9768594; http://dx.doi.org/10.1016/S0091-6749(98)70141-1
63. Mullings RE, Wilson SJ, Puddicombe SM, Lordan JL, Bucchieri F, Djukanovic R, et al. Signal transducer and activator of transcription 6 (STAT-6) expression and function in asthmatic bronchial epithelium. J Allergy Clin Immunol 2001; 108:832-8; PMID:11692112; http://dx.doi.org/10.1067/mai.2001.119554
64. Phipps S, Benyahia F, Ou TT, Barkans J, Robinson DS, Kay AB. Acute allergen-induced airway remodeling in atopic asthma. Am J Respir Cell Mol Biol 2004; 31:626-32; PMID:15333330; http://dx.doi.org/10.1165/rcmb.2004-0193OC
65. Gao PS, Mao XQ, Roberts MH, Arinobu Y, Akaiwa M, Enomoto T, et al. Variants of STAT6 (signal transducer and activator of transcription 6) in atopic asthma. J Med Genet 2000; 37:380-2; PMID:10905892; http://dx.doi.org/10.1136/jmg.37.5.380a

66. Amoli MM, Hand S, Hajeer AH, Jones KP, Rolf S, Sting C, et al. Polymorphism in the STAT6 gene encodes risk for nut allergy. Genes Immun 2002; 3: 220-4; PMID:12058257; http://dx.doi.org/10.1038/sj.gene.6363872
67. Tamura K, Arakawa H, Suzuki M, Kobayashi Y, Mochizuki H, Kato M, et al. Novel dinucleotide repeat polymorphism in the first exon of the STAT-6 gene is associated with allergic diseases. Clin Exp Allergy 2001; 31:1509-14; PMID:11678849; http://dx.doi.org/10.1046/j.1365-2222.2001.01191.x
68. Shao C, Suzuki Y, Kamada F, Kanno K, Tamari M, Hasegawa K, et al. Linkage and association of childhood asthma with the chromosome 12 genes. J Hum Genet 2004; 49:115-22; PMID:14767694; http://dx.doi.org/10.1007/s10038-003-0118-z
69. Duetsch G, Illig T, Loesgen S, Rohde K, Klopp N, Herbon N, et al. STAT6 as an asthma candidate gene: polymorphism-screening, association and haplotype analysis in a Caucasian sib-pair study. Hum Mol Genet 2002; 11:613-21; PMID:11912176; http://dx.doi.org/10.1093/hmg/11.6.613
70. Nagarkatti R. C BR, Vijayan V, Sharma SK, Ghosh B. Signal transducer and activator of transcription 6 haplotypes and asthma in the Indian population. Am J Respir Cell Mol Biol 2004; 31:317-21; PMID:15105161; http://dx.doi.org/10.1165/rcmb.2003-0128OC
71. Gao PS, Heller NM, Walker W, Chen CH, Moller M, Plunkett B, et al. Variation in dinucleotide (GT) repeat sequence in the first exon of the STAT6 gene is associated with atopic asthma and differentially regulates the promoter activity in vitro. J Med Genet 2004; 41:535-9; PMID:15235025; http://dx.doi.org/10.1136/jmg.2003.015842
72. Kavalar MS, Balantic M, Silar M, Kosnik M, Korosec P, Rijavec M. Association of ORMDL3, STAT6 and TBXA2R gene polymorphisms with asthma. Int J Immunogenet 2012; 39:20-5; PMID:22017802; http://dx.doi.org/10.1111/j.1744-313X.2011.01051.x
73. Weidinger S, Klopp N, Wagenpfeil S, Rummler L, Schedel M, Kabesch M, et al. Association of a STAT 6 haplotype with elevated serum IgE levels in a population based cohort of white adults. J Med Genet 2004; 41:658-63; PMID:15342695; http://dx.doi.org/10.1136/jmg.2004.020263
74. Schedel M, Carr D, Klopp N, Woitsch B, Illig T, Stachel D, et al. A signal transducer and activator of transcription 6 haplotype influences the regulation of serum IgE levels. J Allergy Clin Immunol 2004; 114:1100-5; PMID:15536416; http://dx.doi.org/10.1016/j.jaci.2004.07.048
75. Kabesch M, Schedel M, Carr D, Woitsch B, Fritzsch C, Weiland SK, et al. IL-4/IL-13 pathway genetics strongly influence serum IgE levels and childhood asthma. J Allergy Clin Immunol 2006; 117:269-74; PMID:16461126; http://dx.doi.org/10.1016/j.jaci.2005.10.024
76. Leung TF, Chan IH, Wong GW, Li CY, Tang NL, Yung E, et al. Association between candidate genes and lung function growth in Chinese asthmatic children. Clin Exp Allergy 2007; 37:1480-6; PMID:17883727
77. Howell MD, Gao P, Kim BE, Lesley LJ, Streib JE, Taylor PA, et al. The signal transducer and activator of transcription 6 gene (STAT6) increases the propensity of patients with atopic dermatitis toward disseminated viral skin infections. J Allergy Clin Immunol 2011; 128:1006-14; PMID:21762972; http://dx.doi.org/10.1016/j.jaci.2011.06.003
78. Howell MD, Gallo RL, Boguniewicz M, Jones JF, Wong C, Streib JE, et al. Cytokine milieu of atopic dermatitis skin subverts the innate immune response to vaccinia virus. Immunity 2006; 24:341-8; PMID:16546102; http://dx.doi.org/10.1016/j.immuni.2006.02.006
79. Chen H, Sun H, You F, Sun W, Zhou X, Chen L, et al. Activation of STAT6 by STING Is Critical for Antiviral Innate Immunity. Cell 2011; 147:436-46; PMID:22000020; http://dx.doi.org/10.1016/j.cell.2011.09.022
80. Kuperman D, Schofield B, Wills-Karp M, Grusby MJ. Signal transducer and activator of transcription factor 6 (Stat6)-deficient mice are protected from antigen-induced airway hyperresponsiveness and mucus production. J Exp Med 1998; 187:939-48; PMID:9500796; http://dx.doi.org/10.1084/jem.187.6.939
81. Akimoto T, Numata F, Tamura M, Takata Y, Higashida N, Takashi T, et al. Abrogation of bronchial eosinophilic inflammation and airway hyperreactivity in signal transducers and activators of transcription (STAT)6-deficient mice. J Exp Med 1998; 187:1537-42; PMID:9565645; http://dx.doi.org/10.1084/jem.187.9.1537
82. Tomkinson A, Kanehiro A, Rabinovitch N, Joetham A, Cieslewicz G, Gelfand EW. The failure of STAT6-deficient mice to develop airway eosinophilia and airway hyperresponsiveness is overcome by interleukin-5. Am J Respir Crit Care Med 1999; 160:1283-91; PMID:10508820
83. Trifilieff A, El-Hasim A, Corteling R, Owen CE. Abrogation of lung inflammation in sensitized Stat6-deficient mice is dependent on the allergen inhalation procedure. Br J Pharmacol 2000; 130:1581-8; PMID:10928961; http://dx.doi.org/10.1038/sj.bjp.0703501
84. Tachdjian R, Mathias C, Al Khatib S, Bryce PJ, Kim HS, Blaeser F, et al. Pathogenicity of a disease-associated human IL-4 receptor allele in experimental asthma. J Exp Med 2009; 206:2191-204; PMID:19770271; http://dx.doi.org/10.1084/jem.20091480
85. Yang M, Hogan SP, Henry PJ, Matthaei KI, McKenzie AN, Young IG, et al. Interleukin-13 mediates airways hyperreactivity through the IL-4 receptor-alpha chain and STAT-6 independently of IL-5 and eotaxin. Am J Respir Cell Mol Biol 2001; 25:522-30; PMID:11694459
86. Kuperman DA, Huang X, Koth LL, Chang GH, Dolganov GM, Zhu Z, et al. Direct effects of interleukin-13 on epithelial cells cause airway hyperreactivity and mucus overproduction in asthma. Nat Med 2002; 8:885-9; PMID:12091879
87. Fujiwara M, Hirose K, Kagami S, Takatori H, Wakashin H, Tamachi T, et al. T-bet inhibits both TH2 cell-mediated eosinophil recruitment and TH17 cell-mediated neutrophil recruitment into the airways. J Allergy Clin Immunol 2007; 119:662-70; PMID:17336616; http://dx.doi.org/10.1016/j.jaci.2006.12.643
88. Jeon SG, Oh SY, Park HK, Kim YS, Shim EJ, Lee HS, et al. TH2 and TH1 lung inflammation induced by airway allergen sensitization with low and high doses of double-stranded RNA. J Allergy Clin Immunol 2007; 120:803-12; PMID:17610940; http://dx.doi.org/10.1016/j.jaci.2007.05.030
89. Tekkanat KK, Maassab HF, Cho DS, Lai JJ, John A, Berlin A, et al. IL-13-induced airway hyperreactivity during respiratory syncytial virus infection is STAT6 dependent. J Immunol 2001; 166:3542-8; PMID:11207314
90. Foster PS, Webb DC, Yang M, Herbert C, Kumar RK. Dissociation of T helper type 2 cytokine-dependent airway lesions from signal transducer and activator of transcription 6 signalling in experimental chronic asthma. Clin Exp Allergy 2003; 33:688-95; PMID:12752600; http://dx.doi.org/10.1046/j.1365-2222.2003.01647.x
91. Blease K, Schuh JM, Jakubzick C, Lukacs NW, Kunkel SL, Joshi BH, et al. Stat6-deficient mice develop airway hyperresponsiveness and peribronchial fibrosis during chronic fungal asthma. Am J Pathol 2002; 160:481-90; PMID:11839568; http://dx.doi.org/10.1016/S0002-9440(10)64867-5
92. Chapoval SP, Dasgupta P, Smith EP, DeTolla LJ, Lipsky MM, Kelly-Welch AE, et al. STAT6 expression in multiple cell types mediates the cooperative development of allergic airway disease. J Immunol 2011; 186:2571-83; PMID:21242523; http://dx.doi.org/10.4049/jimmunol.1002567
93. Mathew A, MacLean JA, DeHaan E, Tager AM, Green FH, Luster AD. Signal transducer and activator of transcription 6 controls chemokine production and T helper cell type 2 cell trafficking in allergic pulmonary inflammation. J Exp Med 2001; 193:1087-96; PMID:11342593; http://dx.doi.org/10.1084/jem.193.9.1087
94. Tomkinson A, Duez C, Lahn M, Gelfand EW. Adoptive transfer of T cells induces airway hyperresponsiveness independently of airway eosinophilia but in a signal transducer and activator of transcription 6-dependent manner. J Allergy Clin Immunol 2002; 109:810-6; PMID:11994705; http://dx.doi.org/10.1067/mai.2002.123531
95. Hoshino A, Tsuji T, Matsuzaki J, Jinushi T, Ashino S, Teramura T, et al. STAT6-mediated signaling in Th2-dependent allergic asthma: critical role for the development of eosinophilia, airway hyper-responsiveness and mucus hypersecretion, distinct from its role in Th2 differentiation. Int Immunol 2004; 16:1497-505; PMID:15351784; http://dx.doi.org/10.1093/intimm/dxh151
96. Daniel C, Salvekar A, Schindler U. A gain-of-function mutation in STAT6. J Biol Chem 2000; 275:14255-9; PMID:10747856; http://dx.doi.org/10.1074/jbc.C000129200
97. Bruns HA, Schindler U, Kaplan MH. Expression of a constitutively active Stat6 in vivo alters lymphocyte homeostasis with distinct effects in T and B cells. J Immunol 2003; 170:3478-87; PMID:12646608
98. Kim BE, Leung DY, Boguniewicz M, Howell MD. Loricrin and involucrin expression is down-regulated by Th2 cytokines through STAT-6. Clin Immunol 2008; 126:332-7; PMID:18166499; http://dx.doi.org/10.1016/j.clim.2007.11.006
99. DaSilva SC, Sahu RP, Konger RL, Perkins SM, Kaplan MH, Travers JB. Increased skin barrier disruption by sodium lauryl sulfate in mice expressing a constitutively active STAT6 in T cells. Arch Dermatol Res 2012; 304:65-71; PMID:21959772; http://dx.doi.org/10.1007/s00403-011-1168-2
100. Yagi R, Nagai H, Iigo Y, Akimoto T, Arai T, Kubo M. Development of atopic dermatitis-like skin lesions in STAT6-deficient NC/Nga mice. J Immunol 2002; 168:2020-7; PMID:11823539
101. Sherman MA, Secor VH, Lee SK, Lopez RD, Brown MA. STAT6-independent production of IL-4 by mast cells. Eur J Immunol 1999; 29:1235-42; PMID:10229091; http://dx.doi.org/10.1002/(SICI)1521-4141(199904)29:04<1235::AID-IMMU1235>3.0.CO;2-0
102. Ryan JJ, DeSimone S, Klisch G, Shelburne C, McReynolds LJ, Han K, et al. IL-4 inhibits mouse mast cell Fc epsilonRI expression through a STAT6-dependent mechanism. J Immunol 1998; 161:6915-23; PMID:9862725

103. Gillespie SR, DeMartino RR, Zhu J, Chong HJ, Ramirez C, Shelburne CP, et al. IL-10 inhibits Fc epsilon RI expression in mouse mast cells. J Immunol 2004; 172:3181-8; PMID:14978125
104. Bailey DP, Kashyap M, Mirmonsef P, Bouton LA, Domen J, Zhu J, et al. Interleukin-4 elicits apoptosis of developing mast cells via a Stat6-dependent mitochondrial pathway. Exp Hematol 2004; 32:52-9; PMID:14725901; http://dx.doi.org/10.1016/j.exphem.2003.10.011
105. Kweon MN, Yamamoto M, Kajiki M, Takahashi I, Kiyono H. Systemically derived large intestinal CD4(+) Th2 cells play a central role in STAT6-mediated allergic diarrhea. J Clin Invest 2000; 106:199-206; PMID:10903335; http://dx.doi.org/10.1172/JCI8490
106. Forbes EE, Groschwitz K, Abonia JP, Brandt EB, Cohen E, Blanchard C, et al. IL-9- and mast cell-mediated intestinal permeability predisposes to oral antigen hypersensitivity. J Exp Med 2008; 205:897-913; PMID:18378796; http://dx.doi.org/10.1084/jem.20071046
107. Mishra A, Rothenberg ME. Intratracheal IL-13 induces eosinophilic esophagitis by an IL-5, eotaxin-1, and STAT6-dependent mechanism. Gastroenterology 2003; 125:1419-27; PMID:14598258; http://dx.doi.org/10.1016/j.gastro.2003.07.007
108. McCusker CT, Wang Y, Shan J, Kinyanjui MW, Villeneuve A, Michael H, et al. Inhibition of experimental allergic airways disease by local application of a cell-penetrating dominant-negative STAT-6 peptide. J Immunol 2007; 179:2556-64; PMID:17675518
109. Nagashima S, Hondo T, Nagata H, Ogiyama T, Maeda J, Hoshii H, et al. Novel 7H-pyrrolo[2,3-d]pyrimidine derivatives as potent and orally active STAT6 inhibitors. Bioorg Med Chem 2009; 17:6926-36; PMID:19747833; http://dx.doi.org/10.1016/j.bmc.2009.08.021
110. Igawa K, Satoh T, Yokozeki H. A therapeutic effect of STAT6 decoy oligodeoxynucleotide ointment in atopic dermatitis: a pilot study in adults. Br J Dermatol 2009; 160:1124-6; PMID:19292714; http://dx.doi.org/10.1111/j.1365-2133.2009.09070.x
111. Odaka M, Matsukura S, Kuga H, Kokubu F, Kasama T, Kurokawa M, et al. Differential regulation of chemokine expression by Th1 and Th2 cytokines and mechanisms of eotaxin/CCL-11 expression in human airway smooth muscle cells. Int Arch Allergy Immunol 2007; 143(Suppl 1):84-8; PMID:17541284; http://dx.doi.org/10.1159/000101412
112. Peng Q, Matsuda T, Hirst SJ. Signaling pathways regulating interleukin-13-stimulated chemokine release from airway smooth muscle. Am J Respir Crit Care Med 2004; 169:596-603; PMID:14670803; http://dx.doi.org/10.1164/rccm.200307-888OC
113. Matsukura S, Stellato C, Plitt JR, Bickel C, Miura K, Georas SN, et al. Activation of eotaxin gene transcription by NF-kappa B and STAT6 in human airway epithelial cells. J Immunol 1999; 163:6876-83; PMID:10586089
114. Matsukura S, Stellato C, Georas SN, Casolaro V, Plitt JR, Miura K, et al. Interleukin-13 upregulates eotaxin expression in airway epithelial cells by a STAT6-dependent mechanism. Am J Respir Cell Mol Biol 2001; 24:755-61; PMID:11415942
115. Hoeck J, Woisetschlager M. STAT6 mediates eotaxin-1 expression in IL-4 or TNF-alpha-induced fibroblasts. J Immunol 2001; 166:4507-15; PMID:11254707
116. Huang F, Wachi S, Thai P, Loukoianov A, Tan KH, Forteza RM, Wu R. Potentiation of IL-19 expression in airway epithelia by IL-17A and IL-4/IL-13: important implications in asthma. J Allergy Clin Immunol 2008; 121:1415-21; PMID:18539194; http://dx.doi.org/10.1016/j.jaci.2008.04.016
117. Park KS, Korfhagen TR, Bruno MD, Kitzmiller JA, Wan H, Wert SE, et al. SPDEF regulates goblet cell hyperplasia in the airway epithelium. J Clin Invest 2007; 117:978-88; PMID:17347682; http://dx.doi.org/10.1172/JCI29176
118. Hoeck J, Woisetschlager M. Activation of eotaxin-3/CCLl26 gene expression in human dermal fibroblasts is mediated by STAT6. J Immunol 2001; 167:3216-22; PMID:11544308
119. Lim EJ, Lu TX, Blanchard C, Rothenberg ME. Epigenetic regulation of the IL-13-induced human eotaxin-3 gene by CREB-binding protein-mediated histone 3 acetylation. J Biol Chem 2011; 286:13193-204; PMID:21325281; http://dx.doi.org/10.1074/jbc.M110.210724
120. Miyazaki Y, Satoh T, Nishioka K, Yokozeki H. STAT-6-mediated control of P-selectin by substance P and interleukin-4 in human dermal endothelial cells. Am J Pathol 2006; 169:697-707; PMID:16877367; http://dx.doi.org/10.2353/ajpath.2006.051211
121. Conrad DJ, Lu M. Regulation of human 12/15-lipoxygenase by Stat6-dependent transcription. Am J Respir Cell Mol Biol 2000; 22:226-34; PMID:10657944