

Distinct roles for IL-13 and IL-4 via IL-13 receptor $\alpha 1$ and the type II IL-4 receptor in asthma pathogenesis

A. Munitz*, E. B. Brandt*, M. Mingler*, F. D. Finkelman[†], and M. E. Rothenberg*[‡]

*Division of Allergy and Immunology, Cincinnati Children's Hospital Medical Center, 3333 Burnet Avenue, Cincinnati, OH 45229; and [†]Department of Medicine, Cincinnati Veterans Affairs Medical Center, 3200 Vine Street, Cincinnati, OH 45220

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IL-13 and IL-4 are central T helper 2 (Th2) cytokines in the immune system and potent activators of inflammatory responses and fibrosis during Th2 inflammation. Recent studies using *Il13ra1*^{-/-} mice have demonstrated a critical role for IL-13 receptor (IL-13R) $\alpha 1$ in allergen-induced airway responses. However, these observations require further attention especially because IL-4 can induce similar lung pathology to IL-13, independent of IL-13, and is still present in the allergic lung. Thus, we hypothesized that IL-13R $\alpha 1$ regulates IL-4-induced responses in the lung. To dissect the role of IL-13R $\alpha 1$ and the type I and II IL-4Rs in experimental asthma, we examined lung pathology induced by allergen, IL-4, and IL-13 challenge in *Il13ra1*^{-/-} mice. We report that IL-13R $\alpha 1$ is essential for baseline IgE production, but Th2 and IgE responses to T cell-dependent antigens are IL-13R $\alpha 1$ -independent. Furthermore, we demonstrate that increased airway resistance, mucus, TGF- β , and eotaxin(s) production, but not cellular infiltration, are critically dependent on IL-13R $\alpha 1$. Surprisingly, our results identify a CCR3- and IL-13R $\alpha 1$ -independent pathway for lung eosinophilia. Global expression profiling of lungs from mice stimulated with allergen or IL-4 demonstrated that marker genes of alternatively activated macrophages are differentially regulated by the type I and type II IL-4R. Taken together, our data provide a comprehensive mechanistic analysis of the critical role by which IL-13R $\alpha 1$ mediates allergic lung pathology and highlight unforeseen roles for the type II IL-4R.

inflammation | cytokines | eosinophils | chemokines | mucus

Interleukin 13 is a central immune regulator of many hallmark type 2 disease characteristics, including IgE synthesis, mucus hypersecretion, airway hyperreactivity, and fibrosis (1). IL-13 shares overlapping biological functions with IL-4 (1, 2), and both signal via a complex network of receptors. IL-4 mediates its effects through either the type I IL-4 receptor (IL-4R) (i.e., IL-4R α and the common γ chain) or the type II IL-4R (i.e., IL-4R α and IL-13R $\alpha 1$). In contrast, IL-13 is hypothesized to execute its IL-4R α -dependent effects solely through the type II IL-4R but may use a signaling complex that does not require IL-4R α (3). In addition, IL-13R $\alpha 2$, an IL-13 decoy receptor (4), has been recently reported to also mediate IL-13 signaling and induce TGF- β production (5, 6). Thus, the assumption that IL-13R $\alpha 1$ is the main signaling receptor for IL-13 needs definitive proof.

Although IL-4 and IL-13 initiate similar intracellular signaling cascades, IL-13 is capable of exerting specific and IL-4-independent signals (4, 7). In addition, IL-4 can induce lung pathology even in the absence of IL-13, and treatment with an IL-13 antagonist does not inhibit the effects of IL-4 (8). Yet it is currently unknown whether these IL-13-independent effects of IL-4 are mediated via the type I or type II IL-4R (9).

A valuable way to distinguish the role of these two receptors is by genetic deletion of the IL-13R $\alpha 1$ chain, because such genetically engineered mice would harbor a functional deletion of the type II IL-4R but have an intact type I IL-4R.

A recent study using *Il13ra1*^{-/-} mice has shown that these mice are protected from *Schistosoma mansoni* egg antigen-induced airway hyperreactivity and mucus hypersecretion (10). However, these

results require further clarification especially because IL-4 is still up-regulated in the lungs of these mice and could potentially induce airway hyperreactivity and mucus production (8).

Results

***Il13ra1*^{-/-} Mice Display Enhanced Circulating Soluble IL-13R $\alpha 2$ and IL-13.** Soluble IL-13R $\alpha 2$ (sIL-13R $\alpha 2$) has been proposed to counterregulate IL-13 activities (4, 11–13) via an autoregulatory pathway initiated by IL-13R $\alpha 1$ (5). Contrary to the current paradigm, which would have predicted decreased levels of sIL-13R $\alpha 2$ (5, 14), *Il13ra1*^{-/-} [supporting information (SI) Fig. S1] mice had elevated circulating total sIL-13R $\alpha 2$, increased sIL-13R $\alpha 2$:IL-13 complexes (indicating increased IL-13 levels), and markedly increased saturation of IL-13R $\alpha 2$ with IL-13 (Fig. 1 A–C).

IL-13R $\alpha 1$ Is Critical for Maintenance of Homeostatic IgE Independent of Changes in IL-4. *Il13ra1*^{-/-} mice had barely detectable IgE (Fig. 1 D and E) and displayed a minor increase in IgG2a levels but no changes in IgA, IgM, IgG1, IgG3, or IgG2b levels (Fig. S2 A–F).

Given the crucial role of IL-4 in IgE production (15–17), we examined IL-4 levels and signaling components in *Il13ra1*^{-/-} mice. Serum IL-4 and IFN γ levels, IL-4R α expression, and STAT6 phosphorylation in response to IL-4 were comparable between WT and *Il13ra1*^{-/-} splenocytes (Fig. S3 A–E). In response to CD3/CD28 stimulation, *Il13ra1*^{-/-} splenocytes produced normal amounts of IL-4 whereas IFN γ production was decreased (Fig. S3 F–H).

IL-13R $\alpha 1$ Is Dispensable for Polarized T helper 2 (Th2) Responses *in Vivo*. Subsequently, we examined the ability of *Il13ra1*^{-/-} mice to manifest an acquired Th2 response. After treatment with goat anti-mouse IgD (G α M-IgD), a potent Th2 polarizing agent (18), both *Il13ra1*^{-/-} and WT mice had comparable serum IL-4 and IFN- γ levels (Fig. 1 F) and, accordingly, similar serum IgE levels (Fig. 1 G).

Assessment of IL-13R $\alpha 1$ -Mediated Responses in a Model of IL-13-Induced Airway Inflammation. To directly define the role of IL-13R $\alpha 1$ in IL-13-induced lung responses, IL-13 was administered intratracheally to *Il13ra1*^{-/-} mice and lung inflammation was assessed. IL-13 strongly induced chemokine expression (i.e., CCL2, CCL17, CCL11, and CCL24) in WT mice but not *Il13ra1*^{-/-} mice (Fig. 2 A–D).

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[‡]To whom correspondence should be addressed. E-mail: rothenberg@cchmc.org.

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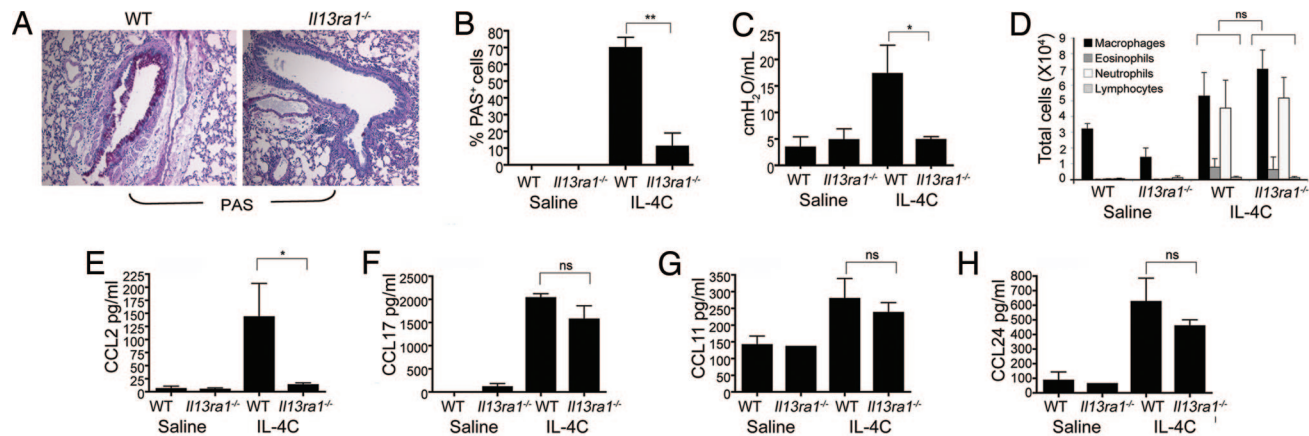


Fig. 4. Assessment of IL-13Rα1-mediated responses in a model of IL-4-induced airway inflammation. Twenty-four hours after the final IL-4C challenge the mice were assessed for mucus production (A and B), airway resistance (C), BALF cellular infiltration (D), and chemokine production (E–H). The data, representative of one of two experiments, are presented as mean $\pm$ SD (nine mice per experimental group). *, $P < 0.05$; **, $P < 0.001$.

After allergen challenge, the expression of 1,049 genes was changed ≥ 2 -fold in WT mice (compared with saline-treated WT mice). In contrast, 608 transcripts were changed in allergen-challenged *Il13ra1*^{-/-} mice (compared with saline-treated *Il13ra1*^{-/-} mice) (Fig. 5A).

Comparison of allergen-challenged WT with *Il13ra1*^{-/-} mice identified a set of 205 IL-13Rα1-dependent genes (e.g., dysregulated ≥ 2 -fold between allergen-challenged WT and *Il13ra1*^{-/-} mice) (Fig. 5B). These genes were segregated into the following four clusters: cluster 1, up-regulated genes (in *Il13ra1*^{-/-} mice but to a lesser extent, i.e., less responsive); cluster 2, unaltered genes (in *Il13ra1*^{-/-} mice, i.e., nonresponsive); cluster 3, down-regulated genes (only in *Il13ra1*^{-/-} mice); and cluster 4, down-regulated genes in WT mice but unaltered in *Il13ra1*^{-/-} mice (Fig. 5B).

Among the less responsive genes, various CC chemokines, mucin-associated genes, and alternatively activated macrophage (aaMΦ) marker genes such as *Arg-1* (arginase 1) and *Chi3l3* (YM1) were identified. Interestingly, the expression of other aaMΦ markers, including *Retnla* (Relm-α) and *Mgl1* (macrophage galactose-type calcium-type lectin 1/CD301a), were independent of IL-13Rα1

(Table 1 and Table S1) (21, 22). Furthermore, numerous other genes including *Sppr2a*, *Corin*, and *Il13ra2* (Fig. 5C and Tables S2–S5) were unresponsive to induction by allergen challenge in *Il13ra1*^{-/-} mice.

In addition, we identified 367 genes that were IL-13Rα1-independent (i.e., similarly regulated in both OVA-challenged *Il13ra1*^{-/-} and WT mice). These included C-X-C chemokines such as CXCL9 and CXCL10, which are up-regulated ≈ 13 - and 8.5-fold, respectively, in both WT and *Il13ra1*^{-/-} mice (Table S1). Interestingly, 15-lipoxygenase (15-LO), a recently described IL-13-induced gene and a key enzyme in arachidonic acid metabolism, was also IL-13Rα1-independent (23).

Identification of IL-4-Induced IL-13Rα1-Dependent Genes. Next, we aimed to identify the relative contribution of the different IL-4R chains in IL-4-induced lung responses. By means of a global microarray approach, we compared the genetic signature of IL-4-treated WT mice with IL-4-treated *Il13ra1*^{-/-} mice (Fig. 5D). This comparison identified a set of 63 genes that were induced by IL-4 and dependent on IL-13Rα1 (e.g., dysregulated ≥ 2 -fold between WT and *Il13ra1*^{-/-} mice) (Fig. 5D). Interestingly, most genes associated with aaMΦ, such as *Chi3l3*, *Arg-1*, and *Retnla*, were independent of *Il13ra1* expression (Table 1). In contrast, IL-4-induced *Chia* (chitinase) expression was dependent on IL-13Rα1. Consistent with our findings, several mucus-associated genes such

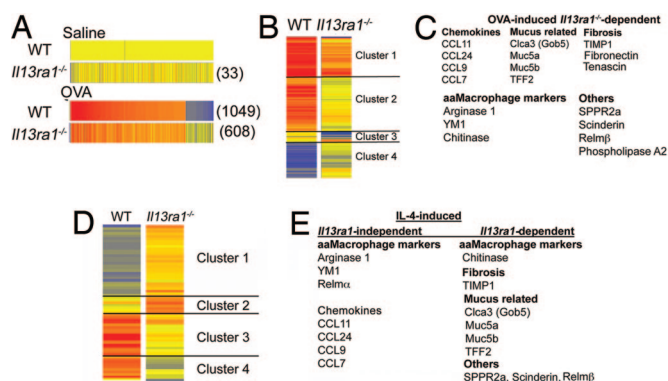


Fig. 5. Identification of IL-13Rα1-dependent genes. DNA microarray analysis of allergen-challenged lungs from *Il13ra1*^{-/-} or WT mice identifies 33 altered genes (shown in parentheses) at baseline in the *Il13ra1*^{-/-} mice, 1,049 altered genes in the allergen-challenged WT mice, and 608 altered genes in the allergen-challenged *Il13ra1*^{-/-} mice (in comparison with saline-treated mice) (A). Comparison of microarray data obtained from the lungs of allergen-challenged mice reveals a subset of 205 altered genes that are depicted in four clusters as shown (B). Several allergen-induced IL-13Rα1-dependent genes are shown (C). Comparison of microarray data obtained from IL-4-challenged mice reveals a subset of 63 altered genes that are dysregulated (D). Several IL-4-induced IL-13Rα1-dependent genes are depicted (E).

Table 1. Comparison of allergen- and IL-4-induced aaMΦ markers in WT and *Il13ra1*^{-/-} mice

Description	Gene symbol	OVA	IL-4
Scinderin	<i>Scin</i>	16.13	16.27
Resistin-like β	<i>Retnlb</i>	8.88	8.84
Chitinase, acidic	<i>Chia</i>	6.705	6.77
Similar to gel-forming mucin	<i>Muc5ac</i>	6.896	7.02
Small proline-rich protein 2A	<i>Sppr2a</i>	5.09	5.78
Intelectin a	<i>Itlna</i>	5.225	5.48
Calpain 9	<i>Capn9</i>	3.775	3.8
Solute carrier member 1	<i>Slc5a1</i>	5.856	5.98
Tissue inhibitor of metalloproteinase 1	<i>Timp1</i>	3.65	NC
Dentin matrix protein 1	<i>Dmp1</i>	5.205	NI
Corin	<i>Corin</i>	7.165	NI

Comparison of allergen (OVA)-induced and IL-4 induced gene expression between WT mice relative to their expression in *Il13ra1*^{-/-} mice. Values are expressed by increased fold change. NC, not changed; NI, not induced.

Table 2. Summary of the differential regulation of various pathological changes in the lung and their dependency on the type I or type II IL-4Rs

Stimuli	AR	Mucus	PF	CC chemokines	Eosinophilia	aaMac genes		
						<i>Arg</i>	<i>Retnla</i>	<i>Chia</i>
IL-13	Type II	Type II	Type II	Type II	Type II	Type II	Type II	Type II
OVA	Type II	Type II	Type II	Type II	Type I	Type II	Type I	Type II
IL-4	Type II	Type II	?	Type I	Type I	Type I	Type I	Type II

AR, airway resistance; PF, profibrogenic mediators; aaMac, alternatively activated macrophages.

as *Muc5ac*, *Clca3* (Gob5), and *Tff2* were dependent on *Il13ra1* (i.e., found on clusters 3 or 4) whereas CC chemokine induction (but not *Ccl2*) was largely independent of *Il13ra1* (Fig. 5E). Furthermore, other genes implicated in asthma pathogenesis such as *Scin* (scinderin), *Itlna* (intelectin), and *Sppr2a* were also dependent on IL-13R α 1 (Fig. 5E and Tables S6–S9).

Comparison of Allergen- and IL-4-Induced IL-13R α 1-Dependent Genes.

Because both allergen- and IL-4-induced airway resistance and mucus production were dependent on the type II IL-4R, we identified IL-13R α 1-dependent genes that were similarly regulated after IL-4 and OVA (Table S10). These genes include *Chia*, *Scin*, *Retnlb* (Relm- β), *Itlna*, and *Capn9* (Calpain 9). Although IL-13R α 1 commonly regulated several allergen- and IL-4-induced genes, our analysis revealed several pathways that were differentially regulated. Furthermore, by examining aaM Φ signature genes (10, 21), we identified a subset of genes that were dependent on IL-13R α 1 after allergen challenge (i.e., *Arg-1* and *Chia*) but not after IL-4 challenge (i.e., *Arg-1*, *MglI*, and *Retnla*) (Table 1).

Discussion

The pathological effects of IL-4 and IL-13 in Th2 immunity have been a focus of intense research in the last decade (1, 7, 17, 19). Even so, the receptor–ligand interactions responsible for the central roles of IL-4 and IL-13 remain to be elucidated. To fully dissect the molecular mechanisms that are regulated by IL-13R α 1 in the lung in response to allergen challenge and the relative contribution of this receptor to IL-13- and IL-4-induced pathology, we examined diverse Th2 responses in *Il13ra1*^{−/−} mice. We report that IL-13R α 1 regulates baseline IgE (independent of changes in IL-4). However, IgE responses to T cell-dependent antigens are IL-13R α 1-independent. Integrating the data obtained from the *in vivo* models with global microarray analysis of allergen- and IL-4-challenged lungs enabled us to conclude the following: (i) IL-13R α 1 is the chief receptor for IL-13 in the lung; (ii) airway resistance, mucus production, and profibrogenic mediator induction are nearly totally dependent on IL-13R α 1, which serves as a signaling molecule for both IL-4 and IL-13; (iii) IL-13 and IL-13R α 1 dependence of the CC chemokine response (especially eotaxin generation) predominantly reflects greater production of IL-13 than IL-4; (iv) IL-4 efficiently utilizes the type I IL-4R to induce inflammatory cell recruitment, even though IL-4 is present at lower levels than IL-13; and (v) aaM Φ induction (defined by the expression of their classic gene products) depends on both the type I and type II IL-4Rs (see Table 2). In addition, we demonstrate that key pathogenic molecules associated with asthma severity, such as chitinase (24), are entirely dependent on IL-13R α 1.

Our data demonstrate that baseline IgE expression depends on IL-13R α 1. Nevertheless, *Il13ra1*^{−/−} mice can still mount a normal Th2 cytokine and IgE response. Baseline natural, but not antigen-specific, IgE has been recently attributed to a unique population of ϵ -germ-line transcript-positive B2 cells (25). We propose that IL-13R α 1 may control natural IgE production by this subpopulation of B cells, which may express the IL-13R α 1 and type II IL-4R along with or instead of γ_c and the type I IL-4R.

Our studies also evaluated the relative roles of IL-13R α 1 and IL-13R α 2 in induction of TGF- β . IL-13 mediated TGF- β induction, and TGF- β production in liver fibrosis after *S. mansoni* infection has been proposed to be independent of IL-13R α 1 (10). Yet findings demonstrate that IL-13- and allergen-induced TGF- β production is completely dependent on IL-13R α 1. The finding that IL-13R α 1 is the key regulator of TGF- β production has therapeutic implications related to allergen-driven fibrotic reactions.

Importantly, mucus production and increased airway resistance were nearly completely dependent on IL-13R α 1. Similarly, whereas the chemokine response (except for CCL2) elicited by IL-4 was mostly IL-13R α 1-independent, all of the examined CC chemokines in the allergen-stimulated lung were IL-13R α 1-dependent. This dependency suggests that the CC chemokine response is regulated mostly by IL-13 and not IL-4, presumably because both the type I and type II IL-4Rs can induce CC chemokine production and more IL-13 is produced than IL-4. Despite this, and consistent with observations made with *stat6*^{−/−} and *Il13ra1*^{−/−} mice (10, 26), BALF eosinophilia was only modestly affected in allergen-challenged *Il13ra1*^{−/−} mice.

Because therapeutic targeting of IL-4R α substantially decreases eosinophilia in response to allergen challenge (27), an IL-13R α 1-independent pathway for eosinophil recruitment that is efficiently induced by IL-4 through the type I IL-4R must exist. Our *in vitro* chemotaxis studies support the existence of a CC chemokine-independent pathway for eosinophil recruitment under these conditions. Arachidonic acid metabolites produced in response to IL-4 such as induction of 15-LO may be responsible for CC chemokine-independent pulmonary eosinophilia. Although 15-LO can be induced by IL-13 (23), its induction is independent of IL-13R α 1 after IL-4 or allergen challenge. This suggests 15-LO production by a unique cell type compared with mucus- and eotaxin-producing cells and that these 15-LO-producing cells have more type I than type II IL-4R.

Incorporating global transcript expression analysis of the lungs of allergen- and IL-4-challenged *Il13ra1*^{−/−} mice provided an opportunity to dissect the contribution of IL-13R α 1 and the type II IL-4R to the asthma phenotype. Remarkably, the expression of *Chia*, a marker of aaM Φ development and a marker and causative molecule for asthma severity (10, 22, 24), was entirely dependent on IL-13R α 1. This result is of particular interest because *Arg-1*, a hallmark aaM Φ gene (21, 22), was independent of IL-13R α 1 after IL-4 administration but dependent on IL-13R α 1 after allergen challenge whereas other genes (i.e., *Retnla* and *MglI*) were entirely independent of IL-13R α 1. Thus, IL-4 may require both the type I and type II IL-4Rs to induce full development of aaM Φ in the lung. Furthermore, these data demonstrate that the precise phenotype of aaM Φ in the allergic lung depends on the stoichiometric relationship between IL-4 and IL-13. This could explain the different results regarding lung expression of arginase 1 in our study (in which IL-13 levels are greater than IL-4 levels) and that by Ramalingam *et al.* (10) (in which IL-4 levels were greater than IL-13 levels). Alternatively, it is possible that subsets of aaM Φ exist that produce either arginase 1 or chitinase or that the latter molecule is produced by other cells in the lung such as epithelial cells.

In addition, we identified a subset of genes that were induced by allergen challenge and IL-4 and were commonly regulated by IL-13R α 1 such as *Scin* (Scnderin), *Capn9* (Calpain 9), and solute carrier family member 1 (*Slc5a1*). These newly identified pathways may be important for regulating airway resistance and mucus production in the asthmatic lung.

In summary, our results establish that the critical role for IL-13R α 1 in asthma pathogenesis is mediated by its interactions with both IL-4 and IL-13. Furthermore, we dissociate mechanisms that stimulate cellular infiltration from those that induce airway resistance and goblet cell hyperplasia and emphasize IL-13R α 1 blockade as a potent target for the treatment of increased airway resistance, mucus production, and fibrosis in asthma. As such, these data highlight IL-13R α 1 as a dominant target for disrupting IL-13-, IL-4-, and allergen-mediated effects in the lung.

Materials and Methods

Measurement of the IL-13/Soluble IL-13R α 2 Complex. Total sIL-13R α 2 and serum levels of IL-13/sIL-13R were measured (28).

Serum IL-4 and IFN- γ Level Determination. Determination of serum IL-4 and IFN- γ levels were assessed by the *in vivo* cytokine capture assay (29).

Th2 Polarization. Goat anti-mouse IgD was injected i.p. (18), and serum IL-4, IFN- γ , and IgE levels were assessed (29).

Cytokine-Induced Airway Inflammation. Three doses (10 μ g per mouse) of IL-13 were administered intratracheally every other day for 4 days. A long-acting form of IL-4 produced by mixing recombinant mouse IL-4 (PeproTech) with a neutralizing mAb (BVD4-1D11) at a 2:1 molar ratio (IL-4C) was administered every other day for 7 days.

Allergen-Induced Airway Inflammation. Experimental asthma was induced as described (19). Twenty-four hours after the final challenge, the mice were anesthetized and the trachea was cannulated for airway resistance measurements. Subsequently, bronchoalveolar lavage was performed, and the lungs were excised for histological measurements.

Ig and Mediator Assessment. Serum Igs and BALF cytokines were measured with kits purchased from the following sources: IgA, IgM, IgG1, IgG2a, IgG2b, and IgG3

from Southern Biotech (lower detection limits: 7.8, 15.6, 31.2, 7.8, 7.8, and 15.6 pg/ml, respectively); IgE from BD Bioscience (lower detection limit: 15 pg/ml); and CCL11, CCL24, CCL2, CCL17, IL-4, IL-13, IL-5, IL-10, and active TGF- β from R & D Systems (lower detection limits: 15.6, 15.6, 3.9, 31.2, 6.25, 31.2, 15.6, 31.2, and 31.2 pg/ml, respectively).

Airway Resistance and Compliance Measurements. Airway resistance was measured by using the flexiVent system (Scireq Scientific Respiratory Equipment). Briefly, the mice were anesthetized, a tracheotomy was performed, and a cannula was inserted. A positive end-expiratory pressure of 0.2 kPa was established. Saline aerosol followed by β -methylcholine (Sigma-Aldrich; 25–100 mg/ml) established control baseline. Aerosols were generated with an ultrasonic nebulizer (DeVilbiss UltraNeb 2000) and delivered to the inspiratory line of the flexiVent. Each aerosol was delivered for 20 seconds during which time regular ventilation was maintained. Five measurements were made at 25-second intervals, and three peak responses were compared to the mean response of the saline aerosol.

Lung Histopathologic Changes. Hematoxylin and eosin or periodic acid Schiff (PAS) staining was performed (30).

Microarray Data Analysis. Whole-lung RNA was extracted by using TRIzol Reagent (Invitrogen Life Technologies). Microarray hybridization to mouse expression array (MOE430 2.1) was performed by the Affymetrix Gene Chip Core facility at Cincinnati Children's Hospital Medical Center (19).

Chemotaxis Assays. Chemotaxis was assessed by using eosinophils obtained from CD2-IL-5 transgenic mice as described (31). Cells (1.5×10^6) were either untreated or treated with anti-CCR3 or an isotype-matched antibody control (50 μ g/ml at 4°C for 30 min) (R & D Systems). Thereafter, the cells were washed and placed in the upper chamber, and 30% BALF (in HBSS) from WT or *Il13ra1*^{−/−} mice was placed in the lower chamber. After 3 h, total eosinophils in the lower chamber were assessed by using a hemacytometer.

Statistical Analysis. Data were analyzed by ANOVA followed by the Tukey post hoc test using GraphPad Prism 4. Data are presented as mean $\pm$ SD, and values of $P < 0.05$ were considered statistically significant.

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Supporting Information

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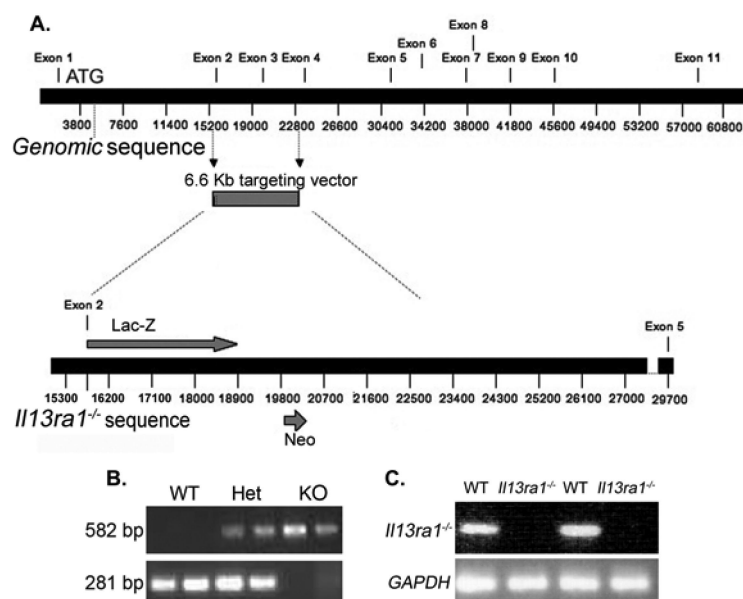


Fig. S1. Generation of *Il13ra1*^{-/-} mice. The IL-13R α 1 gene was replaced by a reporter-selection cassette, which consists of a β -galactosidase enzyme gene and a neomycin resistance gene. The knockout-reporter construct was created by means of bacterial homologous recombination into a bacterial artificial chromosome encoding IL-13R α 1 and was constructed so that the β -galactosidase gene was placed in frame with the ATG of IL-13R α 1 (Fig. 1A). The diagram shows the WT murine IL-13R α 1 gene locus and the gene-targeted locus. The construct deletes amino acids 15,824 through 22,414 of IL-13R α 1 contained in exons 2–4 of the gene. The mice were identified as heterozygotes and homozygotes by means of the TaqMan assay with probes for the Neo and LacZ genes and the IL-13R α 1 loss-of-allele probes. The mice were genotyped by means of PCR displaying a WT band of 281 bp and a targeted band of 582 bp (Fig. 1B). Het, heterozygote mice; KO, *Il13ra1*^{-/-} mice. Each lane represents a separate animal. The absence of mRNA transcript encoding for IL-13R α 1 was validated by PCR (Fig. 1C). Reverse transcription of whole-lung RNA was subjected to PCR using IL-13R α 1-specific primers and GAPDH control primers. Each lane represents a separate animal. Male *Il13ra1*^{-/-} mice were generated from BALB/c $\times$ C57BL/6 mice and were backcrossed onto the BALB/c and C57BL/6 strains (10 and seven generations, respectively). Littermate controls at the identical stage of backcrossing were used in all experiments and are referred to as WT.

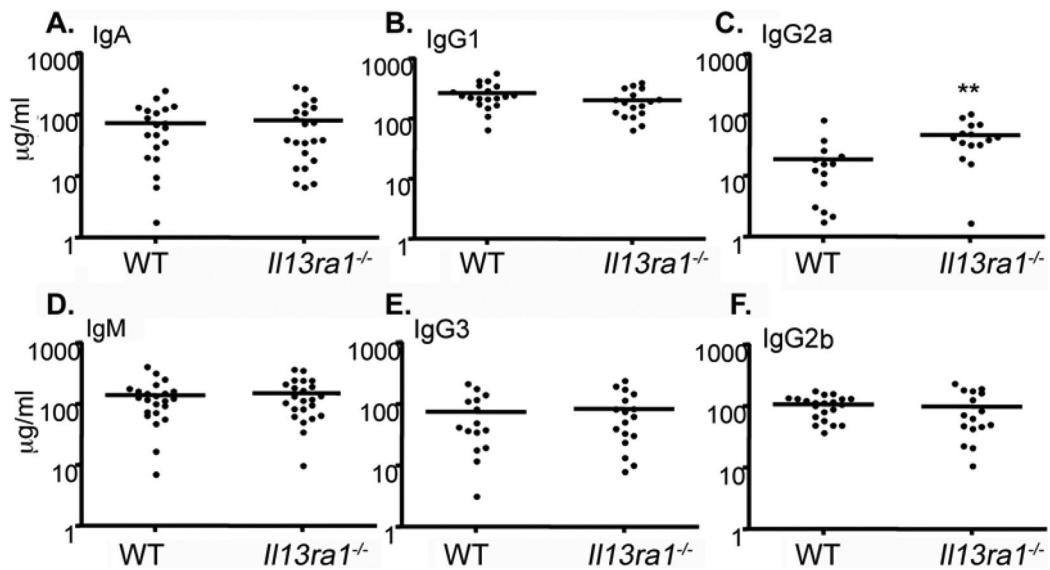
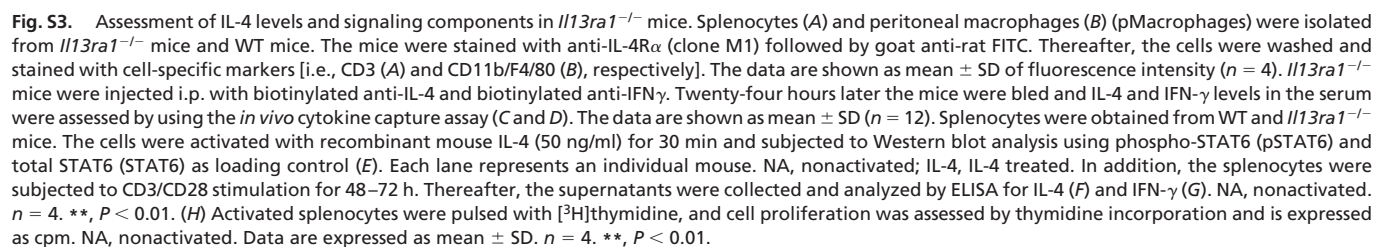


Fig. S2. Assessment of Ig levels in *Il13ra1*^{-/-} mice. Total serum Ig concentrations were determined in *Il13ra1*^{-/-} and WT mice using a standard ELISA (A–F). Each dot represents a different mouse; the horizontal line represents the mean. **, $P < 0.01$.



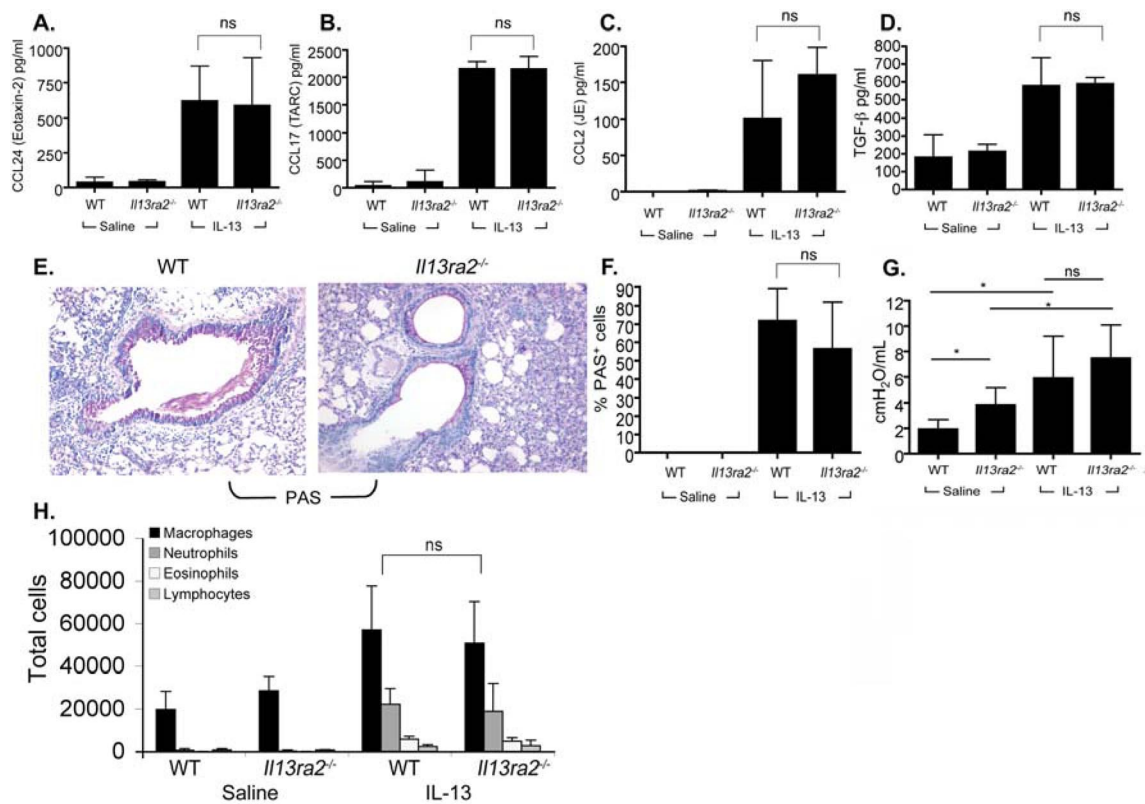


Fig. S4. Assessment of IL-13R α 2-mediated responses in a murine model of IL-13-induced airway inflammation. IL-13 was administered three times every other day to *Il13ra2*^{-/-} mice and WT control mice. Forty-eight hours after the final challenge, the mice were assessed for BALF chemokine levels (A–C), active TGF- β production (D), mucus production (as assessed by PAS staining) (E and F), airway resistance (G), and BALF cellular infiltration (H). The data are presented as mean $\pm$ SD ($n = 3$, eight to 10 mice per experimental group). ns, nonsignificant. *, $P < 0.05$.

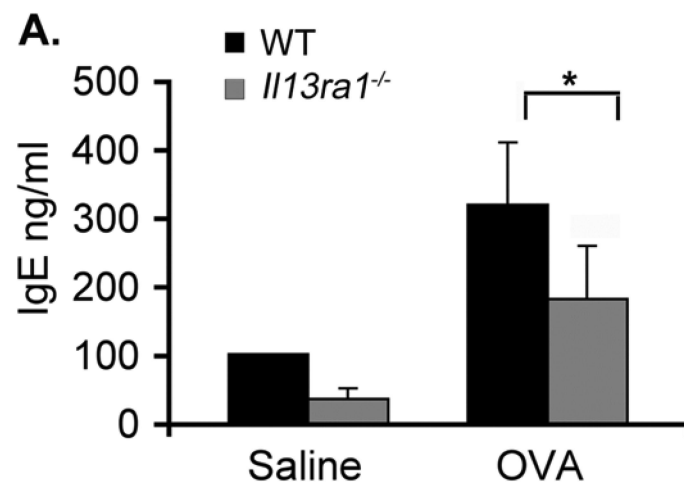


Fig. S5. Assessment of Th2 responses toward T cell-dependent antigens. After OVA/alum sensitization, serum was obtained from WT and *Il13ra1*^{-/-} mice and assessed for total IgE levels (A). The data are expressed as mean $\pm$ SD. $n = 3$ (four mice per experimental group). *, $P < 0.05$.

Table S1. OVA-induced *Il13ra1*-independent genes (i.e., induced similarly in WT and *Il13ra1*^{-/-} mice)

Affymetrix number/probe set	Gene description
AFFX-18SRNAMur/X00686_5.at	Mannosidase 2, alpha B2
1460423_x.at	Ig kappa chain
1460197_a.at	STEAP family member 4
1460187_at	secreted frizzled-related sequence protein 1
1460011_at	cytochrome P450, family 26, subfamily b, polypeptide 1
1459923_at	RIKEN cDNA B020003O03 gene
1458504_at	Zinc finger CCCH-type containing 12D
1458299_s.at	nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor, epsilon
1458148_at	RIKEN cDNA D230007K08 gene
1458007_at	myosin IB
1457823_at	cysteine rich protein 61
1457644_s.at	chemokine (C-X-C motif) ligand 1
1457296_at	cartilage intermediate layer protein, nucleotide pyrophosphohydrolase
1456632_at	B-cell CLL/lymphoma 11A (zinc finger protein)
1456442_at	RAB3A interacting protein (rab3)-like 1
1456402_at	RIKEN cDNA A330076H08 gene
1456328_at	B-cell scaffold protein with ankyrin repeats 1
1456280_at	Clasp homolog (Xenopus laevis)
1455834_x.at	transforming, acidic coiled-coil containing protein 3
1455332_x.at	Fc receptor, IgG, low affinity IIb
1455132_at	RIKEN cDNA A430107D22 gene
1455000_at	G protein-coupled receptor 68
1454744_at	RIKEN cDNA F630043A04 gene
1454694_a.at	topoisomerase (DNA) II alpha
1453472_a.at	SLAM family member 7
1453462_at	RIKEN cDNA 1110067M19 gene
1453416_at	growth arrest-specific 2 like 3
1453107_s.at	forkhead box M1; phosphatidylethanolamine binding protein
1452954_at	ubiquitin-conjugating enzyme E2C
1452917_at	replication factor C (activator 1) 5
1452912_at	RIKEN cDNA 2600005O03 gene
1452881_at	RIKEN cDNA 4833427B12 gene
1452598_at	RIKEN cDNA 2810418N01 gene
1452463_x.at	Immunoglobulin lambda chain, mAb 667
1452458_s.at	peptidylprolyl isomerase (cyclophilin) like 5
1452349_x.at	Interferon activated gene 205; myeloid cell nuclear differentiation antigen
1452314_at	kinesin family member 11
1452279_at	Properdin factor, complement
1452242_at	RIKEN cDNA 1200008O12 gene
1452087_at	epithelial stromal interaction 1 (breast)
1452040_a.at	cell division cycle associated 3
1451941_a.at	Fc receptor, IgG, low affinity IIb
1451905_a.at	myxovirus (influenza virus) resistance 1
1451780_at	B-cell linker
1451721_a.at	histocompatibility 2, class II antigen A, beta 1
1451716_at	v-maf musculoaponeurotic fibrosarcoma oncogene family, protein B (avian)
1451358_a.at	Rac GTPase-activating protein 1
1451246_s.at	Aurora kinase B
1451174_at	leucine rich repeat containing 33
1451128_s.at	kinesin family member 22
1451064_a.at	phosphoserine aminotransferase 1
1451054_at	orosomucoid 1
1450920_at	cyclin B2
1450912_at	membrane-spanning 4-domains, subfamily A, member 1
1450886_at	germ cell-specific gene 2
1450842_a.at	centromere autoantigen A
1450826_a.at	Serum amyloid A 3
1450652_at	cathepsin K
1450496_a.at	RIKEN cDNA 2810433K01 gene
1450355_a.at	capping protein (actin filament), gelsolin-like
1450297_at	interleukin 6
1450291_s.at	membrane-spanning 4-domains, subfamily A, member 4C
1450241_a.at	Ecotropic viral integration site 2a
1450234_at	membrane-spanning 4-domains, subfamily A, member 6C

Affymetrix number/probe set	Gene description
1450009_at	Lactotransferrin
1449864_at	interleukin 4
1449835_at	programmed cell death 1
1449705_x_at	
1449699_s_at	RIKEN cDNA C330027C09 gene
1449580_s_at	histocompatibility 2, class II, locus Mb1; histocompatibility 2, class II, locus Mb2
1449538_a_at	glucosaminyl (N-acetyl) transferase 1, core 2
1449452_a_at	glycoprotein 2 (zymogen granule membrane)
1449401_at	complement component 1, q subcomponent, gamma polypeptide
1449393_at	SH2 domain protein 1A
1449360_at	Colony stimulating factor 2 receptor, beta 2, low-affinity (granulocyte-macrophage)
1449269_at	coagulation factor V
1449227_at	cholesterol 25-hydroxylase
1449207_a_at	kinesin family member 20A
1449171_at	Ttk protein kinase
1449164_at	CD68 antigen
1449049_at	toll-like receptor 1
1449015_at	resistin like alpha
1448901_at	carboxypeptidase X 1 (M14 family)
1448899_s_at	RAD51 associated protein 1
1448883_at	Legumain
1448859_at	chemokine (C-X-C motif) ligand 13
1448777_at	minichromosome maintenance deficient 2 mitotin (<i>S. cerevisiae</i>)
1448627_s_at	PDZ binding kinase
1448466_at	cell division cycle associated 5
1448441_at	CDC28 protein kinase 1b
1448409_at	lymphoid-restricted membrane protein
1448380_at	lectin, galactoside-binding, soluble, 3 binding protein
1448314_at	cell division cycle 2 homolog A (<i>S. pombe</i>)
1448239_at	heme oxygenase (decycling) 1
1448226_at	ribonucleotide reductase M2
1448205_at	cyclin B1, related sequence 1; cyclin B1
1448123_s_at	transforming growth factor, beta induced
1448061_at	
1447891_at	
1447792_x_at	PREDICTED: similar to putative purinergic receptor FKSG79 [<i>Mus musculus</i>]
1447621_s_at	RIKEN cDNA 2610307O08 gene
1447363_s_at	budding uninhibited by benzimidazoles 1 homolog, beta (<i>S. cerevisiae</i>)
1447284_at	triggering receptor expressed on myeloid cells 1
1445365_at	membrane-spanning 4-domains, subfamily A, member 7
1444299_at	RIKEN cDNA A430093F15 gene
1444040_at	leukocyte-associated Ig-like receptor 1
1443687_x_at	
1443686_at	
1442544_at	immunoglobulin heavy chain 4 (serum IgG1)
1442118_at	Transcribed locus
1442082_at	complement component 3a receptor 1
1440275_at	Peptidylprolyl isomerase B, mRNA (cDNA clone MGC:6241 IMAGE:3483267)
1439695_a_at	M-phase phosphoprotein 1
1439595_at	TCR alpha C = T cell receptor alpha {Alternatively Spliced, C region}
1439566_at	RIKEN cDNA C030038J10 gene
1439510_at	shugoshin-like 1 (<i>S. pombe</i>)
1439436_x_at	inner centromere protein
1439377_x_at	cell division cycle 20 homolog (<i>S. cerevisiae</i>)
1439323_a_at	mitogen activated protein kinase kinase kinase 1
1439067_at	leukocyte-associated Ig-like receptor 1
1439040_at	Centromere protein E
1438434_at	Rho GTPase activating protein 11A
1438009_at	Histone 1, H2ae, mRNA (cDNA clone MGC:90847 IMAGE:5713252)
1437811_x_at	Coactosin-like 1 (<i>Dictyostelium</i>) (Cot1), mRNA
1437726_x_at	complement component 1, q subcomponent, beta polypeptide
1437716_x_at	kinesin family member 22
1437611_x_at	kinesin family member 2C
1437370_at	shugoshin-like 2 (<i>S. pombe</i>)
1437356_at	Epstein-Barr virus induced gene 2

Affymetrix number/probe set	Gene description
1437025_at	CD28 antigen
1436922_at	
1436847_s_at	cell division cycle associated 8
1436808_x_at	minichromosome maintenance deficient 5, cell division cycle 46 (<i>S. cerevisiae</i>)
1436723_at	FSH primary response 1
1436649_at	PREDICTED: zinc finger protein, subfamily 1A, 3 (<i>Aiolos</i>) [<i>Mus musculus</i>]
1436598_at	inducible T-cell co-stimulator
1436576_at	RIKEN cDNA A630077B13 gene
1436419_a_at	RIKEN cDNA 1700097N02 gene
1436312_at	zinc finger protein, subfamily 1A, 1 (<i>Ikaros</i>)
1436186_at	E2F transcription factor 8
1436003_at	vascular cell adhesion molecule 1
1435955_at	sialic acid binding Ig-like lectin 10
1435945_a_at	potassium intermediate/small conductance calcium-activated channel, subfamily N
1435773_at	RIKEN cDNA 4930547N16 gene
1435597_at	RIKEN cDNA C130052G03 gene
1435575_at	kinetochore associated 1
1435477_s_at	Fc receptor, IgG, low affinity IIb
1435476_a_at	Fc receptor, IgG, low affinity IIb
1435330_at	expressed sequence AI447904; cDNA sequence BC094916
1435306_a_at	kinesin family member 11
1435172_at	Eomesodermin homolog (<i>Xenopus laevis</i>)
1435114_at	WD repeat and HMG-box DNA binding protein 1
1435005_at	centromere protein E
1434955_at	membrane-associated ring finger (C3HC4) 1
1434850_at	IQ motif containing GTPase activating protein 3
1434767_at	expressed sequence C79407
1434748_at	cytoskeleton associated protein 2
1434695_at	denticleless homolog (<i>Drosophila</i>)
1434437_x_at	Ribonucleotide reductase M2
1434366_x_at	complement component 1, q subcomponent, beta polypeptide
1434295_at	RAS guanyl releasing protein 1
1434152_at	RIKEN cDNA 2210421G13 gene
1434068_s_at	expressed sequence AI662270
1434067_at	expressed sequence AI662270
1433963_a_at	cDNA sequence BC032204
1433893_s_at	Sperm associated antigen 5
1433892_at	Sperm associated antigen 5
1433862_at	extra spindle poles-like 1 (<i>S. cerevisiae</i>)
1432361_a_at	RIKEN cDNA 1700022C02 gene
1431705_a_at	mucolipin 2
1431609_a_at	Acid phosphatase 5, tartrate resistant
1431320_a_at	myosin Va
1430811_a_at	cell division cycle associated 1
1430802_at	histocompatibility 2, Q region locus 8
1430574_at	cyclin-dependent kinase inhibitor 3
1430570_at	kynureninase (L-kynurenine hydrolase)
1430523_s_at	immunoglobulin lambda chain, variable 1
1430447_a_at	leukocyte-associated Ig-like receptor 1
1430352_at	RIKEN cDNA 8430417A20 gene
1429954_at	C-type lectin domain family 4, member a3
1429947_a_at	Z-DNA binding protein 1
1429381_x_at	immunoglobulin heavy chain (J558 family)
1429295_s_at	thyroid hormone receptor interactor 13
1429172_a_at	RIKEN cDNA 5730507H05 gene
1429171_a_at	RIKEN cDNA 5730507H05 gene
1428947_at	RIKEN cDNA 2010001M09 gene
1428787_at	NCK associated protein 1 like
1428481_s_at	cell division cycle associated 8
1428480_at	cell division cycle associated 8
1428391_at	RAB3A interacting protein (rabin3)-like 1
1428304_at	establishment of cohesion 1 homolog 2 (<i>S. cerevisiae</i>)
1428136_at	secreted frizzled-related sequence protein 1
1428114_at	Solute carrier family 14 (urea transporter), member 1
1428111_at	Solute carrier family 38, member 4

Affymetrix number/probe set	Gene description
1428105_at	TPX2, microtubule-associated protein homolog (Xenopus laevis)
1428104_at	TPX2, microtubule-associated protein homolog (Xenopus laevis)
1427911_at	RIKEN cDNA 2610307O08 gene
1427892_at	myosin IG
1427870_x_at	immunoglobulin heavy chain 4 (serum IgG1)
1427860_at	IgG light chain gene, V region
1427820_at	Mus musculus, clone IMAGE:3983821
1427756_x_at	immunoglobulin heavy chain 4 (serum IgG1)
1427747_a_at	lipocalin 2
1427455_x_at	immunoglobulin kappa chain, constant region; immunoglobulin kappa chain variable 28 (V28); similar to anti-glycoprotein-B of human Cytomegalovirus immunoglobulin VI chain; similar to anti-PRSV coat protein monoclonal antibody PRSV-L 3-8 immunoglobulin light chain variable region
1427351_s_at	immunoglobulin heavy chain 6 (heavy chain of IgM)
1427329_a_at	immunoglobulin heavy chain 6 (heavy chain of IgM)
1427161_at	centromere autoantigen F
1427094_at	polymerase (DNA directed), epsilon 2 (p59 subunit)
1426936_at	hypothetical LOC433593
1426817_at	antigen identified by monoclonal antibody K _i 67
1426652_at	minichromosome maintenance deficient 3 (<i>S. cerevisiae</i>)
1426183_a_at	CD209d antigen
1426169_a_at	linker for activation of T cells family, member 2
1426168_a_at	T-cell receptor alpha chain; RIKEN cDNA A430107P09 gene
1426112_a_at	CD72 antigen
1425815_a_at	hyaluronan mediated motility receptor (RHAMM)
1425753_a_at	uracil DNA glycosylase
1425477_x_at	histocompatibility 2, class II antigen A, beta 1
1425406_at	C-type lectin domain family 4, member a2
1425385_a_at	immunoglobulin heavy chain 1a (serum IgG2a)
1425324_x_at	immunoglobulin heavy chain 4 (serum IgG1)
1425294_at	SLAM family member 8
1425247_a_at	immunoglobulin heavy chain 4 (serum IgG1)
1425214_at	pyrimidinergic receptor P2Y, G-protein coupled, 6
1425145_at	interleukin 1 receptor-like 1
1425086_a_at	SLAM family member 6
1425025_at	RIKEN cDNA 0610008L10 gene
1424998_at	EGF-like module containing, mucin-like, hormone receptor-like sequence 4
1424971_at	RIKEN cDNA 2600001J17 gene
1424965_at	Leupaxin
1424931_s_at	immunoglobulin lambda chain, variable 1
1424775_at	2'-5' oligoadenylate synthetase 1A
1424754_at	membrane-spanning 4-domains, subfamily A, member 7
1424727_at	chemokine (C-C motif) receptor 5
1424629_at	Breast cancer 1
1424542_at	S100 calcium binding protein A4
1424511_at	Aurora kinase A
1424509_at	CD177 antigen
1424305_at	immunoglobulin joining chain
1424292_at	DEP domain containing 1a
1424278_a_at	baculoviral IAP repeat-containing 5
1424208_at	prostaglandin E receptor 4 (subtype EP4)
1424144_at	retroviral integration site 2
1424143_a_at	retroviral integration site 2
1424128_x_at	Aurora kinase B
1424118_a_at	spindle pole body component 25 homolog (<i>S. cerevisiae</i>)
1424046_at	budding uninhibited by benzimidazoles 1 homolog (<i>S. cerevisiae</i>)
1423954_at	complement component 3
1423847_at	RIKEN cDNA 2810406C15 gene
1423809_at	transcription factor 19
1423775_s_at	protein regulator of cytokinesis 1
1423774_a_at	protein regulator of cytokinesis 1
1423524_at	microtubule associated serine/threonine kinase-like
1423463_a_at	DNA segment, Chr 2, ERATO Doi 750, expressed
1423182_at	Tumor necrosis factor receptor superfamily, member 13b
1423092_at	inner centromere protein

Affymetrix number/probe set	Gene description
1422957_at	chemokine (C-C motif) receptor 3
1422932_a.at	vav 1 oncogene
1422903_at	lymphocyte antigen 86
1422814_at	asp (abnormal spindle)-like, microcephaly associated (Drosophila)
1422812_at	chemokine (C-X-C motif) receptor 6
1422535_at	cyclin E2
1422476_at	interferon gamma inducible protein 30
1422462_at	ubiquitin-conjugating enzyme E2T (putative)
1422460_at	MAD2 (mitotic arrest deficient, homolog)-like 1 (yeast)
1422430_at	fidgetin-like 1
1422260_x.at	chemokine (C-C motif) receptor 5
1422259_a.at	chemokine (C-C motif) receptor 5
1422046_at	integrin alpha M
1422016_a.at	centromere autoantigen H
1422013_at	C-type lectin domain family 4, member a2
1421813_a.at	Prosaposin
1421731_a.at	flap structure specific endonuclease 1
1421546_a.at	Rac GTPase-activating protein 1
1421307_at	carbonic anhydrase 13
1421211_a.at	class II transactivator
1421188_at	chemokine (C-C motif) receptor 2
1421187_at	chemokine (C-C motif) receptor 2
1421186_at	chemokine (C-C motif) receptor 2
1421173_at	interferon regulatory factor 4
1421073_a.at	prostaglandin E receptor 4 (subtype EP4)
1421038_a.at	potassium intermediate/small conductance calcium-activated channel, subfamily N, member 4
1420664_s.at	protein C receptor, endothelial
1420380_at	chemokine (C-C motif) ligand 2
1420338_at	arachidonate 15-lipoxygenase
1420081_s.at	DNA segment, Chr 2, ERATO Doi 750, expressed
1420028_s.at	minichromosome maintenance deficient 3 (<i>S. cerevisiae</i>)
1419943_s.at	cyclin B1
1419754_at	myosin Va
1419728_at	chemokine (C-X-C motif) ligand 5
1419684_at	chemokine (C-C motif) ligand 8
1419605_at	macrophage galactose N-acetyl-galactosamine specific lectin 1
1419561_at	chemokine (C-C motif) ligand 3
1419483_at	complement component 3a receptor 1
1419482_at	complement component 3a receptor 1
1419334_at	cytotoxic T-lymphocyte-associated protein 4
1419282_at	chemokine (C-C motif) ligand 12
1419254_at	methylenetetrahydrofolate dehydrogenase (NAD + dependent)
1419219_at	cytochrome P450, family 4, subfamily f, polypeptide 18
1419209_at	chemokine (C-X-C motif) ligand 1
1419202_at	cystatin F (leukocystatin)
1419153_at	RIKEN cDNA 2810417H13 gene
1419152_at	RIKEN cDNA 2810417H13 gene
1419128_at	integrin alpha X
1419100_at	Serine (or cysteine) peptidase inhibitor, clade A, member 3N
1419082_at	Serine (or cysteine) peptidase inhibitor, clade B, member 2
1419043_a.at	interferon inducible GTPase 1
1419042_at	interferon inducible GTPase 1
1418998_at	kynurenine 3-monooxygenase (kynurenine 3-hydroxylase)
1418930_at	chemokine (C-X-C motif) ligand 10
1418907_at	coagulation factor V
1418666_at	pentraxin related gene
1418652_at	chemokine (C-X-C motif) ligand 9
1418641_at	lymphocyte cytosolic protein 2
1418353_at	CD5 antigen
1418281_at	RAD51 homolog (<i>S. cerevisiae</i>)
1418264_at	SoxLZ/Sox6 leucine zipper binding protein in testis
1418204_s.at	allograft inflammatory factor 1
1418026_at	exonuclease 1
1417939_at	RAD51 associated protein 1

Affymetrix number/probe set	Gene description
1417938_at	RAD51 associated protein 1
1417929_at	Solute carrier family 7 (cationic amino acid transporter, y + system), member 8
1417928_at	PDZ and LIM domain 4
1417926_at	leucine zipper protein 5
1417911_at	cyclin A2
1417910_at	cyclin A2
1417876_at	Fc receptor, IgG, high affinity I
1417870_x_at	cathepsin Z
1417869_s_at	cathepsin Z
1417851_at	chemokine (C-X-C motif) ligand 13
1417822_at	DNA segment, Chr 17, human D6S56E 5
1417821_at	DNA segment, Chr 17, human D6S56E 5
1417676_a_at	protein tyrosine phosphatase, receptor type, O
1417601_at	regulator of G-protein signaling 1
1417541_at	helicase, lymphoid specific
1417450_a_at	transforming, acidic coiled-coil containing protein 3
1417445_at	kinetochore associated 2
1417381_at	complement component 1, q subcomponent, alpha polypeptide
1417346_at	PYD and CARD domain containing
1417314_at	histocompatibility 2, complement component factor B
1417244_a_at	interferon regulatory factor 7
1417063_at	complement component 1, q subcomponent, beta polypeptide
1417019_a_at	cell division cycle 6 homolog (<i>S. cerevisiae</i>)
1416957_at	POU domain, class 2, associating factor 1
1416956_at	potassium voltage-gated channel, shaker-related subfamily, beta member 2
1416827_at	thromboxane A synthase 1, platelet
1416802_a_at	cell division cycle associated 5
1416757_at	Zwisch, kinetochore associated, homolog (<i>Drosophila</i>)
1416664_at	cell division cycle 20 homolog (<i>S. cerevisiae</i>)
1416641_at	ligase I, DNA, ATP-dependent
1416575_at	cell division cycle 45 homolog (<i>S. cerevisiae</i>)-like
1416558_at	maternal embryonic leucine zipper kinase
1416514_a_at	Fascin homolog 1, actin bundling protein(<i>Strongylocentrotus purpuratus</i>)
1416333_at	docking protein 2
1416309_at	nucleolar and spindle associated protein 1
1416299_at	Shc SH2-domain binding protein 1
1416258_at	thymidine kinase 1
1416251_at	minichromosome maintenance deficient 6 (MIS5 homolog, <i>S. pombe</i>) (<i>S. cerevisiae</i>)
1416120_at	ribonucleotide reductase M2
1416076_at	cyclin B1, related sequence 1; cyclin B1
1416030_a_at	minichromosome maintenance deficient 7 (<i>S. cerevisiae</i>)
1416016_at	transporter 1, ATP-binding cassette, sub-family B (MDR/TAP)
1416002_x_at	coactosin-like 1 (<i>Dictyostelium</i>)
1415945_at	minichromosome maintenance deficient 5, cell division cycle 46 (<i>S. cerevisiae</i>)
1415871_at	transforming growth factor, beta induced
1415811_at	ubiquitin-like, containing PHD and RING finger domains, 1
1415810_at	ubiquitin-like, containing PHD and RING finger domains, 1;

The genes are listed by order of fold change.

Table S2. Cluster 1: Up-regulated in *Il13ra1*^{-/-} to a lesser extent (i.e., down-regulated genes)

Affymetrix number/probe set	Gene description
1432329_a.at	megakaryocyte-associated tyrosine kinase
1416306_at	chloride channel calcium activated 3
1425295_at	eosinophil-associated, ribonuclease A family, member 11
1448872_at	regenerating islet-derived 3 gamma
1423719_at	cDNA sequence U46068
1439423_x.at	cDNA sequence U46068
1422448_at	trefoil factor 2 (spasmolytic protein 1)
1426911_at	desmocollin 2
1427626_at	mucin 5, subtype B, tracheobronchial
1450488_at	chemokine (C-C motif) ligand 24
1418993_s.at	coagulation factor X
1440173_x.at	Selectin, platelet (Selp), mRNA
1426008_a.at	solute carrier family 7 (cationic amino acid transporter, y + system), member 2
1428034_a.at	tumor necrosis factor receptor superfamily, member 9
1417795_at	cell adhesion molecule with homology to L1CAM
1417256_at	matrix metalloproteinase 13
1450290_at	programmed cell death 1 ligand 2
1452014_a.at	insulin-like growth factor 1
1436223_at	PREDICTED: integrin beta 8 [Mus musculus], mRNA sequence
1419519_at	insulin-like growth factor 1
1453076_at	RIKEN cDNA 9130211I03 gene
1425386_at	RIKEN cDNA 4833422F24 gene
1437244_at	PREDICTED: similar to growth arrest-specific 2 like 3 [Mus musculus], mRNA sequence
1451289_at	double cortin and calcium/calmodulin-dependent protein kinase-like 1
1424271_at	double cortin and calcium/calmodulin-dependent protein kinase-like 1
1419549_at	arginase 1, liver
1420558_at	selectin, platelet
1448898_at	chemokine (C-C motif) ligand 9
1449305_at	coagulation factor X
1417936_at	chemokine (C-C motif) ligand 9
1421075_s.at	cytochrome P450, family 7, subfamily b, polypeptide 1
1434046_at	expressed sequence AA467197
1460227_at	tissue inhibitor of metalloproteinase 1
1451798_at	interleukin 1 receptor antagonist
1420582_at	Cd209e antigen
1435409_at	Transcribed locus
1438148_at	gene model 1960, (NCBI)
1418992_at	coagulation factor X
1425663_at	interleukin 1 receptor antagonist
1427221_at	X transporter protein 3 similar 1 gene
1450060_at	polymeric immunoglobulin receptor
1449906_at	selectin, platelet
1421921_at	serine (or cysteine) peptidase inhibitor, clade A, member 3M
1455490_at	upstream binding transcription factor, RNA polymerase I
1421977_at	matrix metalloproteinase 19
1449153_at	matrix metalloproteinase 12
1421228_at	chemokine (C-C motif) ligand 7
1423569_at	glycine amidinotransferase (L-arginine:glycine amidinotransferase)
1419599_s.at	membrane-spanning 4-domains, subfamily A, member 11
1435190_at	cell adhesion molecule with homology to L1CAM
1424923_at	serine (or cysteine) peptidase inhibitor, clade A, member 3G
1419192_at	interleukin 4 induced 1
1422873_at	proteoglycan 2, bone marrow
1452794_x.at	spermatogenesis associated glutamate (E)-rich protein 1
1421074_at	cytochrome P450, family 7, subfamily b, polypeptide 1
1419598_at	membrane-spanning 4-domains, subfamily A, member 6D
1455980_a.at	PREDICTED: similar to growth arrest-specific 2 like 3 [Mus musculus], mRNA sequence
1425450_at	chitinase 3-like 4

The genes are listed by order of fold change.

Affymetrix number/probe set	Gene description
1438665_at	sphingomyelin phosphodiesterase 3, neutral
1455531_at	RIKEN cDNA A930031D07 gene
1449285_at	cystatin 9
1416286_at	regulator of G-protein signaling 4
1422340_a.at	Actin, gamma 2, smooth muscle, enteric
1449896_at	Melanophilin
1448285_at	regulator of G-protein signaling 4
1427511_at	Beta-2 microglobulin (B2m), mRNA
1452250_a.at	procollagen, type VI, alpha 2
1460604_at	Cytochrome b reductase 1, mRNA
1441818_at	Transcribed locus
1426872_at	Fc fragment of IgG binding protein
1431900_a.at	forkhead box A3
1450276_a.at	Scinderin
1430899_at	similar to secreted gel-forming mucin
1418207_at	FXYP domain-containing ion transport regulator 4
1448789_at	aldehyde dehydrogenase family 1, subfamily A3
1418547_at	tissue factor pathway inhibitor 2
1455770_at	Tryptophan 2,3-dioxygenase, mRNA (cDNA clone MGC:25811 IMAGE:4159877)
1417789_at	small chemokine (C-C motif) ligand 11
1419017_at	Corin
1459889_at	chloride channel calcium activated 3
1443408_at	Polo-like kinase 1 (Drosophila)
1422177_at	Interleukin 13 receptor, alpha 2
1418368_at	resistin like beta
1419093_at	tryptophan 2,3-dioxygenase
1425122_at	Open reading frame 9
1450618_a.at	small proline-rich protein 2A
1422876_at	calpain 9 (nCL-4)
1439016_x.at	small proline-rich protein 2A
1416200_at	RIKEN cDNA 9230117N10 gene
1424187_at	RIKEN cDNA 2610001E17 gene
1416456_a.at	chitinase, acidic
1421564_at	serine (or cysteine) peptidase inhibitor, clade A, member 3C
1455802_x.at	anterior gradient 2 (Xenopus laevis)
1458484_at	RIKEN cDNA A730020M07 gene
1443746_x.at	dentin matrix protein 1
1449028_at	ras homolog gene family, member U
1443745_s.at	dentin matrix protein 1
1417642_at	aldehyde dehydrogenase family 1, subfamily A3
1419725_at	solute carrier family 26, member 4
1451416_a.at	transglutaminase 1, K polypeptide
1419268_at	anterior gradient 2 (Xenopus laevis)
1419413_at	chemokine (C-C motif) ligand 17
1419057_at	solute carrier family 5 (sodium/glucose cotransporter), member
11433600_at	
1450139_at	endoplasmic reticulum (ER) to nucleus signalling 2
1455431_at	solute carrier family 5 (sodium/glucose cotransporter), member 1
1440681_at	Adult male cortex cDNA, RIKEN full-length enriched library,
1436712_at	phospholipase A2, group IVC (cytosolic, calcium-independent)
1418165_at	intelectin a
1420413_at	solute carrier family 7 (cationic amino acid transporter, y + system), member 1
1434583_at	transmembrane protein 26
1418724_at	complement component factor i
1445626_at	Lectin, galactose binding, soluble 3 (Lgals3), mRNA
1416342_at	tenascin C
1448470_at	fructose biphosphatase 1
1418649_at	EGL nine homolog 3 (C. elegans)
1438467_at	macrophage galactose N-acetyl-galactosamine specific lectin 2
1450616_at	eosinophil-associated, ribonuclease A family, member 5
1431970_at	transmembrane 7 superfamily member 4
1445642_at	LEM domain containing 1
1422648_at	solute carrier family 7 (cationic amino acid transporter, y + system), member 2
1457483_at	Modulator recognition factor 2 (Mrf2)

Affymetrix number/probe set	Gene description
1437218_at	fibronectin 1
1458781_at	Potassium channel, subfamily K, member 13 (Kcnk13), mRNA
1443536_at	solute carrier family 7 (cationic amino acid transporter, y + system), member 11
1441836_x_at	PREDICTED: hypothetical protein LOC74174 [Mus musculus], mRNA sequence
1449451_at	serine (or cysteine) peptidase inhibitor, clade B (ovalbumin), member 11
1416125_at	FK506 binding protein 5
1428195_at	RIKEN cDNA 4631427C17 gene
1450185_a_at	Potassium inwardly-rectifying channel, subfamily J, member 15
1419610_at	chemokine (C-C motif) receptor 1
1435354_at	Potassium inwardly-rectifying channel, subfamily J, member 15
1423523_at	aminoadipate-semialdehyde synthase
1442025_a_at	
1419874_x_at	zinc finger and BTB domain containing 16
1442026_at	
1439163_at	PREDICTED: zinc finger and BTB domain containing 16 [Mus musculus], mRNA sequence

The genes are listed by order of fold change.

Table S4. Cluster 3: Altered only in *Il13ra1*^{-/-} mice

Affymetrix number/probe set	Gene description
1427381_at	immunoresponsive gene 1
1455318_at	T-cell immunoglobulin and mucin domain containing 4
1419532_at	Interleukin 1 receptor, type II
1447227_at	Solute carrier family 40 (iron-regulated transporter), member 1
1440104_at	RAN binding protein 2
1458089_at	FK506 binding protein 5, mRNA (cDNA clone MGC:18417 IMAGE:4237766)
1441799_at	FK506 binding protein 5, mRNA (cDNA clone MGC:18417 IMAGE:4237766)
1422892_s_at	histocompatibility 2, class II antigen E alpha
1418438_at	fatty acid binding protein 2, intestinal
1417065_at	early growth response 1
1431577_at	
1420357_s_at	X-linked lymphocyte-regulated 3A; X-linked lymphocyte-regulated 3B
1434449_at	Aquaporin 4

The genes are listed by order of fold change.

Table S5. Cluster 4: Up-regulated genes (i.e., down-regulated only in WT mice)

Affymetrix number/probe set	Gene description
1449280_at	endothelial cell-specific molecule 1
1424479_at	cystatin 8 (cystatin-related epididymal spermatogenic)
1422168_a.at	brain derived neurotrophic factor
1448785_at	CBFA2T1 identified gene homolog (human)
1433836_a.at	RIKEN cDNA 8430408G22 gene
1433837_at	RIKEN cDNA 8430408G22 gene
1426433_at	myc target 1
1448973_at	sulfotransferase family 1D, member 1
1418138_at	sulfotransferase family 1D, member 1
1436528_at	Kazal-type serine peptidase inhibitor domain 1
1416040_at	Lipase, gastric
1444480_at	Protein kinase, AMP-activated, gamma 3 non-catalytic subunit (Prkag3)
1416505_at	nuclear receptor subfamily 4, group A, member 1
1452260_at	cell death-inducing DFFA-like effector c
1455392_at	RIKEN cDNA 9630019K15 gene
1429953_at	RIKEN cDNA 2210011C24 gene
1416077_at	Adrenomedullin
1449498_at	macrophage receptor with collagenous structure

The genes are listed by order of fold change.

Affymetrix number/probe set	Gene name	Gene description
1456062_at	Nppa	natriuretic peptide precursor type A
1418373_at	Pgam2	phosphoglycerate mutase 2
1427735_a.at	Acta1	Actin, alpha 1, skeletal muscle
1449218_at	Cox8b	cytochrome c oxidase, subunit VIIIb
1452651_a.at	Myl1	myosin, light polypeptide 1
1436867_at	Srl	Sarcalumenin
1418951_at	Txlnb	taxilin beta
1448327_at	Actn2	actinin alpha 2
1455374_at		
1450952_at	Pln	Phospholamban
1417889_at	Apobec2	apolipoprotein B editing complex 2
1420347_at	Plunc	palate, lung, and nasal epithelium carcinoma associated
1441111_at		15 days embryo embryonic body below diaphragm cDNA, RIKEN full-length enriched library, clone:8230401F05 product:unclassifiable, full insert sequence
1451203_at	Mb	Myoglobin
1428722_at	Ckmt2	creatine kinase, mitochondrial 2
1426615_s.at	Ndrp4	N-myc downstream regulated gene 4
1422536_at	Tnni3	troponin I, cardiac
1420884_at	Sln	Sarcolipin
1427446_s.at	Ttn	Titin
1437482_at	Srd5a2l2	steroid 5 alpha-reductase 2-like 2
1450123_at	Ryr2	ryanodine receptor 2, cardiac
1427445_a.at	Ttn	Titin
1453351_at	Tbx20	T-box 20
1418370_at	Tnni3	troponin C, cardiac/slow skeletal
1460332_at	Pln	Phospholamban
1420465_s.at	Mup1; Mup2	major urinary protein 1; major urinary protein 2
1422973_a.at	Thrsp	thyroid hormone responsive SPOT14 homolog (Rattus)

The genes are listed by order of fold change.

Table S7. Cluster 2: Up-regulated genes

Affymetrix number/probe set	Gene name	Gene description
1431642_at	Eif2 s3y	Eukaryotic translation initiation factor 2, subunit 3, structural gene Y-linked
1441799_at	Fkbp5	FK506 binding protein 5, mRNA (cDNA clone MGC:18417 IMAGE:4237766)
1417464_at	Tnnc2	troponin C2, fast
1427868_x_at	Myh1	myosin, heavy polypeptide 1, skeletal muscle, adult
1427520_a_at	Myh1	myosin, heavy polypeptide 1, skeletal muscle, adult
1450060_at	Pigr	Polymeric immunoglobulin receptor

The genes are listed by order of fold change.

Table S8. Cluster 3: Unaltered genes

Affymetrix number/probe set	Gene name	Gene description
1419268_at	Agr2	anterior gradient 2 (Xenopus laevis)
1450139_at	Ern2	endoplasmic reticulum (ER) to nucleus signalling 2
1426872_at	Fcgbp	Fc fragment of IgG binding protein
1439016_x_at	Sprr2a	small proline-rich protein 2A
1440409_at	2210401J11Rik	RIKEN cDNA 2210401J11 gene
1450276_a_at	Scin	Scinderin
1459889_at	Clca3	chloride channel calcium activated 3
1441818_at		Transcribed locus
1422876_at	Capn9	calpain 9 (nCL-4)
1418165_at	Itlna	intelectin a
1430899_at	LOC546020	similar to secreted gel-forming mucin
1450618_a_at	Sprr2a	small proline-rich protein 2A
1433600_at		
1422448_at	Tff2	trefoil factor 2 (spasmolytic protein 1)

The genes are listed by order of fold change.

Table S9. Cluster 4: Down-regulated genes

Affymetrix number/probe set	Gene name	Gene description
1422448_at	Tff2	trefoil factor 2 (spasmolytic protein 1) Erythroid differentiation regulator 1, mRNA (cDNA clone MGC:69587 IMAGE:6820436)
1439200_x_at		
1451775_s_at	Il13ra1	interleukin 13 receptor, alpha 1
1427164_at	Il13ra1	interleukin 13 receptor, alpha 1
1454783_at	Il13ra1	interleukin 13 receptor, alpha 1
1427165_at	Il13ra1	interleukin 13 receptor, alpha 1
1455431_at	Slc5a1	solute carrier family 5 (sodium/glucose cotransporter) member 1
1438239_at	Mid1	midline 1
1434292_at	E130013N09Rik	RIKEN cDNA E130013N09 gene
1416456_a_at	Chia	chitinase, acidic
1435761_at	Stfa3	stefin A3

The genes are listed by order of fold change.

OVA		IL-4	
<i>Il13ra1</i> -dependent	<i>Il13ra1</i> -independent	<i>Il13ra1</i> -dependent	<i>Il13ra1</i> -independent
<i>Chi3l3</i> *	<i>Retnla</i>	<i>Chia</i>	<i>Mgl1</i>
<i>Arg1</i>	<i>Mgl1</i>		<i>Arg1</i>
<i>Chia</i>			<i>Chi3l3</i>
			<i>Retnla</i>

* Not induced by OVA challenge.