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T helper 2 (Th2) cell differentiation, type 2 innate lymphoid cell (ILC2) development and regulation of interleukin-4 (IL-4) and IL-13 production

Jinfang Zhu

Molecular and Cellular Immunoregulation Unit, Laboratory of Immunology, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, MD 20892, USA

Abstract

Interleukin-4 (IL-4), IL-5 and IL-13, the signature cytokines that are produced during type 2 immune responses, are critical for protective immunity against infections of extracellular parasites and are responsible for asthma and many other allergic inflammatory diseases. Although many immune cell types within the myeloid lineage compartment including basophils, eosinophils and mast cells are capable of producing at least one of these cytokines, the production of these “type 2 immune response-related” cytokines by lymphoid lineages, CD4 T helper 2 (Th2) cells and type 2 innate lymphoid cells (ILC2s) in particular, are the central events during type 2 immune responses. In this review, I will focus on the signaling pathways and key molecules that determine the differentiation of naïve CD4 T cells into Th2 cells, and how the expression of Th2 cytokines, especially IL-4 and IL-13, is regulated in Th2 cells. The similarities and differences in the differentiation of Th2 cells, IL-4-producing T follicular helper (Tfh) cells and ILC2s as well as their relationships will also be discussed.

Keywords

Type 2 T helper cells (Th2); Type 2 innate lymphoid cells (ILC2); Interleukin-4; Interleukin-13; Transcriptional regulation; GATA3

1. Introduction

CD4 T helper (Th) cells are the key players during adaptive immune responses[1, 2]. Almost thirty years ago, it was first recognized by Mossman and Coffman that there are different subsets of Th cells, namely type 1 T helper (Th1) and type 2 T helper (Th2) cells, which are critically involved in distinct types of immune responses[3–5]. With the discovery of a third

Corresponding author at: 10 Center Drive, Building 10, Room 11D02, Bethesda, MD 20892-1892, USA. Tel: (+1) 301-402-6662, jfzhu@niaid.nih.gov.

Declaration of Interest

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major CD4 T effector cell lineage, Th17 cells that produce interleukin-17 (IL-17), and a regulatory T cell subset (induced regulatory T (iTreg) or peripherally induced regulatory T (pTreg)) that can be differentiated from naive CD4 T cells outside of the thymus, either in vitro or in vivo, the members of CD4 T cell family have expanded[2, 6–9].

Th2 cells are the crucial lymphocytes during adaptive immune responses to the infections of extracellular parasites such as helminths; Th2 cells are also responsible for development of some asthmatic and allergic inflammatory diseases. By secreting IL-4, Th2 cells instruct B cells to produce IgG1 and IgE[10]; by producing IL-4 and IL-13, Th2 cells induce alternatively activated macrophage[11]. Th2 cells also recruit eosinophils via IL-5 production[12] and directly act on epithelial cells and smooth muscle cells through IL-13 production[13–15]. Therefore, IL-4, IL-5 and IL-13 are the major effector cytokines produced by Th2 cells during type 2 immune responses. All three cytokine genes, *Il4*, *Il5* and *Il13*, are located within a genomic segment, which is subjected to the regulation of locus control region (LCR) in the *Rad50* gene[16, 17].

During CD4 T cell activation through T cell receptor (TCR)-mediated signaling and co-stimulation, cytokine signals received by the activated T cells are deterministic in T cell fate commitment. For example, together with TCR ligation, IL-4-mediated activation of the signal transducer and activator of transcription 6 (STAT6) plays an important role during Th2 cell differentiation [18–22], although IL-4-independent Th2 cell differentiation may occur in the absence of IL-4 signaling under certain conditions in vivo [23–27]. Both IL-4-dependent and IL-4-independent Th2 differentiation requires the key transcription factor GATA-3 (Figure 1), which is responsible for epigenetic changes in many Th2-specific gene loci and for direct transcription activation[28–31]. In addition, IL-2-mediated activation of STAT5 is indispensable for the production of Th2 cytokines possibly through chromatin remodeling of the Th2 cytokine locus as well as maintaining GATA3 expression in already differentiated Th2 cells [18, 32–34]. Therefore, GATA3 up-regulation and STAT5 activation are the two key events for Th2 cell differentiation. Th2 cell differentiation and the induction of Th2 cytokines are also regulated by many other transcription factors including NFAT, NFκb and AP-1 family members. Mechanisms for the reinforcement of Th2 cell differentiation include positive feedbacks, inhibition of other alternative lineage choices and selective growth of differentiated Th2 cells.

Besides Th2 cells, other lymphoid cells including subsets of γδ T cells, NKT cells, T follicular helper (Tfh) cells and type 2 innate lymphoid cells (ILC2s) are also capable of producing IL-4 and/or IL-13. In fact, in the steady state, ILC2s are the major IL-5-producing cells [35,36]. ILC2s exert similar functions as Th2 cells during type 2 immune responses [37,38]. In fact, the production of IL-13 by T cells is dispensable for type 2 immunity suggesting that there is another importance source of IL-13, most likely from ILC2s [39]. While this review will primarily focus on Th2 cell differentiation and the regulation of IL-4/IL-13 production in Th2 cells, the relationships among conventional Th2 cells, IL-4-producing Tfh cells and ILC2s, as well as the regulation of cytokine production in these cell subsets will be also discussed.

2. Signaling pathways involved in Th2 cell differentiation

2.1. IL-4-mediated signaling pathway

IL-4 promotes Th2 cell differentiation mainly by activating STAT6 through tyrosine phosphorylation[20–22]. Naïve STAT6-deficient CD4 T cells fail to up-regulate GATA3 expression and thus are not able to develop into Th2 cells in vitro even when IL-4 is exogenously provided. On the other hand, constitutively active STAT6 mutants are capable of replacing IL-4 in driving Th2 cell differentiation[40, 41]. In addition to up-regulating GATA3 expression, STAT6 is also responsible for chromatin remodeling at the *Il4/Il13* LCR region[16]. The source of IL-4 during initial immune responses that lead to the differentiation of Th2 cells could be NKT cells[42], basophils[43, 44] and CD4 T cells themselves[45, 46].

However, under certain circumstances, Th2 cell differentiation can occur in the absence of the IL-4/STAT6 pathway in vivo[23–26], even though STAT6 is still important for Th2 cells migrating to the lung tissue[26], for optimal expression of GATA3[47], and for the maintenance of Th2 memory cells[24]. IL-4-independent Th2 cell differentiation could be induced by low strength of TCR stimulation with co-stimulation, Notch signaling and/or IL-2-mediated STAT5 activation which will be discussed below.

2.2. TCR-mediated signaling pathway

Strength of TCR signaling is an important factor for determining the fate of T helper cell differentiation[48]. Although cytokine environment can often be the deterministic factor for T helper cell differentiation during infections, TCR signaling strength has a dominant effect over the Th1- or Th2-inducing adjuvants[49–51]. Naive CD4 T cells cultured in vitro with low dose peptide stimulation undergo IL-4/STAT6-independent induction of GATA3 within 24 hours, which is essential for the early IL-4 production by these cells[45]. GATA3 is not up-regulated when naïve CD4 T cells receive high dose peptide stimulation. TCR signaling also induces the activation of p38 kinase, which phosphorylates and regulates GATA3 function[52]. The inhibition of GATA3 up-regulation is due to a strong Erk activation by high levels of TCR activation. This is consistent with the finding that resting dendritic cells (DCs) preferentially induce differentiation of Th2 cells[53]. In fact, omega-1, an important glycoprotein in the *Schistosoma mansoni* egg, inhibits the activation of DCs thus limits the strength of T cell activation; this may be an important mechanism for how *Schistosoma mansoni* egg antigen induces Th2 responses in vivo[54, 55]. Similarly, some in vivo Th2 cell differentiation may depend on basophils, which serves as Th2-promoting antigen presenting cells possibly because of low expression levels of MHC class II on these cells[44, 56, 57].

2.3. Co-stimulation of T cell activation

Although TCR co-stimulation mediated by CD28 is required for optimal T cell activation and Th2 cell differentiation, the CD28 effect is mainly through the induction of IL-2 production[58]. IL-2-STAT5 pathway, which will be further discussed below, is important for Th2 cell differentiation at least in vitro[32, 33]. Whether diminished Th2 responses in CD28-deficient mice are due to reduced IL-2 production is not clear[59, 60]. On the other hand, CTLA-4 is a negative regulator of Th2 cell differentiation[61, 62]. ICOS was originally thought

to co-stimulate Th2 cell differentiation[63, 64], however, ICOSL/ICOS interaction is also known to be important for other T helper cell differentiation, especially for Tfh cells[9]. In addition, OX40 co-stimulation promotes Th2 cell differentiation[65, 66]. Interestingly, TSLP induces the expression of OX40 ligand on DCs thus to promote Th2 response[67].

T cell activation and co-stimulation also activate PI3K pathway, which leads to mTOR (mammalian target of rapamycin) activation; mTOR regulates Th cell fate determination[68, 69]. While mTORC1 signaling is important for Th1 and Th17 differentiation, mTORC2 signaling promotes Th2 cell differentiation partly through activating SGK1 to stabilize JunB protein (an important AP-1 transcription factor for Th2 cell differentiation)[70–73]. However, the dispensable role of mTORC1 during Th2 cell differentiation has been called into question. The conclusion that mTORC1 is dispensable for Th2 cell differentiation is based on the results obtained with Rheb-deficient cells (Rheb is considered as an important upstream activator of mTORC1)[72], however, a study using Raptor-deficient cells (Raptor is the component of mTORC1) indicates that mTORC1 is also important for Th2 cell differentiation[74]. Rheb-deficient cells cultured under Th2 conditions divide slower than wild type cells and there is a Rheb-independent mTORC1 activation; these results are consistent with the notion that mTORC1 is required for metabolic reprogramming and cell proliferation during T cell activation. In addition, PI3K/mTOR pathway is involved in translational regulation of GATA3 protein[75]. Whether the dose of antigen affects preferential activation of either mTORC1 or mTORC2 is not known.

2.4. Notch-mediated signaling pathway

Notch signaling is involved in Th2 cell differentiation[76–78]. It seems that different notch ligand may have different function since Dll1/Notch interaction induces Th1 but Jag1/Notch interaction induces Th2 cell differentiation[76]. After activation, intracellular portion of Notch is translocated to the nucleus and forms a complex with CSL protein to induce gene expression. A Notch/CSL binding site is identified in the 3' of the *Il4* gene suggesting that Notch signaling may directly regulate IL-4 expression[76]. In addition, Notch/CSL complex binds to the distal promoter of *Gata3* gene[78]. Therefore, Notch signaling may promote Th2 cell differentiation by inducing the expression of both GATA3 and IL-4. Notch signaling may also modulate co-stimulation by affecting PI3K pathway to allow naïve CD4 T cells to respond to low dose of antigen[79]. Furthermore, Notch-mediated signaling pathway regulates the expression of IL-2 and IL-2 receptor α chain[79, 80]; IL-2-mediated signaling pathway, discussed in detail below, is one of the key signaling pathways for Th2 cell differentiation.

2.5. STAT5-activating signaling pathways

STAT5 family includes STAT5A and STAT5B, which are encoded by two genetically linked genes[81]. While low amount of STAT5 activation is necessary and sufficient for the proliferation and survival of all the CD4 T cell subsets[32, 82], high levels of STAT5 activation is essential for Th2 cell differentiation both in vitro and in vivo[32, 33, 83, 84]. Enforced expression of a constitutively active form of STAT5A in Th1 cells results in IL-4 production in these cells without up-regulating GATA3 expression[33].

Several cytokines including IL-2, IL-7 and TSLP can activate STAT5 in T cells. TSLP, produced by epithelial cells, dendritic cells (DCs), and/or basophils, may mediate the initiation of Th2 responses in vivo[43, 67, 85, 86]. Although the major role of TSLP is to regulate the functions of dendritic cells[67, 87], TSLP may also directly act on T cells to promote Th2 cell differentiation[88, 89]. Other potent STAT5 activators are IL-2 and IL-7. While IL-7 is capable of inducing and/or promoting Th2 differentiation in vitro, its function in vivo has not been carefully studied. IL-2 is mainly a product of activated T cells and thus may serve as an important autocrine cytokine during Th2 differentiation. However, due to a broader function of IL-2, including its effect on regulatory T cells, it is difficult to assess the precise role of IL-2 during Th2 cell differentiation in vivo.

It is known that TCR-mediated signaling transiently suppresses cytokine signaling during initial T cell activation[90]. Both IL-2/STAT5 and IL-4/STAT6 signaling pathways are inhibited 4–6 hours of T cell activation. Interestingly, IL-2-mediated STAT5 activation is detected at the early stages of T cell activation with low dose but not high dose of TCR stimulation[45]. As mentioned above, Notch signaling induces the expression of IL-2 and IL-2 receptor α chain during T cell activation. In fact, all five Th2-promoting signaling pathways triggered by antigen, co-stimulation, Notch, IL-4 and IL-2, respectively, may crosstalk with each other and collaborate to induce Th2 cell differentiation.

2.6. Th2 cell differentiation via a default program

IFNγ produced by Th1 cells is an important cytokine that promotes Th1 cell differentiation[91–93]. IFNγ is also produced by NK cells and/or ILC1s, which are the first responders at the innate phase of type 1 immune responses[93–96]. In addition, IL-12 produced by antigen presenting cells including DCs plays an important role in driving Th1 cell differentiation both in vitro and in vivo through activating STAT4 in CD4 T cells[97–99]. On the other hand, an equivalent DC-derived cytokine that induces Th2 cell differentiation has not been found. Instead, IL-2 and IL-4, two known cytokines driving Th2 cell differentiation, are mainly produced by T cells themselves. Thus, the determination of Th1 and Th2 cell fate appears to be asymmetrical. Even more interestingly, because GATA3 is critical for the development of CD4 T cells at multiple stages[100], it is already expressed by naïve CD4 T cells[28, 101]. Furthermore, STAT5 is usually activated either by IL-7 at naïve stage or by IL-2 upon T cell activation.

Given that a Th2-biased differentiation program (basal GATA3 expression and STAT5 activation) has pre-existed in naïve CD4 T cells, IL-4/STAT6 signaling, which possibly serve as a reinforcement and/or amplification of Th2 responses, may not be necessary for Th2 cell differentiation, especially at early stages, in many situations in vivo. Indeed, *Nippostrongylus brasiliensis* and *Schistosoma mansoni* infection exclusively elicit IL-4-independent Th2 responses[23–27]. The requirement of IL-4/STAT6 during some *in vivo* Th2 responses such as in response to *Trichuris muris* infection[102] may simply because such infection generates a mixture of Th1 and Th2 responses and IL-4 is required to suppress Th1 responses. Consistent with the notion that Th2 cell differentiation can occur through a default pathway when the major Th1-inducing signals are absent, mice deficient in either Th1 master regulator T-bet[103] or STAT4/IFNγR doubly deficient mice developed Th2

responses in response to *Toxoplasma gondii* infection[104], which usually induces robust Th1 responses. However, in the absence of IL-12 alone, T cells fail to default to a Th2 program[105], suggesting that both IL-12 and IFNγ are involved in suppressing the endogenous Th2 program in T cells. Indeed, either IL-12 or IFNγ is able to induce T-bet expression[104].

3. Key transcription factors required for Th2 cell differentiation

3.1. GATA3

GATA3 has been recognized as the master regulator of Th2 cells[28–31]. GATA3 is expressed by naïve CD4 T cells but its expression levels are induced during Th2 differentiation[29, 106, 107]. GATA3 is sufficient to induce the Th2 phenotype since retrovirus-mediated enforced expression of GATA3 in Th1 cultures induces IL-4 production and endogenous GATA3 expression[106, 108]. On the other hand, knocking down or deletion of GATA3 results in diminished Th2 cell differentiation, whether it is IL-4-dependent or IL-4-independent, both in vitro and in vivo[28, 30, 31, 109].

GATA3 promotes Th2 cell differentiation through multiple mechanisms[110]. The key role of GATA3 is to directly act on the *Il4/Il13* gene locus at various sites. Chromatin immune-precipitation followed by high throughput sequencing (ChIP-Seq) analysis with anti-GATA3 reveals genome wide pattern of GATA3 binding[101]. In Th2 cells, GATA3 binds to the promoters of the *Il5* and the *Il13* gene including the CGRE site in the *Il13* distal promoter[111, 112]; GATA3 also binds to multiple sites at the LCR region in the *Rad50*[113, 114] and several enhancer regulatory elements for the *Il4* genes, such as DNase I hypersensitivity II (HSII)[112, 115] and HSV sites[116]. GATA3 regulates epigenetic modifications in the Th2 cytokine locus during Th2 cell differentiation. However, in already differentiated Th2 cells, GATA3 is dispensable for IL-4 production although it is still essential for IL-5 and IL-13 production possibly because GATA3 is critical for activating the promoters of these two genes[30]. Besides its direct action on the Th2 cytokine locus, GATA3 also directly regulate many Th2 specific genes such as *Il1rl1* encoding T1/ST2, the IL-33 receptor, and many chemokine receptor genes such as *Ccr8*[101].

3.2. STAT5

As discussed above, a strong STAT5 activation is another key element for Th2 cell differentiation. Co-expression of GATA-3 and a constitutively active form of STAT5A in non-Th2 cells results in a larger proportion of IL-4-producing cells compared to that induced by either GATA3 or active STAT5A alone[33]. On the other hand, GATA3 has very limited IL-4-inducing capacity when STAT5 activation is inhibited by anti-IL-2 treatment[32] and the active STAT5A is unable to induce IL-4 in *Gata3*-deficient CD4 T cells[30]. Therefore, both GATA3 induction and STAT5 activation are essential for Th2 cell differentiation; high STAT5 activation levels may reduce the requirement of GATA3 expression levels for inducing IL-4 expression[117].

Mechanistically, STAT5 binds to the HSII and HSIII sites of the *Il4* gene, through which it regulates the accessibility of these sites[32, 33]. ChIP-Seq results confirming STAT5 binding at the HSII site has also revealed another STAT5 binding at the LCR region, which may

contribute to inducing IL-4[118]. Besides directly regulating the *Il4/Il13* locus, IL-2/STAT5 induces the expression of IL-4Rα especially at the initial stage of Th2 cell differentiation so that activated T cells may respond to IL-4 to complete the differentiation process[118].

3.3. Other transcription factors

In addition to GATA3 and STAT5, there are several other transcription factors that regulate IL-4 production and/or Th2 cell differentiation (Figure 1). IRF-4 is highly up-regulated during T cell activation and it is important for the differentiation and functions of virtually any CD4 T helper subsets including Th2 cells[119–128]. In Th2 cells, one important function of IRF4 is to bind to the *Il4* promoter to regulate its expression[121, 122].

It has been recently reported that c-Maf is highly expressed by Th17 cells[126, 129, 130] and Tfh cells[131]; c-Maf regulates the expression of IL-10, IL-21 and IL-22. However, c-Maf was initially considered as a Th2 specific transcription factor by comparing its expression in Th1 and Th2 cells. Indeed, it controls the production of IL-4 but not other Th2 cytokines[132]. C-Maf directly binds to the promoter of the *Il4* gene. C-Maf expression is negatively regulated by a LincRNA, linc-MAF-4, which is highly expressed in human Th1 cells[133].

Klf13 collaborates with c-Maf in regulating IL-4 production in T cells[134, 135]. The action of c-Maf at the *Il4* promoter also relies on JunB, which is critical for Th2 cell differentiation[70]. The induction of JunB expression partially depends on Dec2, another Th2-specific transcription factor[136]. Nfil3 (E4BP4), a transcription factor that is critical for the development of NK cells and ILCs[137–142], is highly expressed by Th2 cells and responsible for regulating IL-10 production[143]. In addition, YY1 physically interacts with GATA3 and thus collaborates with GATA3 in chromatin remodeling and cytokine production[144].

IL-4 production and Th2 cell differentiation can also be negatively regulated by many transcription factors. One typical example is Runx3, which binds to the HSIV site of the *Il4* gene and suppresses IL-4 production[145–147]. Both Eomes, which is expressed by a subset of memory Th2 cells, and Sox4 that is induced by TGFβ suppress GATA3 activity and Th2 cytokine expression[148, 149]. PU.1, a critical transcription factor regulating myeloid and lymphoid cell development at early stages, is also expressed by subsets of T cells. In Th2 cells, PU.1 is preferentially expressed by IL-4 non-producers suggesting that PU.1 negatively regulates IL-4 production[150].

GATA3 is expressed by some activated regulatory T cells (Tregs) at an intermediate level[151–154], however, such cells do not produce IL-4 suggesting that Foxp3 may suppress the GATA3 function in inducing Th2 cytokines. Interestingly, reduced Foxp3 expression in Tregs results in Th2 cytokine production by these cells[155]. Such Foxp3 effect may be particularly important in humans since activated human effector T cells can transiently express Foxp3[156, 157].

Therefore, Th2 cell differentiation and cytokine production depend on a network of many transcription factors, which consists of GATA3, STAT5, IRF4, c-Maf, JunB, Dec2, Klf13, E4BP4, YY1, Runx3 and PU.1 etc. Changes in the balance and/or the ratio of these transcription factors may lead to an alteration in Th2 cell fate determination.

4. Mechanisms of reinforcing Th2 cell differentiation

4.1. Positive feedback

A basic principle of T helper cell differentiation is positive feedback mechanism[2]. During Th2 cell differentiation, IL-4 produced by developing Th2 cells may instruct IL-4 non-producers to produce IL-4, and/or enhance the IL-4-producing capacity of the IL-4-producing cells in a paracrine and/or autocrine manner (Figure 1). Not only can IL-4 induce GATA3 expression and thus IL-4 production, it also enhances the sensitivity of developing Th2 cells to IL-4 stimulation by up-regulates IL-4Rα expression. Therefore, IL-4/GATA3/IL-4 and IL-4/IL-4R constitute two powerful positive feedback loops during Th2 differentiation.

It has been reported that GATA3 auto-regulates its expression[108]. Our ChIP-Seq data suggest that GATA3 binds to multiple regions at the *Gata3* locus across 1 Mb length of DNA[101]. Therefore, GATA3 may regulate its own expression directly. However, in the absence of functional GATA3 protein, the truncated GATA3 mRNA expression is not reduced when T cells are activated in the presence of IL-4[101]. This result suggests that GATA3 auto-regulation is minimal when IL-4/STAT6 signaling is present. Nevertheless, GATA3 auto-regulation may be critical to sustain Th2 phenotype when IL-4/STAT6 signaling ceases. Among the GATA3 binding sites at the *Gata3* gene, one co-localizes with a T cell specific enhancer for GATA3 expression[158]. Even more interesting, this enhancer region overlaps with the dual promoter that mediates the transcription of two long intergenic non-coding RNAs (LincRNAs) into opposite directions[159]. Whether the expression of these two LincRNAs is related to the “enhancer” activity needs to be further studied.

GATA3 may indirectly regulate itself by inducing other transcription factors such as Dec2 and IRF4; the expression of Dec2 and IRF4 is reduced when GATA3 is acutely deleted from Th2 cells[101, 136]. On the other hand, both Dec2 and IRF4 are able to enhance GATA3 expression levels in Th2 cells[121, 136].

Positive feedback regulations are also found between the two critical pathways, IL-2/STAT5 and IL-4/STAT6/GATA3, during Th2 cell differentiation. Besides inducing CD25 (IL-2 receptor α chain) expression[160], IL-2-mediated STAT5 activation up-regulates the expression of IL-4Rα during the initial T cell activation[118]. Furthermore, STAT5 activation is required to maintain GATA3 expression in differentiated Th2 cells[34]. On the other hand, GATA3 regulates optimal expression levels of CD25 by T effector cells as well as Tregs[101, 151].

4.2. Suppression of other lineage fates

CD4 T cells can differentiate into different effector lineages including Th1, Th2 and Th17 cells. As mentioned above, IL-12/STAT4 and IFNγ/STAT1 pathways, both of which induce the master regulator T-bet, are important for Th1 cell differentiation[92, 97, 98, 103, 161]. Therefore, STAT1/STAT4 activation and T-bet induction are the key events during Th1 cell differentiation. Similarly, Th17 cell differentiation requires STAT3 activation and the induction of RORγt expression[1, 8].

Th2 cell differentiation is accompanied by the suppression of other lineage fates. GATA3 deficiency leads to de-repression of STAT4 expression and enforced expression of GATA-3 down-regulates the expression of STAT4 in Th1 cells[146, 162]. Interestingly, *Gata3* deletion during Th2 cell differentiation results in an IL-12/STAT4 and IFNγ/T-bet independent IFNγ production, which is mediated by Runx3/Eomesodermin (Eomes) pathway[30, 145–147, 163]. It has been shown that T-bet inhibits GATA-3 transcription[107] and suppresses its function through protein-protein interaction[164]. T-bet and GATA3 co-expressing cells can be generated in response to parasite infection[165]. The physical interaction between GATA3 and T-bet may allow them to suppress each other's function. Indeed, T-bet and GATA3 binding sites co-localize at many critical Th1- or Th2-specific genes[166]. Furthermore, GATA3 is able to silence *Tbx21* (encoding T-bet) locus in Th2 cells by its direct binding to a site in the *Tbx21* locus with suppressive epigenetic modifications; removing GATA3 results in a substantial reduction of such suppressive modifications at the binding site [101]. Thus, GATA3 suppresses Th1 cell fate through multiple mechanisms[110].

Strong STAT5 activation is necessary for Th2 differentiation as discussed above. Interestingly, constitutively active STAT5a suppresses T-bet expression when introduced into developing Th1 cells [33]. STAT5 activation also suppresses Th17 differentiation and Tfh cell differentiation [167–171]. Besides GATA3 and STAT5, another transcription factor, growth factor independent 1 (Gfi-1), which is preferentially expressed in Th2 cells, inhibits IFNγ and IL-17 production in Th1 and Th17 cells, respectively [172–174].

4.3. Selective growth of Th2 cells

IL-12 receptor β2 chain is selectively up-regulated during Th1 cell differentiation [175], thus, IL-12 plays an important role in selecting committed Th1 cells to grow [176]. However, although IL-4 receptor α chain can be up-regulated by IL-4 [177], naïve CD4 T cells as well as non-Th2 cells are also able to respond to IL-4 because of constitutive expression of IL-4 receptor on T cells [90]. Therefore, IL-4 receptor up-regulation is not the major mechanism for selective outgrowth of Th2 cells in response to IL-4 stimulation. Nevertheless, IL-4 up-regulates Gfi-1 expression through STAT6 activation; Gfi-1 preferentially induces the expansion of GATA-3^{hi} Th2 cells through its actions upstream and downstream of STAT5 activation[172, 174].

In fully differentiated Th2 cells, IL-33 receptor α chain (T1/ST2 or IL-1R–like 1) is highly expressed[178, 179]. Since IL-33 is released during Th2 responses, IL-33 may play a role in promoting the survival and/or expansion of fully differentiated Th2 cells. Indeed, blocking IL-33 signaling results in attenuated eosinophilic airway inflammation and decreased responses to *Schistosoma mansoni*[180, 181]. Strikingly, continuous IL-33Rα expression in differentiated Th2 cells depends on both GATA3 expression and STAT5 activation[34].

5. Chromatin remodeling and epigenetic modifications at the *Il4/Il13* locus

The *Il4* and *Il13* genes, located at chromosome 5 in human and chromosome 11 in mouse, respectively, are flanked by the *Rad50* and *Kif3a* genes. The locus control region (LCR) of the *Il4-Il13* locus is within the 3' of *Rad50* gene[113]; the *Il5* gene is on the other side of the *Rad50*. There are GATA3-mediated intrachromosomal interactions between the LCR and

the Th2 cytokine gene promoters[182]. A potential regulatory element may reside within the *Kif3a* gene based on the specific-lineage epigenetic markers in Th2 cells in this region[101]. Transcriptional regulation of *Il4/Il13* expression, like transcriptional regulation of any other lineage specific genes, highly depends on chromatin remodeling and epigenetic modifications[17, 183, 184].

During Th2 differentiation, several Th2-specific DNase I hypersensitivity (HS) sites are induced at the *Il4-Il13* locus[185]. These include the HSII site in the intron 2 of the *Il4* gene[112, 186], HSV and HSVa[116] at 3' of the *Il4* coding region, conserved non-coding sequence 1 (CNS1)[187, 188] which is located at the intergenic region of *Il4* and *Il13*, and several HS sites within the LCR in the *Rad50* gene[113, 189]. Interestingly, the HS IV site is accessible in both Th1 and Th2 cells[185]; Runx3 binds to this region in Th1 cells to suppress IL-4 production[145, 190]. Deletion of the RHS7 (HS7 site within the LCR in the *Rad50*) site results in reduced IL-4 and IL-13 production but not IL-5 in Th2 cells[191]. However, deletion of the RHS6 site or the whole LCR affects the expression of all three cytokines[192, 193]. The requirement of these elements to induce cytokine production seems to vary when different immunization protocols are followed[192] suggesting that the LCR contains both IL-4-depedent and IL-4-independent regulatory elements, consist with the findings that STAT6 binds to both the RHS6 and RHS7 sites[16], but GATA3 only binds to RHS5 and RHS6 not RHS7 of the LCR[101]. Activated STAT5 binds to the HSII site and induces the accessibility at this region[33]. ChIP-Seq analysis also indicates that GATA3 binds the HSII site[101]. Our unpublished data suggest GATA3 is responsible for the induction and maintenance of HSII accessibility.

Different histone modifications are associated with either gene activation or repression[194]. Tri-methylation and di-methylation at the lysine position 4 of histone 3 (H3K4me2 and 3) are usually indicative of active gene loci, whereas, H3K27me3 is a marker for repressed gene loci. The *Il4/Il13* locus displays H3K4me3 modification in Th2 cells and H3K27me3 in Th1 cells[195]. MLL, a histone H3K4 methyltransferase, is important for maintaining H3K4 methylation at the *Il4/Il13* locus in differentiated Th2 cells[196]. On the other hand, Ezh2, an H3K27 methyltransferase, is responsible for mediating H3K27me3 modification at the *Il4/Il13* and *Gata3* loci in Th1 cells[197–199]. Deletion of Ezh2 in T cells results in reduced H3K27me3 at the *Tbx21* and *Gata3* loci, and thus enhanced Th1 and Th2 differentiation. Ezh2 deletion also causes reduced stability of differentiated cells and enhances their plasticity. Besides histone methylation modifications, histone acetylation at the lineage-specific cytokine gene loci has also been described[200].

Although GATA3 may regulate the expression of IL-5 and IL-13 through its direct binding to the promoters of the *Il5* and *Il13* genes[30, 111, 201, 202], a main function of GATA3 in Th2 cell fate determination seems to mediate histone modifications at Th1 and Th2 specific gene loci including the *Ifng* and *Il4/Il13* locus[108, 203, 204]. In the absence of GATA3, H3K27me3 marks around the GATA3 binding site at the *Ifng* locus is greatly reduced. On the other hand, H3K4me2 marks at the *Il4/Il13* locus, especially at the LCR region where GATA3 binds, are diminished when GATA3 is absent. At a genome level, GATA3 binds to ∼2000 genes in Th2 cells[101]. When GATA3 is deleted, ∼100 genes among these 2000 genes have altered their expression level, however, alteration of histone modifications occurs at the loci

of ~900 genes[101]. This strongly indicates that GATA3-dependent alterations in epigenetic modifications is not a result of gene expression change.

DNA CpG methylation also affects gene expression[205]. Deletion of Dnmt-1, a DNA methyltrasferase, or of methyl CpG-binding domain protein-2 (MBD2), results in aberrant expression of IL-4 without affecting GATA3 expression[206, 207]. Interestingly, GATA3 inhibits the binding of MBD2 to methyl CpG. STAT5 activation is partially involved in de-methylation of the RHS7 site in the LCR during Th2 differentiation[189].

Collectively, both GATA3 and STAT5 strongly bind to key regulatory elements of the *Il4/Il13* locus including the HSII site[112] and HS sites within the LCR[191]; both GATA-3 and STAT5 are involved in many aspects of chromatin remodeling at the *Il4/Il13* locus including chromatin accessibility, histone modifications and DNA methylation.

6. Induction of IL-4 and IL-13 expression by Th2 cells

TCR-mediated signaling plays an important role in T helper cell differentiation as discussed above. Another critical function of TCR-mediated signaling is to induce cytokine production in already differentiated T helper cells. The most efficient way to stimulate Th2 cells to produce cytokines is through their TCRs or by chemicals such as PMA and ionomycin to mimic TCR-mediated signaling. TCR stimulation activates NFAT family members in a Ca^{2+} dependent manner[208]. Upon TCR re-stimulation of differentiated Th2 cells, NFAT translocates to the nucleus and binds to the *Il4* promoter as well as HSVa region to regulate IL-4 expression[122, 209]. Besides inducing cytokine production, NFAT proteins are also involved in Th2 cell differentiation[210].

T helper cells can also produce cytokines in response to cytokine stimulation. IL-18 together with IL-12 induces IFNγ production independent of TCR stimulation[211, 212]. Similarly, IL-33 in combination of STAT5 activators, such as IL-2, IL-7 and TSLP, induces IL-13 but not IL-4 expression in differentiated Th2 cells[178, 179]. In addition, Th2 cells express cysteinyl leukotriene receptor 1 and are able to respond to leukotrienes to produce IL-13 possibly through calcium-dependent pathway[213, 214].

Whether Th2 cells are stimulated by TCR to produce IL-4 and IL-13, or by cytokines to produce IL-13, not all Th2 cells actually produce IL-4 and/or IL-13 upon stimulation. Although heterogeneity of Th2 cells can be a result of differential epigenetic status of each cytokine allele[215, 216], the actual cytokine production in a given cell upon a single round of stimulation may be a stochastic event[217]. Heterogeneous activation of transcription factors such as NFAT or differential assembly of functional transcription factor complex needed for inducing cytokine production may be one of the main explanations[217, 218]. Thus, IL-4- and/or IL-13-non-producing cells identified after stimulation at one time may express these cytokines in response to another round of stimulation. Therefore, both cytokine-producing and non-producing cells may represent fully differentiated cells. Indeed, IL-4-producers and non-producers from a same Th2 culture express similar levels of GATA3 and have identical chromatin accessibility at the *Il4/Il13* locus[217]. Interestingly, this may not be entirely true for IFNγ production in Th1 cells since low- and high-IFNγ-producing cells express different levels of T-bet and seem to have a quantitative memory to produce IFNγ[219].

7. Relationship between Th2 cells and IL-4-producing Tfh cells

Tfh cells, many found in the germinal centers in the B cell follicle, are critical helper T cells in helping B cells produce antibodies and undergo immunoglobulin (Ig) class switching[9]. IL-4 is essential for the Ig class switching to IgE[10], suggesting that some Tfh cells need to produce IL-4[220–222]. Indeed, IL-4-producing CD4 T cells are mainly Tfh cells[222]. Interestingly, Tfh cells do not produce IL-13[47].

GATA3-deficient mice fail to generate type 2 response including IgE class switching[30] suggesting that GATA3 is also required for Tfh cells to produce IL-4. However, Tfh cells containing IL-4-producers express low levels of GATA3 suggesting that small amount of GATA3 is sufficient for IL-4 production in Tfh cells[47, 223]. On the other hand, IL-13-producing Th2 cells are GATA3 high expressors[47]. GATA3 at a low level is sufficient to induce IL-4 but not IL-13 production when there is a strong STAT5 activation[33]. However, Tfh cells express low levels of IL-2Rα, and STAT5 activation is paradoxically suppressive for Tfh cell differentiation[168, 224].

Since c-Maf is highly expressed by Tfh cells[131] and it is important for IL-4 production in Th2 cells[132], it is possible that high levels of c-Maf expression by Tfh cells may compensate low GATA3 expression and/or low STAT5 activity for inducing IL-4. Interestingly, deletion of a DNA segment containing both HSV and HSVA results in impaired IL-4 production in both Th2 cells and mast cells[116], however, only deleting the CNS2 region at the HSV site dramatically affects IL-4 production in Tfh cells and in naïve CD4 T cells but has a small effect on IL-4 production in conventional effector Th2 cells in tissue[225, 226]. Consequently, IgE induction is completely abolished in CNS2 deficient mice. Furthermore, CNS2 region is much more active in Tfh cells[225]. Since Notch/CSL complex directly binds the CNS2 region[76], Notch signaling may be constitutively active and critical for IL-4 production in Tfh cells.

The relationship between classical Th2 cells and IL-4-producing Tfh cells is still not certain. An in vitro study suggests that T cells can sequentially gain cytokine producing capacity and follicular helper phenotype[227]. That is, conventional Th2 cells can be converted to IL-4-producing Tfh cells. Similarly, Tfh cells can be further differentiated to produce IL-4. In a transfer model, it has been shown that activated T cells that have gained IL-4-producing capacity can later on display a Tfh cell phenotype in a B cell dependent manner[221]. However, this does not rule out the possibility that some Tfh cells gain IL-4-producing capacity after they have entered germinal center. Alternatively, activated T cells may gain IL-4-producing capacity and features of Tfh cells simultaneously, and their fate to become either classical effector Th2 cells or IL-4-producing Tfh cells may be determined during early T cell differentiation. Indeed, it has been shown that there is an early lineage commitment to either T effector cells or Tfh cells based on their Bcl6 and IL-2Rα expression two days after immunization[168]. Bcl6 is transiently induced during Th1 cell differentiation supporting the model of early fate determination[228], however, whether Bcl6-expressed/expressing cells will become Tfh cells later is unknown.

IL-4-producing cells are mainly found in the B cell follicle whereas IL-13-producing cells are found in the tissue[47]; this suggests that Th2 effector cells and IL-4-producing Tfh cells may adopt distinct differentiation programs which involves regulation of GATA3 levels and other transcription factors collaborating with GATA3. Interestingly, it has also been shown that memory T helper cells may originate from Tfh cells[229]. A careful genome-wide assessment of the epigenetic status in conventional Th2 cells, IL-4-producing Tfh cells and Th2 memory cells may help understand the relationship between these cells.

8. Relationship between Th2 cells and ILC2

ILC2s are important players during type 2 immune responses particularly at early stages[230–233] and their development requires transcription factors RORα[234, 235] and GATA3[38, 236–240]. Gfi-1, also highly expressed by ILC2s, is involved in the development and/or maintenance of ILC2s[241]; without Gfi-1, ILC2s aberrantly express IL-17 similar to the finding in Gfi-1-deficient T cells[173]. ILC2s not only contribute to the expulsion of helminths, they are also involved in allergic lung and skin inflammation[242–246]. ILC2s are also involved in adipocyte differentiation and energy metabolism[35, 247, 248]; whether Th2 cells have similar functions is an important question of interest.

When pharmacologically stimulated by cytokines such as IL-25 or IL-33, ILC2s are able to expel worms in the absence of adaptive immune system. During activation, ILC2s transiently produce IL-9, which in turn maintains ILC2 survival in an autocrine manner[249, 250]. IL-9 receptor is highly expressed on ILC2s and such expression depends on GATA3[38, 250]. GATA3 is also critical for the expression of many ILC2-specific genes[38, 47], including IL-5 and IL-13 that are also shared by Th2 cells, and GATA3 is indispensable for the maintenance of ILC2 cell numbers in vivo[38, 237].

ILC2s and Th2 cells have identical functions and similar requirement of transcriptional machinery in cytokine production (Figure 2) [37, 251]. However, due to the lack of an antigen receptor and toll-like receptor, ILC2s mainly respond to cytokines such as IL-33 to produce type 2 cytokines, IL-5 and IL-13, but not IL-4.

ILC2s also respond to cysteinyl leukotrienes to produce cytokines[252]. Unlike IL-33, cysteinyl leukotriene D4 induces the expression of IL-4 in addition to IL-5 and IL-13. PGD2, a product of mast cells after IgE-mediated degranulation, can induce ILC2 migration and cytokine production through its interaction with CRTH2 expressed by ILC2s[253]. Interestingly, basophil-derived IL-4 is able to induce ILC2 proliferation[245] and IL-13 production by ILC2s[254]; this direct effect of IL-4 on ILC2s to produce IL-13 requires further investigation and confirmation by other groups. On the other hand, lipoxin A4 and E-cadherin (a ligand for KLRG1) suppress cytokine production by ILC2s[255, 256].

ILC2s and Th2 cells collaborate during type 2 immune responses[257–260]. In a lung inflammation model, papain-induced IL-33 activates ILC2s to produce IL-13, which induces the migration and activation of lung dendritic cells (DCs) to the draining lymph nodes[260]; these migratory DCs then induce Th2 cell differentiation in an IL-4-independent manner. Some ILC2s also express MHC class II through which activate T cells to produce IL-2, and IL-2 acts back on ILC2s to produce type 2 cytokines[257, 258]. IL-2 has been reported to

induce ILC2 proliferation and cytokine release[246, 249, 261]. Co-culture ILC2s with naïve CD4 T cells results in ILC2 proliferation and cytokine production, which is mediated by IL-2 produced by T cells[258]. While it is clear that ILC2s are capable of inducing Th2 cell differentiation in vitro possibly through IL-4 production[259], whether ILC2 and T cell direct interaction is critical for initiating and promoting Th2 cell differentiation in vivo, or this interaction mainly reflects collaborative relationship of ILC2s and Th2 cells at the effector stage requires further investigation.

9. Summary and conclusions

During Th2 cell differentiation, multiple signaling pathways result in two major critical events: up-regulation of GATA3 expression and activation of STAT5 proteins. Both GATA3 expression and STAT5 activation are essential for Th2 cell differentiation. GATA3 and STAT5 directly act on the *Il4/Il13* locus through chromatin remodeling and epigenetic modifications. Several other transcription factors are also involved in Th2 cell differentiation. Th2 cell lineage commitment, determined by a network of transcription factors, is accompanied by blockage of alternative lineage fates and selective growth of already differentiated Th2 cells. Once Th2 cells are fully differentiated, the production of Th2 cytokines including IL-4, IL-5 and IL-13 requires re-stimulation by antigens through TCR, by cytokines such as IL-33, or possibly by other inflammatory molecules such as cysteinyl leukotrienes. Some transcription factors that are important for Th2 lineage commitment may also be involved in regulating Th2 cytokine production upon restimulation.

We have gained much knowledge of Th2 cell differentiation mainly from *in vitro* studies. However, our understanding of Th2 cell differentiation *in vivo* is far from complete. A particular example is that while IL-4/STAT6 pathway is absolutely necessary for *in vitro* Th2 cell differentiation, this pathway is only minimally involved in many *in vivo* Th2 responses. The complexity of Th2 responses *in vivo* is partially due to the involvement of other cytokines such as TSLP, IL-25 and IL-33[85, 262, 263], and the participation of many immune cells including DCs, ILC2s, NKT cells, basophils, eosinophils and mast cells[264]. In the future, it is critical to further understand the relationship between conventional Th2 cells and IL-4-producing Tfh cells, and the functional similarity and difference between Th2 cells and ILC2s, particularly during type 2 immune responses in vivo. Two particular intriguing questions are: how GATA3 with various amounts differentially regulates IL-4 and IL-13 in conventional Th2 cells and Tfh cells, respectively; why Th2 cells express both IL-4 and IL-13 but ILC2s mainly produce IL-13 but not IL-4. By simultaneously studying Th2 cells and ILC2s, common and unique pathways/molecules that are involved in regulating type 2 immune responses will be identified; these pathways and molecules may be considered as drug targets for treating Th2- and/or ILC2-related diseases including allergy and asthma.

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References

1. Zhu J, Paul WE. CD4 T cells: fates, functions, and faults. Blood. 2008; 112:1557–1569. [PubMed: 18725574]
2. Zhu J, Yamane H, Paul WE. Differentiation of effector CD4 T cell populations. Annual review of immunology. 2010; 28:445–489.
3. Mosmann TR, Cherwinski H, Bond MW, Giedlin MA, Coffman RL. Two types of murine helper T cell clone. I. Definition according to profiles of lymphokine activities and secreted proteins. J Immunol. 1986; 136:2348–2357. [PubMed: 2419430]
4. Mosmann TR, Coffman RL. TH1 and TH2 cells: different patterns of lymphokine secretion lead to different functional properties. Annual review of immunology. 1989; 7:145–173.
5. Paul WE, Seder RA. Lymphocyte responses and cytokines. Cell. 1994; 76:241–251. [PubMed: 7904900]
6. Bilate AM, Lafaille JJ. Induced CD4+Foxp3+ regulatory T cells in immune tolerance. Annual review of immunology. 2012; 30:733–758.
7. Abbas AK, et al. Regulatory T cells: recommendations to simplify the nomenclature. Nature immunology. 2013; 14:307–308. [PubMed: 23507634]
8. Korn T, Bettelli E, Oukka M, Kuchroo VK. IL-17 and Th17 Cells. Annual review of immunology. 2009; 27:485–517.
9. Crotty S. Follicular helper CD4 T cells (TFH). Annual review of immunology. 2011; 29:621–663.
10. Kopf M, et al. Disruption of the murine IL-4 gene blocks Th2 cytokine responses. Nature. 1993; 362:245–248. [PubMed: 8384701]
11. Gordon S. Alternative activation of macrophages. Nature reviews immunology. 2003; 3:23–35.
12. Coffman RL, Seymour BW, Hudak S, Jackson J, Rennick D. Antibody to interleukin-5 inhibits helminth-induced eosinophilia in mice. Science (New York, N.Y. 1989; 245:308–310.
13. Kuperman DA, et al. Direct effects of interleukin-13 on epithelial cells cause airway hyperreactivity and mucus overproduction in asthma. Nature medicine. 2002; 8:885–889.
14. Wynn TA. IL-13 effector functions. Annual review of immunology. 2003; 21:425–456.
15. Urban JF Jr, et al. IL-13, IL-4Ralpha, and Stat6 are required for the expulsion of the gastrointestinal nematode parasite Nippostrongylus brasiliensis. Immunity. 1998; 8:255–264. [PubMed: 9492006]
16. Lee DU, Rao A. Molecular analysis of a locus control region in the T helper 2 cytokine gene cluster: a target for STAT6 but not GATA3. Proceedings of the National Academy of Sciences of the United States of America. 2004; 101:16010–16015. [PubMed: 15507491]
17. Ansel KM, Djuretic I, Tanasa B, Rao A. Regulation of Th2 differentiation and Il4 locus accessibility. Annual review of immunology. 2006; 24:607–656.
18. Le Gros G, Ben-Sasson SZ, Seder R, Finkelman FD, Paul WE. Generation of interleukin 4 (IL-4)-producing cells in vivo and in vitro: IL-2 and IL-4 are required for in vitro generation of IL-4-producing cells. The Journal of experimental medicine. 1990; 172:921–929. [PubMed: 2117636]
19. Swain SL, Weinberg AD, English M, Huston G. IL-4 directs the development of Th2-like helper effectors. J Immunol. 1990; 145:3796–3806. [PubMed: 2147202]
20. 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; 4:313–319. [PubMed: 8624821]
21. Shimoda K, et al. Lack of IL-4-induced Th2 response and IgE class switching in mice with disrupted Stat6 gene. Nature. 1996; 380:630–633. [PubMed: 8602264]
22. Takeda K, et al. Essential role of Stat6 in IL-4 signalling. Nature. 1996; 380:627–630. [PubMed: 8602263]
23. Jankovic D, et al. Single cell analysis reveals that IL-4 receptor/Stat6 signaling is not required for the in vivo or in vitro development of CD4+ lymphocytes with a Th2 cytokine profile. J Immunol. 2000; 164:3047–3055. [PubMed: 10706693]
24. Finkelman FD, et al. Stat6 regulation of in vivo IL-4 responses. J Immunol. 2000; 164:2303–2310. [PubMed: 10679064]

25. Min B, et al. Basophils produce IL-4 and accumulate in tissues after infection with a Th2-inducing parasite. The Journal of experimental medicine. 2004; 200:507–517. [PubMed: 15314076]
26. Voehringer D, Shinkai K, Locksley RM. Type 2 immunity reflects orchestrated recruitment of cells committed to IL-4 production. Immunity. 2004; 20:267–277. [PubMed: 15030771]
27. van Panhuys N, et al. In vivo studies fail to reveal a role for IL-4 or STAT6 signaling in Th2 lymphocyte differentiation. Proceedings of the National Academy of Sciences of the United States of America. 2008; 105:12423–12428. [PubMed: 18719110]
28. Zheng W, Flavell RA. The transcription factor GATA-3 is necessary and sufficient for Th2 cytokine gene expression in CD4 T cells. Cell. 1997; 89:587–596. [PubMed: 9160750]
29. Zhang DH, Cohn L, Ray P, Bottomly K, Ray A. Transcription factor GATA-3 is differentially expressed in murine Th1 and Th2 cells and controls Th2-specific expression of the interleukin-5 gene. The Journal of biological chemistry. 1997; 272:21597–21603. [PubMed: 9261181]
30. Zhu J, et al. Conditional deletion of Gata3 shows its essential function in T(H)1-T(H)2 responses. Nature immunology. 2004; 5:1157–1165. [PubMed: 15475959]
31. Pai SY, Truitt ML, Ho IC. GATA-3 deficiency abrogates the development and maintenance of T helper type 2 cells. Proceedings of the National Academy of Sciences of the United States of America. 2004; 101:1993–1998. [PubMed: 14769923]
32. Cote-Sierra J, et al. Interleukin 2 plays a central role in Th2 differentiation. Proceedings of the National Academy of Sciences of the United States of America. 2004; 101:3880–3885. [PubMed: 15004274]
33. Zhu J, Cote-Sierra J, Guo L, Paul WE. Stat5 activation plays a critical role in Th2 differentiation. Immunity. 2003; 19:739–748. [PubMed: 14614860]
34. Guo L, et al. IL-1 family members and STAT activators induce cytokine production by Th2, Th17, and Th1 cells. Proceedings of the National Academy of Sciences of the United States of America. 2009
35. Nussbaum JC, et al. Type 2 innate lymphoid cells control eosinophil homeostasis. Nature. 2013; 502:245–248. [PubMed: 24037376]
36. Horiuchi S, et al. Genome-wide analysis reveals unique regulation of transcription of Th2-specific genes by GATA3. J Immunol. 2011; 186:6378–6389. [PubMed: 21536806]
37. Licona-Limon P, Kim LK, Palm NW, Flavell RA. TH2, allergy and group 2 innate lymphoid cells. Nature immunology. 2013; 14:536–542. [PubMed: 23685824]
38. Yagi R, et al. The transcription factor GATA3 is critical for the development of all IL-7Ralpha-expressing innate lymphoid cells. Immunity. 2014; 40:378–388. [PubMed: 24631153]
39. Voehringer D, Reese TA, Huang X, Shinkai K, Locksley RM. Type 2 immunity is controlled by IL-4/IL-13 expression in hematopoietic non-eosinophil cells of the innate immune system. The Journal of experimental medicine. 2006; 203:1435–1446. [PubMed: 16702603]
40. Kurata H, Lee HJ, O'Garra A, Arai N. Ectopic expression of activated Stat6 induces the expression of Th2-specific cytokines and transcription factors in developing Th1 cells. Immunity. 1999; 11:677–688. [PubMed: 10626890]
41. Zhu J, Guo L, Watson CJ, Hu-Li J, Paul WE. Stat6 is necessary and sufficient for IL-4's role in Th2 differentiation and cell expansion. J Immunol. 2001; 166:7276–7281. [PubMed: 11390477]
42. Yoshimoto T, Paul WE. CD4pos, NK1.1pos T cells promptly produce interleukin 4 in response to in vivo challenge with anti-CD3. The Journal of experimental medicine. 1994; 179:1285–1295. [PubMed: 7908323]
43. Sokol CL, Barton GM, Farr AG, Medzhitov R. A mechanism for the initiation of allergen-induced T helper type 2 responses. Nature immunology. 2008; 9:310–318. [PubMed: 18300366]
44. Sokol CL, et al. Basophils function as antigen-presenting cells for an allergen-induced T helper type 2 response. Nature immunology. 2009; 10:713–720. [PubMed: 19465907]
45. Yamane H, Zhu J, Paul WE. Independent roles for IL-2 and GATA-3 in stimulating naive CD4+ T cells to generate a Th2-inducing cytokine environment. The Journal of experimental medicine. 2005; 202:793–804. [PubMed: 16172258]
46. Noben-Trauth N, Hu-Li J, Paul WE. IL-4 secreted from individual naive CD4+ T cells acts in an autocrine manner to induce Th2 differentiation. European journal of immunology. 2002; 32:1428–1433. [PubMed: 11981831]

47. Liang HE, et al. Divergent expression patterns of IL-4 and IL-13 define unique functions in allergic immunity. Nature immunology. 2012; 13:58–66. [PubMed: 22138715]
48. Tao X, Constant S, Jorritsma P, Bottomly K. Strength of TCR signal determines the costimulatory requirements for Th1 and Th2 CD4+ T cell differentiation. J Immunol. 1997; 159:5956–5963. [PubMed: 9550393]
49. van Panhuys N, Klauschen F, Germain RN. T-cell-receptor-dependent signal intensity dominantly controls CD4(+) T cell polarization In Vivo. Immunity. 2014; 41:63–74. [PubMed: 24981853]
50. Nelson RW, et al. T Cell Receptor Cross-Reactivity between Similar Foreign and Self Peptides Influences Naive Cell Population Size and Autoimmunity. Immunity. 2014
51. Tubo NJ, et al. Single naive CD4+ T cells from a diverse repertoire produce different effector cell types during infection. Cell. 2013; 153:785–796. [PubMed: 23663778]
52. Chen CH, Zhang DH, LaPorte JM, Ray A. Cyclic AMP activates p38 mitogen-activated protein kinase in Th2 cells: phosphorylation of GATA-3 and stimulation of Th2 cytokine gene expression. J Immunol. 2000; 165:5597–5605. [PubMed: 11067915]
53. Stumbles PA, et al. Resting respiratory tract dendritic cells preferentially stimulate T helper cell type 2 (Th2) responses and require obligatory cytokine signals for induction of Th1 immunity. The Journal of experimental medicine. 1998; 188:2019–2031. [PubMed: 9841916]
54. Steinfelder S, et al. The major component in schistosome eggs responsible for conditioning dendritic cells for Th2 polarization is a T2 ribonuclease (omega-1). The Journal of experimental medicine. 2009; 206:1681–1690. [PubMed: 19635859]
55. Everts B, et al. Omega-1, a glycoprotein secreted by Schistosoma mansoni eggs, drives Th2 responses. The Journal of experimental medicine. 2009; 206:1673–1680. [PubMed: 19635864]
56. Yoshimoto T, et al. Basophils contribute to T(H)2-IgE responses in vivo via IL-4 production and presentation of peptide-MHC class II complexes to CD4+ T cells. Nature immunology. 2009; 10:706–712. [PubMed: 19465908]
57. Perrigoue JG, et al. MHC class II-dependent basophil-CD4+ T cell interactions promote T(H)2 cytokine-dependent immunity. Nature immunology. 2009; 10:697–705. [PubMed: 19465906]
58. Seder RA, Germain RN, Linsley PS, Paul WE. CD28-mediated costimulation of interleukin 2 (IL-2) production plays a critical role in T cell priming for IL-4 and interferon gamma production. The Journal of experimental medicine. 1994; 179:299–304. [PubMed: 8270872]
59. Lenschow DJ, et al. CD28/B7 regulation of Th1 and Th2 subsets in the development of autoimmune diabetes. Immunity. 1996; 5:285–293. [PubMed: 8808683]
60. Corry DB, Reiner SL, Linsley PS, Locksley RM. Differential effects of blockade of CD28-B7 on the development of Th1 or Th2 effector cells in experimental leishmaniasis. J Immunol. 1994; 153:4142–4148. [PubMed: 7523517]
61. Oosterwegel MA, et al. The role of CTLA-4 in regulating Th2 differentiation. J Immunol. 1999; 163:2634–2639. [PubMed: 10453003]
62. Bour-Jordan H, et al. CTLA-4 regulates the requirement for cytokine-induced signals in T(H)2 lineage commitment. Nature immunology. 2003; 4:182–188. [PubMed: 12524538]
63. Nurieva RI, et al. Transcriptional regulation of th2 differentiation by inducible costimulator. Immunity. 2003; 18:801–811. [PubMed: 12818161]
64. Watanabe M, et al. ICOS-mediated costimulation on Th2 differentiation is achieved by the enhancement of IL-4 receptor-mediated signaling. J Immunol. 2005; 174:1989–1996. [PubMed: 15699127]
65. Flynn S, Toellner KM, Raykundalia C, Goodall M, Lane P. CD4 T cell cytokine differentiation: the B cell activation molecule, OX40 ligand, instructs CD4 T cells to express interleukin 4 and upregulates expression of the chemokine receptor, Blr-1. The Journal of experimental medicine. 1998; 188:297–304. [PubMed: 9670042]
66. Ohshima Y, et al. OX40 costimulation enhances interleukin-4 (IL-4) expression at priming and promotes the differentiation of naive human CD4(+) T cells into high IL-4-producing effectors. Blood. 1998; 92:3338–3345. [PubMed: 9787171]
67. Ito T, et al. TSLP-activated dendritic cells induce an inflammatory T helper type 2 cell response through OX40 ligand. The Journal of experimental medicine. 2005; 202:1213–1223. [PubMed: 16275760]

68. Pollizzi KN, Powell JD. Regulation of T cells by mTOR: the known knowns and the known unknowns. Trends Immunol. 2015; 36:13–20. [PubMed: 25522665]
69. Chi H. Regulation and function of mTOR signalling in T cell fate decisions. Nature reviews immunology. 2012; 12:325–338.
70. Li B, Tournier C, Davis RJ, Flavell RA. Regulation of IL-4 expression by the transcription factor JunB during T helper cell differentiation. The EMBO journal. 1999; 18:420–432. [PubMed: 9889198]
71. Lee K, et al. Mammalian target of rapamycin protein complex 2 regulates differentiation of Th1 and Th2 cell subsets via distinct signaling pathways. Immunity. 2010; 32:743–753. [PubMed: 20620941]
72. Delgoffe GM, et al. The kinase mTOR regulates the differentiation of helper T cells through the selective activation of signaling by mTORC1 and mTORC2. Nature immunology. 2011; 12:295–303. [PubMed: 21358638]
73. Heikamp EB, et al. The AGC kinase SGK1 regulates TH1 and TH2 differentiation downstream of the mTORC2 complex. Nature immunology. 2014; 15:457–464. [PubMed: 24705297]
74. Yang K, et al. T cell exit from quiescence and differentiation into Th2 cells depend on Raptor-mTORC1-mediated metabolic reprogramming. Immunity. 2013; 39:1043–1056. [PubMed: 24315998]
75. Cook KD, Miller J. TCR-dependent translational control of GATA-3 enhances Th2 differentiation. J Immunol. 2010; 185:3209–3216. [PubMed: 20696860]
76. Amsen D, et al. Instruction of distinct CD4 T helper cell fates by different notch ligands on antigen-presenting cells. Cell. 2004; 117:515–526. [PubMed: 15137944]
77. Tanigaki K, et al. Regulation of alphabeta/gammadelta T cell lineage commitment and peripheral T cell responses by Notch/RBP-J signaling. Immunity. 2004; 20:611–622. [PubMed: 15142529]
78. Amsen D, et al. Direct regulation of Gata3 expression determines the T helper differentiation potential of Notch. Immunity. 2007; 27:89–99. [PubMed: 17658279]
79. Laky K, Evans S, Perez-Diez A, Fowlkes BJ. Notch Signaling Regulates Antigen Sensitivity of Naive CD4(+) T Cells by Tuning Co-stimulation. Immunity. 2015; 42:80–94. [PubMed: 25607460]
80. Adler SH, et al. Notch signaling augments T cell responsiveness by enhancing CD25 expression. J Immunol. 2003; 171:2896–2903. [PubMed: 12960312]
81. Lin JX, Leonard WJ. The role of Stat5a and Stat5b in signaling by IL-2 family cytokines. Oncogene. 2000; 19:2566–2576. [PubMed: 10851055]
82. Moriggl R, et al. Stat5 is required for IL-2-induced cell cycle progression of peripheral T cells. Immunity. 1999; 10:249–259. [PubMed: 10072077]
83. Kagami S, et al. Stat5a regulates T helper cell differentiation by several distinct mechanisms. Blood. 2001; 97:2358–2365. [PubMed: 11290598]
84. Kagami S, et al. Both stat5a and stat5b are required for antigen-induced eosinophil and T-cell recruitment into the tissue. Blood. 2000; 95:1370–1377. [PubMed: 10666213]
85. Zhou B, et al. Thymic stromal lymphopoietin as a key initiator of allergic airway inflammation in mice. Nature immunology. 2005; 6:1047–1053. [PubMed: 16142237]
86. Liu YJ. Thymic stromal lymphopoietin: master switch for allergic inflammation. The Journal of experimental medicine. 2006; 203:269–273. [PubMed: 16432252]
87. Liu YJ, et al. TSLP: an epithelial cell cytokine that regulates T cell differentiation by conditioning dendritic cell maturation. Annual review of immunology. 2007; 25:193–219.
88. He R, et al. TSLP acts on infiltrating effector T cells to drive allergic skin inflammation. Proceedings of the National Academy of Sciences of the United States of America. 2008; 105:11875–11880. [PubMed: 18711124]
89. Omori M, Ziegler S. Induction of IL-4 expression in CD4(+) T cells by thymic stromal lymphopoietin. J Immunol. 2007; 178:1396–1404. [PubMed: 17237387]
90. Zhu J, et al. Transient inhibition of interleukin 4 signaling by T cell receptor ligation. The Journal of experimental medicine. 2000; 192:1125–1134. [PubMed: 11034602]

91. Afkarian M, et al. T-bet is a STAT1-induced regulator of IL-12R expression in naive CD4+ T cells. Nature immunology. 2002; 3:549–557. [PubMed: 12006974]
92. Lighvani AA, et al. T-bet is rapidly induced by interferon-gamma in lymphoid and myeloid cells. Proceedings of the National Academy of Sciences of the United States of America. 2001; 98:15137–15142. [PubMed: 11752460]
93. Martin-Fontecha A, et al. Induced recruitment of NK cells to lymph nodes provides IFN-gamma for T(H)1 priming. Nature immunology. 2004; 5:1260–1265. [PubMed: 15531883]
94. Laouar Y, Sutterwala FS, Gorelik L, Flavell RA. Transforming growth factor-beta controls T helper type 1 cell development through regulation of natural killer cell interferon-gamma. Nature immunology. 2005; 6:600–607. [PubMed: 15852008]
95. Goldszmid RS, et al. NK cell-derived interferon-gamma orchestrates cellular dynamics and the differentiation of monocytes into dendritic cells at the site of infection. Immunity. 2012; 36:1047–1059. [PubMed: 22749354]
96. Klose CS, et al. Differentiation of type 1 ILCs from a common progenitor to all helper-like innate lymphoid cell lineages. Cell. 2014; 157:340–356. [PubMed: 24725403]
97. Kaplan MH, Sun YL, Hoey T, Grusby MJ. Impaired IL-12 responses and enhanced development of Th2 cells in Stat4-deficient mice. Nature. 1996; 382:174–177. [PubMed: 8700209]
98. Thierfelder WE, et al. Requirement for Stat4 in interleukin-12-mediated responses of natural killer and T cells. Nature. 1996; 382:171–174. [PubMed: 8700208]
99. Hsieh CS, et al. Development of TH1 CD4+ T cells through IL-12 produced by Listeria-induced macrophages. Science (New York, N.Y. 1993; 260:547–549.
100. Ho IC, Tai TS, Pai SY. GATA3 and the T-cell lineage: essential functions before and after T-helper-2-cell differentiation. Nature reviews immunology. 2009; 9:125–135.
101. Wei G, et al. Genome-wide analyses of transcription factor GATA3-mediated gene regulation in distinct T cell types. Immunity. 2011; 35:299–311. [PubMed: 21867929]
102. Else KJ, Finkelman FD, Maliszewski CR, Grencis RK. Cytokine-mediated regulation of chronic intestinal helminth infection. The Journal of experimental medicine. 1994; 179:347–351. [PubMed: 8270879]
103. Szabo SJ, et al. A novel transcription factor, T-bet, directs Th1 lineage commitment. Cell. 2000; 100:655–669. [PubMed: 10761931]
104. Zhu J, et al. The transcription factor T-bet is induced by multiple pathways and prevents an endogenous Th2 cell program during Th1 cell responses. Immunity. 2012; 37:660–673. [PubMed: 23041064]
105. Jankovic D, et al. In the absence of IL-12, CD4(+) T cell responses to intracellular pathogens fail to default to a Th2 pattern and are host protective in an IL-10(−/−) setting. Immunity. 2002; 16:429–439. [PubMed: 11911827]
106. Ouyang W, et al. Inhibition of Th1 development mediated by GATA-3 through an IL-4-independent mechanism. Immunity. 1998; 9:745–755. [PubMed: 9846495]
107. Usui T, et al. T-bet regulates Th1 responses through essential effects on GATA-3 function rather than on IFNG gene acetylation and transcription. The Journal of experimental medicine. 2006; 203:755–766. [PubMed: 16520391]
108. Ouyang W, et al. Stat6-independent GATA-3 autoactivation directs IL-4-independent Th2 development and commitment. Immunity. 2000; 12:27–37. [PubMed: 10661403]
109. Zhang DH, et al. Inhibition of allergic inflammation in a murine model of asthma by expression of a dominant-negative mutant of GATA-3. Immunity. 1999; 11:473–482. [PubMed: 10549629]
110. Yagi R, Zhu J, Paul WE. An updated view on transcription factor GATA3-mediated regulation of Th1 and Th2 cell differentiation. Int Immunol. 2011; 23:415–420. [PubMed: 21632975]
111. Yamashita M, et al. Identification of a conserved GATA3 response element upstream proximal from the interleukin-13 gene locus. The Journal of biological chemistry. 2002; 277:42399–42408. [PubMed: 12205084]
112. Tanaka S, et al. The enhancer HS2 critically regulates GATA-3-mediated Il4 transcription in T(H)2 cells. Nature immunology. 2011; 12:77–85. [PubMed: 21131966]

113. Fields PE, Lee GR, Kim ST, Bartsevich VV, Flavell RA. Th2-specific chromatin remodeling and enhancer activity in the Th2 cytokine locus control region. Immunity. 2004; 21:865–876. [PubMed: 15589174]
114. Lee GR, Fields PE, Griffin TJ, Flavell RA. Regulation of the Th2 cytokine locus by a locus control region. Immunity. 2003; 19:145–153. [PubMed: 12871646]
115. Hural JA, Kwan M, Henkel G, Hock MB, Brown MA. An intron transcriptional enhancer element regulates IL-4 gene locus accessibility in mast cells. J Immunol. 2000; 165:3239–3249. [PubMed: 10975840]
116. Solymar DC, Agarwal S, Bassing CH, Alt FW, Rao A. A 3' enhancer in the IL-4 gene regulates cytokine production by Th2 cells and mast cells. Immunity. 2002; 17:41–50. [PubMed: 12150890]
117. Zhu J, Yamane H, Cote-Sierra J, Guo L, Paul WE. GATA-3 promotes Th2 responses through three different mechanisms: induction of Th2 cytokine production, selective growth of Th2 cells and inhibition of Th1 cell-specific factors. Cell research. 2006; 16:3–10. [PubMed: 16467870]
118. Liao W, et al. Priming for T helper type 2 differentiation by interleukin 2-mediated induction of interleukin 4 receptor alpha-chain expression. Nature immunology. 2008; 9:1288–1296. [PubMed: 18820682]
119. Murphy TL, Tussiwand R, Murphy KM. Specificity through cooperation: BATF-IRF interactions control immune-regulatory networks. Nature reviews immunology. 2013; 13:499–509.
120. Huber M, Lohoff M. IRF4 at the crossroads of effector T-cell fate decision. European journal of immunology. 2014; 44:1886–1895. [PubMed: 24782159]
121. Lohoff M, et al. Dysregulated T helper cell differentiation in the absence of interferon regulatory factor 4. Proceedings of the National Academy of Sciences of the United States of America. 2002; 99:11808–11812. [PubMed: 12189207]
122. Rengarajan J, et al. Interferon regulatory factor 4 (IRF4) interacts with NFATc2 to modulate interleukin 4 gene expression. The Journal of experimental medicine. 2002; 195:1003–1012. [PubMed: 11956291]
123. Brustle A, et al. The development of inflammatory T(H)-17 cells requires interferon-regulatory factor 4. Nature immunology. 2007; 8:958–966. [PubMed: 17676043]
124. Zheng Y, et al. Regulatory T-cell suppressor program co-opts transcription factor IRF4 to control T(H)2 responses. Nature. 2009; 458:351–356. [PubMed: 19182775]
125. Staudt V, et al. Interferon-regulatory factor 4 is essential for the developmental program of T helper 9 cells. Immunity. 2010; 33:192–202. [PubMed: 20674401]
126. Ciofani M, et al. A Validated Regulatory Network for Th17 Cell Specification. Cell. 2012
127. Li P, et al. BATF-JUN is critical for IRF4-mediated transcription in T cells. Nature. 2012; 490:543–546. [PubMed: 22992523]
128. Glasmacher E, et al. A genomic regulatory element that directs assembly and function of immune-specific AP-1-IRF complexes. Science (New York, N.Y. 2012; 338:975–980.
129. Xu J, et al. c-Maf regulates IL-10 expression during Th17 polarization. J Immunol. 2009; 182:6226–6236. [PubMed: 19414776]
130. Rutz S, et al. Transcription factor c-Maf mediates the TGF-beta-dependent suppression of IL-22 production in T(H)17 cells. Nature immunology. 2011; 12:1238–1245. [PubMed: 22001828]
131. Bauquet AT, et al. The costimulatory molecule ICOS regulates the expression of c-Maf and IL-21 in the development of follicular T helper cells and TH-17 cells. Nature immunology. 2009; 10:167–175. [PubMed: 19098919]
132. Kim JI, Ho IC, Grusby MJ, Glimcher LH. The transcription factor c-Maf controls the production of interleukin-4 but not other Th2 cytokines. Immunity. 1999; 10:745–751. [PubMed: 10403649]
133. Ranzani V, et al. The long intergenic noncoding RNA landscape of human lymphocytes highlights the regulation of T cell differentiation by linc-MAF-4. Nature immunology. 2015
134. Kwon SJ, et al. KLF13 cooperates with c-Maf to regulate IL-4 expression in CD4+ T cells. J Immunol. 2014; 192:5703–5709. [PubMed: 24821970]

135. Lai D, et al. KLF13 sustains thymic memory-like CD8(+) T cells in BALB/c mice by regulating IL-4-generating invariant natural killer T cells. The Journal of experimental medicine. 2011; 208:1093–1103. [PubMed: 21482696]
136. Yang XO, et al. Requirement for the basic helix-loop-helix transcription factor Dec2 in initial TH2 lineage commitment. Nature immunology. 2009; 10:1260–1266. [PubMed: 19881507]
137. Kamizono S, et al. Nfil3/E4bp4 is required for the development and maturation of NK cells in vivo. The Journal of experimental medicine. 2009; 206:2977–2986. [PubMed: 19995955]
138. Gascoyne DM, et al. The basic leucine zipper transcription factor E4BP4 is essential for natural killer cell development. Nature immunology. 2009; 10:1118–1124. [PubMed: 19749763]
139. Male V, et al. The transcription factor E4bp4/Nfil3 controls commitment to the NK lineage and directly regulates Eomes and Id2 expression. The Journal of experimental medicine. 2014; 211:635–642. [PubMed: 24663216]
140. Geiger TL, et al. Nfil3 is crucial for development of innate lymphoid cells and host protection against intestinal pathogens. The Journal of experimental medicine. 2014; 211:1723–1731. [PubMed: 25113970]
141. Seillet C, et al. Nfil3 is required for the development of all innate lymphoid cell subsets. The Journal of experimental medicine. 2014; 211:1733–1740. [PubMed: 25092873]
142. Yu X, et al. The basic leucine zipper transcription factor NFIL3 directs the development of a common innate lymphoid cell precursor. eLife. 2014; 3
143. Motomura Y, et al. The transcription factor E4BP4 regulates the production of IL-10 and IL-13 in CD4+ T cells. Nature immunology. 2011; 12:450–459. [PubMed: 21460847]
144. Hwang SS, et al. Transcription factor YY1 is essential for regulation of the Th2 cytokine locus and for Th2 cell differentiation. Proceedings of the National Academy of Sciences of the United States of America. 2013; 110:276–281. [PubMed: 23248301]
145. Djuretic IM, et al. Transcription factors T-bet and Runx3 cooperate to activate Ifng and silence Il4 in T helper type 1 cells. Nature immunology. 2007; 8:145–153. [PubMed: 17195845]
146. Yagi R, et al. The transcription factor GATA3 actively represses RUNX3 protein-regulated production of interferon-gamma. Immunity. 2010; 32:507–517. [PubMed: 20399120]
147. Naoe Y, et al. Repression of interleukin-4 in T helper type 1 cells by Runx/Cbf beta binding to the Il4 silencer. The Journal of experimental medicine. 2007; 204:1749–1755. [PubMed: 17646405]
148. Endo Y, et al. Eomesodermin controls interleukin-5 production in memory T helper 2 cells through inhibition of activity of the transcription factor GATA3. Immunity. 2011; 35:733–745. [PubMed: 22118525]
149. Kuwahara M, et al. The transcription factor Sox4 is a downstream target of signaling by the cytokine TGF-beta and suppresses T(H)2 differentiation. Nature immunology. 2012; 13:778–786. [PubMed: 22751141]
150. Chang HC, et al. PU.1 expression delineates heterogeneity in primary Th2 cells. Immunity. 2005; 22:693–703. [PubMed: 15963784]
151. Wohlfert EA, et al. GATA3 controls Foxp3(+) regulatory T cell fate during inflammation in mice. The Journal of clinical investigation. 2011; 121:4503–4515. [PubMed: 21965331]
152. Wang Y, Su MA, Wan YY. An essential role of the transcription factor GATA-3 for the function of regulatory T cells. Immunity. 2011; 35:337–348. [PubMed: 21924928]
153. Rudra D, et al. Transcription factor Foxp3 and its protein partners form a complex regulatory network. Nature immunology. 2012; 13:1010–1019. [PubMed: 22922362]
154. Yu F, Sharma S, Edwards J, Feigenbaum L, Zhu J. Dynamic expression of transcription factors T-bet and GATA-3 by regulatory T cells maintains immunotolerance. Nature immunology. 2015; 16:197–206. [PubMed: 25501630]
155. Wan YSY, Flavell RA. Regulatory T-cell functions are subverted and converted owing to attenuated Foxp3 expression. Nature. 2007; 445:766–770. [PubMed: 17220876]
156. Roncador G, et al. Analysis of FOXP3 protein expression in human CD4+CD25+ regulatory T cells at the single-cell level. European journal of immunology. 2005; 35:1681–1691. [PubMed: 15902688]

157. Tran DQ, Ramsey H, Shevach EM. Induction of FOXP3 expression in naive human CD4+FOXP3 T cells by T-cell receptor stimulation is transforming growth factor-beta dependent but does not confer a regulatory phenotype. Blood. 2007; 110:2983–2990. [PubMed: 17644734]
158. Hosoya-Ohmura S, et al. An NK and T cell enhancer lies 280 kilobase pairs 3' to the gata3 structural gene. Mol Cell Biol. 2011; 31:1894–1904. [PubMed: 21383068]
159. Hu G, et al. Expression and regulation of intergenic long noncoding RNAs during T cell development and differentiation. Nature immunology. 2013; 14:1190–1198. [PubMed: 24056746]
160. Kim HP, Kelly J, Leonard WJ. The basis for IL-2-induced IL-2 receptor alpha chain gene regulation: importance of two widely separated IL-2 response elements. Immunity. 2001; 15:159–172. [PubMed: 11485747]
161. Vrisekoop N, Monteiro JP, Mandl JN, Germain RN. Revisiting thymic positive selection and the mature T cell repertoire for antigen. Immunity. 2014; 41:181–190. [PubMed: 25148022]
162. Usui T, Nishikomori R, Kitani A, Strober W. GATA-3 suppresses Th1 development by downregulation of Stat4 and not through effects on IL-12Rbeta2 chain or T-bet. Immunity. 2003; 18:415–428. [PubMed: 12648458]
163. Pearce EL, et al. Control of effector CD8+ T cell function by the transcription factor Eomesodermin. Science (New York, N.Y. 2003; 302:1041–1043.
164. Hwang ES, Szabo SJ, Schwartzberg PL, Glimcher LH. T helper cell fate specified by kinase-mediated interaction of T-bet with GATA-3. Science (New York, N.Y. 2005; 307:430–433.
165. Peine M, et al. Stable T-bet(+)GATA-3(+) Th1/Th2 hybrid cells arise in vivo, can develop directly from naive precursors, and limit immunopathologic inflammation. PLoS biology. 2013; 11:e1001633. [PubMed: 23976880]
166. Kanhere A, et al. T-bet and GATA3 orchestrate Th1 and Th2 differentiation through lineage-specific targeting of distal regulatory elements. Nature communications. 2012; 3:1268.
167. Laurence A, et al. Interleukin-2 signaling via STAT5 constrains T helper 17 cell generation. Immunity. 2007; 26:371–381. [PubMed: 17363300]
168. Johnston RJ, Choi YS, Diamond JA, Yang JA, Crotty S. STAT5 is a potent negative regulator of TFH cell differentiation. The Journal of experimental medicine. 2012; 209:243–250. [PubMed: 22271576]
169. Ballesteros-Tato A, et al. Interleukin-2 inhibits germinal center formation by limiting T follicular helper cell differentiation. Immunity. 2012; 36:847–856. [PubMed: 22464171]
170. Liao W, Lin JX, Leonard WJ. Interleukin-2 at the crossroads of effector responses, tolerance, and immunotherapy. Immunity. 2013; 38:13–25. [PubMed: 23352221]
171. Yamane H, Paul WE. Cytokines of the gamma(c) family control CD4+ T cell differentiation and function. Nature immunology. 2012; 13:1037–1044. [PubMed: 23080204]
172. Zhu J, Jankovic D, Grinberg A, Guo L, Paul WE. Gfi-1 plays an important role in IL-2-mediated Th2 cell expansion. Proceedings of the National Academy of Sciences of the United States of America. 2006; 103:18214–18219. [PubMed: 17116877]
173. Zhu J, et al. Down-regulation of Gfi-1 expression by TGF-beta is important for differentiation of Th17 and CD103+ inducible regulatory T cells. The Journal of experimental medicine. 2009; 206:329–341. [PubMed: 19188499]
174. Zhu J, et al. Growth factor independent-1 induced by IL-4 regulates Th2 cell proliferation. Immunity. 2002; 16:733–744. [PubMed: 12049724]
175. Szabo SJ, Dighe AS, Gubler U, Murphy KM. Regulation of the interleukin (IL)-12R beta 2 subunit expression in developing T helper 1 (Th1) and Th2 cells. The Journal of experimental medicine. 1997; 185:817–824. [PubMed: 9120387]
176. Mullen AC, et al. Role of T-bet in commitment of TH1 cells before IL-12-dependent selection. Science (New York, N.Y. 2001; 292:1907–1910.
177. Renz H, Domenico J, Gelfand EW. IL-4-dependent up-regulation of IL-4 receptor expression in murine T and B cells. J Immunol. 1991; 146:3049–3055. [PubMed: 2016538]
178. Hwang ES, White IA, Ho IC. An IL-4-independent and CD25-mediated function of c-maf in promoting the production of Th2 cytokines. Proceedings of the National Academy of Sciences of the United States of America. 2002; 99:13026–13030. [PubMed: 12271139]

179. Xu D, et al. Selective expression of a stable cell surface molecule on type 2 but not type 1 helper T cells. The Journal of experimental medicine. 1998; 187:787–794. [PubMed: 9480988]

180. Lohning M, et al. T1/ST2 is preferentially expressed on murine Th2 cells, independent of interleukin 4, interleukin 5, and interleukin 10, and important for Th2 effector function. Proceedings of the National Academy of Sciences of the United States of America. 1998; 95:6930–6935. [PubMed: 9618516]

181. Townsend MJ, Fallon PG, Matthews DJ, Jolin HE, McKenzie AN. T1/ST2-deficient mice demonstrate the importance of T1/ST2 in developing primary T helper cell type 2 responses. The Journal of experimental medicine. 2000; 191:1069–1076. [PubMed: 10727469]

182. Spilianakis CG, Flavell RA. Long-range intrachromosomal interactions in the T helper type 2 cytokine locus. Nature immunology. 2004; 5:1017–1027. [PubMed: 15378057]

183. Wilson CB, Rowell E, Sekimata M. Epigenetic control of T-helper-cell differentiation. Nature reviews immunology. 2009; 9:91–105.

184. Kanno Y, Vahedi G, Hirahara K, Singleton K, O'Shea JJ. Transcriptional and epigenetic control of T helper cell specification: molecular mechanisms underlying commitment and plasticity. Annual review of immunology. 2012; 30:707–731.

185. Agarwal S, Rao A. Modulation of chromatin structure regulates cytokine gene expression during T cell differentiation. Immunity. 1998; 9:765–775. [PubMed: 9881967]

186. Henkel G, et al. A DNase I-hypersensitive site in the second intron of the murine IL-4 gene defines a mast cell-specific enhancer. J Immunol. 1992; 149:3239–3246. [PubMed: 1431102]

187. Loots GG, et al. Identification of a coordinate regulator of interleukins 4, 13, and 5 by cross-species sequence comparisons. Science (New York, N.Y. 2000; 288:136–140.

188. Mohrs M, et al. Deletion of a coordinate regulator of type 2 cytokine expression in mice. Nature immunology. 2001; 2:842–847. [PubMed: 11526400]

189. Kim ST, Fields PE, Flavell RA. Demethylation of a specific hypersensitive site in the Th2 locus control region. Proceedings of the National Academy of Sciences of the United States of America. 2007; 104:17052–17057. [PubMed: 17940027]

190. Ansel KM, et al. Deletion of a conserved Il4 silencer impairs T helper type 1-mediated immunity. Nature immunology. 2004; 5:1251–1259. [PubMed: 15516924]

191. Lee GR, Spilianakis CG, Flavell RA. Hypersensitive site 7 of the TH2 locus control region is essential for expressing TH2 cytokine genes and for long-range intrachromosomal interactions. Nature immunology. 2005; 6:42–48. [PubMed: 15608641]

192. Williams A, et al. Hypersensitive site 6 of the Th2 locus control region is essential for Th2 cytokine expression. Proceedings of the National Academy of Sciences of the United States of America. 2013; 110:6955–6960. [PubMed: 23569250]

193. Koh BH, et al. Th2 LCR is essential for regulation of Th2 cytokine genes and for pathogenesis of allergic asthma. Proceedings of the National Academy of Sciences of the United States of America. 2010; 107:10614–10619. [PubMed: 20483988]

194. Barski A, et al. High-resolution profiling of histone methylations in the human genome. Cell. 2007; 129:823–837. [PubMed: 17512414]

195. Wei G, et al. Global mapping of H3K4me3 and H3K27me3 reveals specificity and plasticity in lineage fate determination of differentiating CD4+ T cells. Immunity. 2009; 30:155–167. [PubMed: 19144320]

196. Yamashita M, et al. Crucial role of MLL for the maintenance of memory T helper type 2 cell responses. Immunity. 2006; 24:611–622. [PubMed: 16713978]

197. Koyanagi M, et al. EZH2 and histone 3 trimethyl lysine 27 associated with Il4 and Il13 gene silencing in Th1 cells. The Journal of biological chemistry. 2005; 280:31470–31477. [PubMed: 16009709]

198. Tumes DJ, et al. The polycomb protein Ezh2 regulates differentiation and plasticity of CD4(+) T helper type 1 and type 2 cells. Immunity. 2013; 39:819–832. [PubMed: 24238339]

199. Onodera A, et al. STAT6-mediated displacement of polycomb by trithorax complex establishes long-term maintenance of GATA3 expression in T helper type 2 cells. The Journal of experimental medicine. 2010; 207:2493–2506. [PubMed: 20956546]

200. Avni O, et al. T-H cell differentiation is accompanied by dynamic changes in histone acetylation of cytokine genes. Nature immunology. 2002; 3:643–651. [PubMed: 12055628]
201. Siegel MD, Zhang DH, Ray P, Ray A. Activation of the interleukin-5 promoter by cAMP in murine EL-4 cells requires the GATA-3 and CLE0 elements. The Journal of biological chemistry. 1995; 270:24548–24555. [PubMed: 7592673]
202. Kishikawa H, Sun J, Choi A, Miaw SC, Ho IC. The cell type-specific expression of the murine IL-13 gene is regulated by GATA-3. J Immunol. 2001; 167:4414–4420. [PubMed: 11591766]
203. Seki N, et al. IL-4-induced GATA-3 expression is a time-restricted instruction switch for Th2 cell differentiation. J Immunol. 2004; 172:6158–6166. [PubMed: 15128803]
204. Lee HJ, et al. GATA-3 induces T helper cell type 2 (Th2) cytokine expression and chromatin remodeling in committed Th1 cells. The Journal of experimental medicine. 2000; 192:105–115. [PubMed: 10880531]
205. Lee DU, Agarwal S, Rao A. Th2 lineage commitment and efficient IL-4 production involves extended demethylation of the IL-4 gene. Immunity. 2002; 16:649–660. [PubMed: 12049717]
206. Makar KW, et al. Active recruitment of DNA methyltransferases regulates interleukin 4 in thymocytes and T cells. Nature immunology. 2003; 4:1183–1190. [PubMed: 14595437]
207. Hutchins AS, et al. Gene silencing quantitatively controls the function of a developmental trans-activator. Molecular cell. 2002; 10:81–91. [PubMed: 12150909]
208. Rao A, Luo C, Hogan PG. Transcription factors of the NFAT family: regulation and function. Annual review of immunology. 1997; 15:707–747.
209. Agarwal S, Avni O, Rao A. Cell-type-restricted binding of the transcription factor NFAT to a distal IL-4 enhancer in vivo. Immunity. 2000; 12:643–652. [PubMed: 10894164]
210. Rengarajan J, Tang B, Glimcher LH. NFATc2 and NFATc3 regulate T(H)2 differentiation and modulate TCR-responsiveness of naive T(H)cells. Nature immunology. 2002; 3:48–54. [PubMed: 11740499]
211. Robinson D, et al. IGIF does not drive Th1 development but synergizes with IL-12 for interferon-gamma production and activates IRAK and NFkappaB. Immunity. 1997; 7:571–581. [PubMed: 9354477]
212. Yang J, Zhu H, Murphy TL, Ouyang W, Murphy KM. IL-18-stimulated GADD45 beta required in cytokine-induced, but not TCR-induced, IFN-gamma production. Nature immunology. 2001; 2:157–164. [PubMed: 11175814]
213. Parmentier CN, et al. Human T(H)2 cells respond to cysteinyl leukotrienes through selective expression of cysteinyl leukotriene receptor 1. J Allergy Clin Immunol. 2012; 129:1136–1142. [PubMed: 22391114]
214. Xue L, et al. Leukotriene E4 activates human Th2 cells for exaggerated proinflammatory cytokine production in response to prostaglandin D2. J Immunol. 2012; 188:694–702. [PubMed: 22174450]
215. Guo L, Hu-Li J, Paul WE. Probabilistic regulation in TH2 cells accounts for monoallelic expression of IL-4 and IL-13. Immunity. 2005; 23:89–99. [PubMed: 16039582]
216. Guo L, et al. In TH2 cells the Il4 gene has a series of accessibility states associated with distinctive probabilities of IL-4 production. Proceedings of the National Academy of Sciences of the United States of America. 2002; 99:10623–10628. [PubMed: 12149469]
217. Guo L, Hu-Li J, Paul WE. Probabilistic regulation of IL-4 production in Th2 cells: accessibility at the Il4 locus. Immunity. 2004; 20:193–203. [PubMed: 14975241]
218. Kock J, et al. Nuclear factor of activated T cells regulates the expression of interleukin-4 in Th2 cells in an all-or-none fashion. The Journal of biological chemistry. 2014; 289:26752–26761. [PubMed: 25037220]
219. Helmstetter C, et al. Individual T helper cells have a quantitative cytokine memory. Immunity. 2015; 42:108–122. [PubMed: 25607461]
220. Reinhardt RL, Liang HE, Locksley RM. Cytokine-secreting follicular T cells shape the antibody repertoire. Nature immunology. 2009; 10:385–393. [PubMed: 19252490]
221. Zaretsky AG, et al. T follicular helper cells differentiate from Th2 cells in response to helminth antigens. The Journal of experimental medicine. 2009; 206:991–999. [PubMed: 19380637]

222. King IL, Mohrs M. IL-4-producing CD4+ T cells in reactive lymph nodes during helminth infection are T follicular helper cells. The Journal of experimental medicine. 2009; 206:1001–1007. [PubMed: 19380638]

223. Yusuf I, et al. Germinal center T follicular helper cell IL-4 production is dependent on signaling lymphocytic activation molecule receptor (CD150). J Immunol. 2010; 185:190–202. [PubMed: 20525889]

224. Nurieva RI, et al. STAT5 protein negatively regulates T follicular helper (Tfh) cell generation and function. The Journal of biological chemistry. 2012; 287:11234–11239. [PubMed: 22318729]

225. Harada Y, et al. The 3' enhancer CNS2 is a critical regulator of interleukin-4-mediated humoral immunity in follicular helper T cells. Immunity. 2012; 36:188–200. [PubMed: 22365664]

226. Vijayanand P, et al. Interleukin-4 production by follicular helper T cells requires the conserved Il4 enhancer hypersensitivity site V. Immunity. 2012; 36:175–187. [PubMed: 22326582]

227. Lu KT, et al. Functional and epigenetic studies reveal multistep differentiation and plasticity of in vitro-generated and in vivo-derived follicular T helper cells. Immunity. 2011; 35:622–632. [PubMed: 22018472]

228. Nakayamada S, et al. Early Th1 cell differentiation is marked by a Tfh celllike transition. Immunity. 2011; 35:919–931. [PubMed: 22195747]

229. Luthje K, et al. The development and fate of follicular helper T cells defined by an IL-21 reporter mouse. Nature immunology. 2012; 13:491–498. [PubMed: 22466669]

230. Moro K, et al. Innate production of T(H)2 cytokines by adipose tissue-associated c-Kit(+)Sca-1(+) lymphoid cells. Nature. 2010; 463:540–544. [PubMed: 20023630]

231. Neill DR, et al. Nuocytes represent a new innate effector leukocyte that mediates type-2 immunity. Nature. 2010; 464:1367–1370. [PubMed: 20200518]

232. Price AE, et al. Systemically dispersed innate IL-13-expressing cells in type 2 immunity. Proceedings of the National Academy of Sciences of the United States of America. 2010; 107:11489–11494. [PubMed: 20534524]

233. Fallon PG, et al. Identification of an interleukin (IL)-25-dependent cell population that provides IL-4, IL-5, and IL-13 at the onset of helminth expulsion. The Journal of experimental medicine. 2006; 203:1105–1116. [PubMed: 16606668]

234. Halim TY, et al. Retinoic-acid-receptor-related orphan nuclear receptor alpha is required for natural helper cell development and allergic inflammation. Immunity. 2012; 37:463–474. [PubMed: 22981535]

235. Wong SH, et al. Transcription factor RORalpha is critical for nuocyte development. Nature immunology. 2012; 13:229–236. [PubMed: 22267218]

236. Hoyler T, et al. The transcription factor GATA-3 controls cell fate and maintenance of type 2 innate lymphoid cells. Immunity. 2012; 37:634–648. [PubMed: 23063333]

237. Mjosberg J, et al. The transcription factor GATA3 is essential for the function of human type 2 innate lymphoid cells. Immunity. 2012; 37:649–659. [PubMed: 23063330]

238. Klein Wolterink RG, et al. Essential, dose-dependent role for the transcription factor Gata3 in the development of IL-5+ and IL-13+ type 2 innate lymphoid cells. Proceedings of the National Academy of Sciences of the United States of America. 2013; 110:10240–10245. [PubMed: 23733962]

239. Yang Q, et al. T cell factor 1 is required for group 2 innate lymphoid cell generation. Immunity. 2013; 38:694–704. [PubMed: 23601684]

240. Furusawa J, et al. Critical role of p38 and GATA3 in natural helper cell function. J Immunol. 2013; 191:1818–1826. [PubMed: 23851685]

241. Spooner CJ, et al. Specification of type 2 innate lymphocytes by the transcriptional determinant Gfi1. Nature immunology. 2013; 14:1229–1236. [PubMed: 24141388]

242. Chang YJ, et al. Innate lymphoid cells mediate influenza-induced airway hyper-reactivity independently of adaptive immunity. Nature immunology. 2011; 12:631–638. [PubMed: 21623379]

243. Halim TY, Krauss RH, Sun AC, Takei F. Lung natural helper cells are a critical source of Th2 cell-type cytokines in protease allergen-induced airway inflammation. Immunity. 2012; 36:451–463. [PubMed: 22425247]

244. Monticelli LA, et al. Innate lymphoid cells promote lung-tissue homeostasis after infection with influenza virus. Nature immunology. 2011; 12:1045–1054. [PubMed: 21946417]

245. Kim BS, et al. Basophils promote innate lymphoid cell responses in inflamed skin. J Immunol. 2014; 193:3717–3725. [PubMed: 25156365]

246. Roediger B, et al. Cutaneous immunosurveillance and regulation of inflammation by group 2 innate lymphoid cells. Nature immunology. 2013; 14:564–573. [PubMed: 23603794]

247. Brestoff JR, et al. Group 2 innate lymphoid cells promote beiging of white adipose tissue and limit obesity. Nature. 2014

248. Lee MW, et al. Activated type 2 innate lymphoid cells regulate beige fat biogenesis. Cell. 2015; 160:74–87. [PubMed: 25543153]

249. Wilhelm C, et al. An IL-9 fate reporter demonstrates the induction of an innate IL-9 response in lung inflammation. Nature immunology. 2011; 12:1071–1077. [PubMed: 21983833]

250. Turner JE, et al. IL-9-mediated survival of type 2 innate lymphoid cells promotes damage control in helminth-induced lung inflammation. The Journal of experimental medicine. 2013; 210:2951–2965. [PubMed: 24249111]

251. Lambrecht BN, Hammad H. The immunology of asthma. Nature immunology. 2015; 16:45–56. [PubMed: 25521684]

252. Doherty TA, et al. Lung type 2 innate lymphoid cells express cysteinyl leukotriene receptor 1, which regulates TH2 cytokine production. J Allergy Clin Immunol. 2013; 132:205–213. [PubMed: 23688412]

253. Xue L, et al. Prostaglandin D2 activates group 2 innate lymphoid cells through chemoattractant receptor-homologous molecule expressed on TH2 cells. J Allergy Clin Immunol. 2014; 133:1184–1194. [PubMed: 24388011]

254. Motomura Y, et al. Basophil-derived interleukin-4 controls the function of natural helper cells, a member of ILC2s, in lung inflammation. Immunity. 2014; 40:758–771. [PubMed: 24837103]

255. Barnig C, et al. Lipoxin A4 regulates natural killer cell and type 2 innate lymphoid cell activation in asthma. Science translational medicine. 2013; 5:174ra126.

256. Salimi M, et al. A role for IL-25 and IL-33-driven type-2 innate lymphoid cells in atopic dermatitis. The Journal of experimental medicine. 2013; 210:2939–2950. [PubMed: 24323357]

257. Oliphant CJ, et al. MHCII-mediated dialog between group 2 innate lymphoid cells and CD4(+) T cells potentiates type 2 immunity and promotes parasitic helminth expulsion. Immunity. 2014; 41:283–295. [PubMed: 25088770]

258. Mirchandani AS, et al. Type 2 innate lymphoid cells drive CD4+ Th2 cell responses. J Immunol. 2014; 192:2442–2448. [PubMed: 24470502]

259. Drake LY, Iijima K, Kita H. Group 2 innate lymphoid cells and CD4+ T cells cooperate to mediate type 2 immune response in mice. Allergy. 2014; 69:1300–1307. [PubMed: 24939388]

260. Halim TY, et al. Group 2 innate lymphoid cells are critical for the initiation of adaptive T helper 2 cell-mediated allergic lung inflammation. Immunity. 2014; 40:425–435. [PubMed: 24613091]

261. Van Gool F, et al. Interleukin-5-producing group 2 innate lymphoid cells control eosinophilia induced by interleukin-2 therapy. Blood. 2014; 124:3572–3576. [PubMed: 25323825]

262. Fort MM, et al. IL-25 induces IL-4, IL-5, and IL-13 and Th2-associated pathologies in vivo. Immunity. 2001; 15:985–995. [PubMed: 11754819]

263. Schmitz J, et al. IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines. Immunity. 2005; 23:479–490. [PubMed: 16286016]

264. Paul WE, Zhu J. How are T(H)2-type immune responses initiated and amplified? Nature reviews immunology. 2010; 10:225–235.

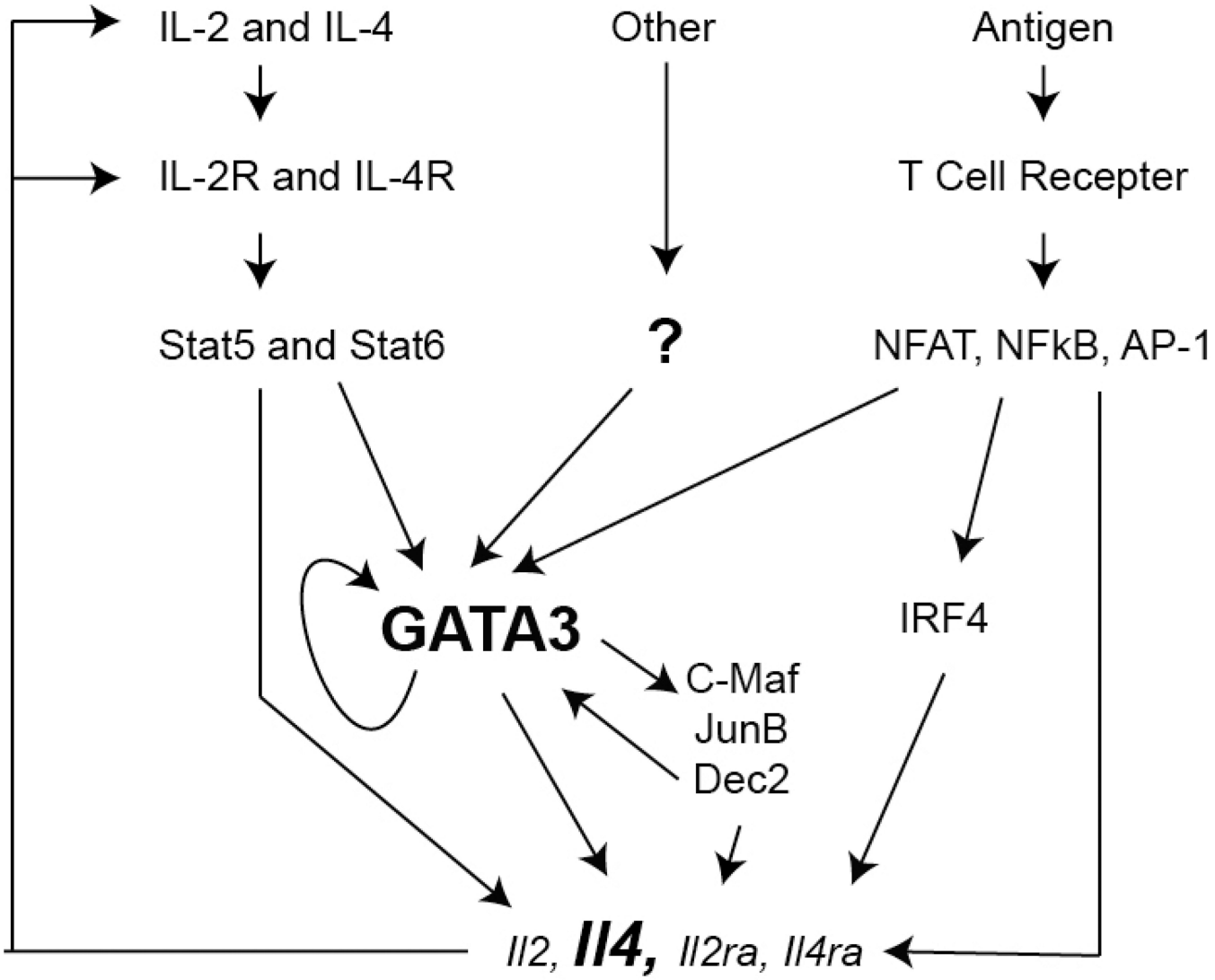

Figure 1. Transcriptional network and positive feedback regulation during Th2 cell differentiation

TCR activation and cytokine-mediated signaling are critical during Th2 cell differentiation. TCR stimulation activates NFAT, NFκb and AP-1 family members, resulting in up-regulation of IRF4 expression, which has a general function in T cell activation. Low dose of antigen stimulation accompanied by the up-regulation of Th2 master regulator GATA3 favors Th2 cell differentiation. IL-4-mediated Stat6 activation and other signaling pathways such as Notch-mediated signaling are also capable of inducing GATA3 expression. GATA3 directly mediates epigenetic modifications at the Th2 cytokine locus and cytokine transcription. GATA3 also indirectly regulates Th2 cytokine expression by inducing other transcription factor some of which may further up-regulate GATA3 expression. GATA3 also regulates its own expression. IL-2-mediated Stat5 activation is another key event for Th2 cytokine production. Activated T cells are able to produce both IL-2 and IL-4, and to up-regulate IL-2 and IL-4 receptors, forming a powerful positive feedback loop.

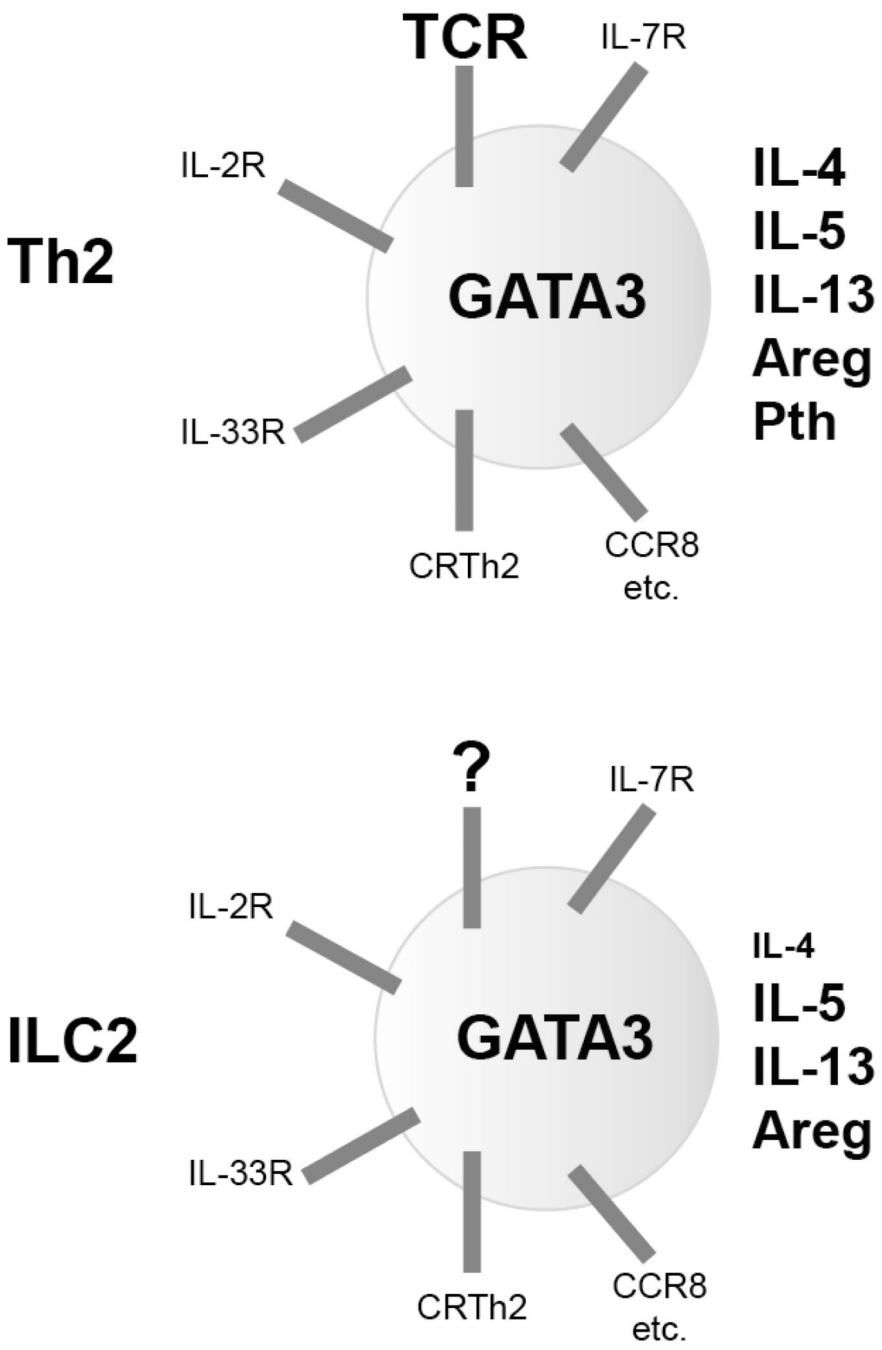

Figure 2. Similarity between Th2 cells and ILC2s

Both Th2 cells and ILC2s are capable of producing a set of cytokines such as IL-5, IL-13 and Amphiregulin (Areg), although ILC2s produce less IL-4 and no parathyroid hormone (Pth). GATA3 is critical for the development, maintenance and functions of both Th2 cells and ILC2s. While ILC2s lack T cell receptor (TCR), they express cytokine receptors found on Th2 cells including IL-33 receptor (T1/ST2), IL-2 receptor and IL-7 receptor. Both Th2 cells and ILC2s produce IL-5 and IL-13 when stimulated through IL-33 and a Stat5 activator such as IL-2 and IL-7. Th2 cells and ILC2s also share expression of specific chemokine

receptors including CRTh2, CCR8, CCR1 etc. Similar gene expression between Th2 cells and ILC2s and their dependence on GATA3 is consistent with their similar functions during type 2 immune responses.