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GATA3: A master of many trades in immune regulation

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Abstract

GATA3 has conventionally been regarded as a transcription factor that drives the differentiation of T helper (Th) 2 cells. Increasing evidence points to function for GATA3 beyond controlling Th2 differentiation. GATA3 regulates T cell development, proliferation and maintenance. Furthermore, recent studies have demonstrated important roles for GATA3 in innate lymphoid cells. Thus GATA3 emerges as a factor with diverse functions in immune regulation, which are in some cases cell-type specific and in others shared by multiple cell types. Here I discuss recent discoveries and the current understanding of the functions of GATA3 in immune regulation.

Keywords

GATA3; T cell; Treg cell; natural killer T cell; natural killer cell; innate lymphoid cell

GATA3 in innate and adaptive immunity

The immune system is constantly challenged by pathogens such as bacteria and viruses. Complex strategies have developed to respond to those insults through innate and adaptive immunity, while also maintaining immune homeostasis and self-tolerance. Elucidating the mechanisms that guide immune function under normal and pathological conditions could aid the design of effective therapies to treat a variety of afflictions, including infectious diseases, autoimmunity, inflammatory disorders, and some types of cancers. A central step towards understanding these mechanisms is to define the role of transcription factors that drive immune cell function.

The transcription factor GATA3 has emerged as a critical regulator of both innate and adaptive immunity. GATA3 belongs to the GATA (GATA-binding protein) family comprised of six members - GATA1 to 6 - in mammals. GATA transcription factors recognize the consensus DNA sequence (W)GATA(R) (W = A or T and R = A or G) [1, 2]. Different GATA family members are expressed in a tissue and cell-type specific fashion. Compared to other GATA family members, GATA3 is highly expressed in the

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hematopoietic compartment, and gene-targeting studies have shown an essential role for GATA3 in the development and function of T cells, B cells, CD1-restricted natural killer T (NKT) cells, natural killer (NK) cells, innate like lymphoid cells (ILCs) ([3–8] and commented in [9]). It should be noted that GATA3 also functions in non-hematopoietic cells in both embryonic and adult tissues including the adrenal glands, kidneys, central nervous system, inner ear, hair follicles, skin and breast tissue. For the purpose of this article, I will focus on the roles of GATA3 in innate and adaptive immunity.

Upon encounter with cognate antigen presented by antigen presenting cells (APCs) and depending on the inflammatory context, CD4⁺ T cells differentiate into distinct effector subsets including T helper 1 (Th1), Th2, Th17 and Th9 cells to direct discrete immune responses (reviewed in [10–13]) (Figure 1), often in combination with cytolytic CD8⁺ T cell effectors (reviewed in [14]). GATA3 function in T cells has been studied more extensively than in other cell types of adaptive immunity including B and NKT cells. Although GATA3 was originally identified as a master regulator for Th2 differentiation of CD4⁺ T cells, increasing evidence suggests that it is critical for the development, differentiation and function of other CD4⁺ T cell subsets, as well as CD8⁺ cells. While this review will discuss how GATA3 functions in B and NKT cells, it will focus on GATA3 controlled T cell function. In addition, recent studies point to roles for GATA3 in innate lymphocyte, which will also be discussed in this review. Furthermore, this article will place the findings in the context of earlier studies so as to delineate similarities and differences that provide insight into the multiple mechanisms of action of this versatile transcription factor in different immune cell types.

GATA3 integrates diverse upstream signals to control target gene expression

High levels of GATA3 expression in Th2 cells were first observed by Zheng and Flavell by a cDNA representational difference analysis that compared gene expression in Th1 and Th2 cells [15]. This study also showed that interleukin 4 (IL4), a Th2 cell polarizing cytokine, promoted GATA3 expression. Later findings revealed the importance of other upstream signals in the regulation of GATA3 expression, expanding on studies showed that GATA3 activation can occur independent of the IL4-STAT6 signal [16–18] (Figure 2A). T cell receptor (TCR) stimulation was shown to trigger GATA3 up-regulation in CD4⁺CD8⁺ DP thymocytes, as well as in mature CD4⁺ and CD8⁺ T cells [19]. IL4 and TCR promote GATA3 expression through different mechanisms: Signaling downstream of IL4, largely mediated by STAT6, enhances the transcription of GATA3 [19], while signaling through the TCR promotes the translation of GATA3 via PI3K-mTOR dependent pathways [20]. IL2 stimulation also up-regulates GATA3 expression in activated CD4⁺ T cells in a STAT5 dependent manner [21]. IL2 promotes GATA3 expression in both activated CD4⁺ and activated CD8⁺ T cells [22]. It remains to be addressed whether IL2 controls GATA3 expression through transcriptional or post-transcriptional mechanisms.

Wnt and Notch signals also regulate GATA3 expression. Activation of Wnt-Frizzles signal induces the accumulation of β -catenin in Th2 cells and promotes GATA3 transcription by recruiting Satb1, β -catenin and the histone acetyltransferase p300 to the GATA3 promoter

[23]. Abrogation of Wnt-Frizzled/ β -catenin signaling by the Wnt antagonist Dkk1 or small interfering RNA (siRNA) targeting of β -catenin leads to reduced GATA3 expression in Th2 cells [23]. Wnt-driven GATA3 expression has also been shown through the induction of TCF1 and β -catenin binding to the TCF1-binding site upstream of GATA3 exon-1b during the initiation of Th2 cell differentiation [18]. Notch signaling in activated CD4⁺ T cells promotes GATA3 transcription and Th2 differentiation independent of IL4-STAT6 signal [24–26].

In addition, GATA3 expression has been shown to diminish in Th2 cells when Notch signal is inhibited by the expression of a dominant negative form of Mastermind-like (MAML), a factor necessary for the assembly of the complex that contains Recombination signal binding protein for immunoglobulin kappa J region (Rbpj) and the cleaved intracellular domain of Notch (ICN), which triggers the transcription of target genes [26]. In addition to regulatory mechanisms at the transcriptional and translational levels, GATA3 function is also regulated through post-translational modifications. p38 phosphorylates GATA3 [27] to promote its nuclear translocation [28]. Pharmacological inhibition of p38 activity impairs the nuclear translocation [28] and transactivation activity [27] of GATA3 with reduced Th2 cytokine production in T cells [27]. In addition, pharmacological inhibition of p38 activity blocked the nuclear translocation of GATA3 in cycling long-term hematopoietic stem cells (LTHSCs) and the self-renewal of these cells [29]. Moreover, in type 2 innate lymphoid cells (ILC2), cytokine stimulation induces p38-mediated GATA3 phosphorylation and inhibition of p38 activity by pharmacological drugs blocked such phosphorylation and the production of IL6 by ILC2 [30]. GATA3 is acetylated on lysine 305 (K305) [31], but how this acetylation affects GATA3 function remains controversial: In transgenic models, expression of a hypoacetylated form of GATA3 (KRR to AAA) resulted in defects in T cell survival and homing [31], decreased Th2 cytokine production, and impaired allergic response [32]. Nonetheless, when a GATA3 with the KRR mutation is retrovirally transduced into activated CD4⁺ T cells, it is able to bind to DNA within the *Il4* locus, induce DNase hypersensitive sites, and drive the production of IL4, albeit at moderately reduced levels compared to wild type GATA3 [33]. It is possible this discrepancy is due to the difference in the timing of GATA3 (KRR) expression: It is expressed earlier in transgenic T cells (before T cell activation) than in retrovirally transduced T cells (after T cell activation). These findings nonetheless demonstrate that GATA3 function is modulated by post-translational modifications. How these and other yet-to-be-identified post-translational modifications integrate information from upstream signaling pathways in different cellular context, and the molecular mechanisms by which they impact GATA3 function are open questions.

It should be noted that while the aforementioned signaling pathways impact GATA3 function in immune cells under specific conditions, their functions extend beyond GATA3 regulation. In fact, all of the above mentioned pathways exert pleiotropic effect on T cells. Notch, for example, sensitizes CD4⁺ T cell to differentiate into multiple Th cell types including Th1, Th2 and Th17 [34]. Notch inhibition by a gamma secretase inhibitor (GSI) or by the expression of dominant negative MAML leads to impaired Th1, Th2 and Th17 differentiation with reduced expression of *Ifn γ* , *Tbx21*, *Il4*, *Gata3*, *Il17a*, and *Rorc*. Notch1

directly binds to *Il4*, *Gata3*, *Tbx21* loci under Th1, Th2 and Th17 polarizing conditions. In addition, GSI inhibits Th cell differentiation more potently in sub-optimal than in optimal Th polarization conditions [34]. These findings suggest that Notch integrates and amplifies cytokine-derived signals to sensitize the differentiation of not only Th2 but also Th1 and Th17 cells [25, 26, 34–36].

Besides being regulated by exogenous stimuli, GATA3 can initiate an auto-activation feedback loop independent of cytokine stimulation. Retrovirus mediated ectopic expression of GATA3 induced strong expression of endogenous GATA3 in both IL4/STAT6 deficient and Th1 CD4⁺ T cells [16, 37–39]. Structural and mutational analysis revealed that GATA3 may promote its own expression by acting on a T-cell-specific cis elements within the *Gata3* locus [39]. Therefore, GATA3, once highly expressed, is able to ‘lock-in’ a GATA3-promoted program to stabilize Th2 function.

Transcriptional regulation by GATA3

GATA3 controls cellular function predominantly through regulating target gene expression (Figure 2B). For example, to promote Th2 differentiation, GATA3 activates the expression of Th2 cytokines by binding directly to the *Il5* and *Il13* promoters, the intragenic regions of *Il4*, and the CGRE region within *Il13* locus [40]. To inhibit Th1 differentiation, GATA3 suppresses the expression of IL12R β 2 [41] and STAT4 [42], both of which are critical for Th1 differentiation [43, 44]. In addition, GATA3 inhibits Eomes expression and IFN- γ production by physically interacting with Runx3 [45], a transcriptional regulator that promotes Th1 differentiation [46]. To reveal targets of GATA3 in T cells, genome-wide analysis has been performed using a combination of ChIP-Seq and RNA-Seq approaches, which enable the identification of DNA sequences bound by GATA3-containing protein complexes as well as the profiling of associated RNA expression [47]. Many GATA3 binding sites, e.g. the ones within *Ctla4* and *Icos* loci, are shared by different T cell subsets including thymocytes, CD4⁺ T, CD8⁺ T, Treg cells, Th1, Th2, Th17 and NKT cells. Nonetheless, GATA3 regulates different transcriptional programs depending on the cellular context [47]. For example, GATA3 directly regulates the expression of Th-POK, Notch1, and TCR subunits specifically in the thymocytes and naïve T cell, and it controls the distinct expression of 91, 90, 7, and 43 genes in Th1, Th2, Th17, and iTreg cells respectively. GATA3 thus deploys shared and unique mechanisms to control the function of different T cell subsets. To achieve diverse function in different cell types, GATA3 associates with various co-factors including ETS, RUNX, AP1, TCF11 and FLI1 in a cell type specific manner to either directly regulate target gene expression or modify epigenetic markers, such as the methylation of the histon [47].

GATA3 in the development of T, B and NKT cells

GATA3 controls the function of both adaptive and innate immune cells (Figure 3). The involvement of GATA3 in adaptive immunity, esp. T cell function, has been studied extensively. Early studies in human cells revealed that GATA3 is expressed by early T cell progenitors, and that it binds the human TCR- α enhancer [48], suggesting a role in T cell development. GATA3 was deleted in mice using gene targeting approaches, but these mice

die during early embryonic development (day 12) [49], precluding an assessment of the role of GATA3 in lymphocyte development in these mice. To study GATA3 function in T cells, chimeric mice were created by supplementing GATA3-deficient embryonic stem cells into immuno-deficient Rag2 null blastocysts. GATA3 deficient T cells failed to develop in the chimeric mice, whereas B cells developed normally, indicating a requirement for GATA3 in T cell development [49]. In this study, it was difficult to rule out an effect of GATA3^{-/-} non-lymphoid tissues on T cell lineage commitment because GATA3 null embryonic stem cells contributed to the formation of nonlymphoid tissues including those in the thymus. In subsequent studies, T and B cell development was evaluated in bone marrow chimeric mice reconstituted with GATA3 null fetal liver cells and in a conditional gene targeted model that enabled the deletion of GATA3 specifically in hematopoietic cells [3, 50]. GATA3 deficiency resulted in failed development of T but not B cells in these models [3, 50]. Furthermore, studies in a mouse model engineered to have green fluorescent protein (GFP) under the control of the endogenous GATA3 promoter revealed that GATA3 is expressed in Lin^{lo}c-Kit^{hi}CD25⁻ early T cell progenitors (ETPs). The generation of ETPs is impaired when GATA3 expression is abated by targeted deletion or an engineered hypomorphism [50]. Therefore, GATA3 is required for T cell lineage commitment, but not for B cell lineage commitment. In fact, GATA3 actively suppresses B cell development. EBF1, a factor critical for B cell development (reviewed in [51]), suppresses GATA3 expression. Blockage of EBF1-mediated suppression of GATA3 leads to elevated GATA3 expression and defective B cell development, which can be rescued by GATA3 deletion [52]. In addition, inducible GATA3 deletion in DN2 thymocytes results in B cell development [53]. Therefore, GATA3 promotes T cell development but suppresses B cell development. These reciprocal functions of GATA3 in T and B cell development appear to be evolutionarily conserved: orthologues of GATA3 are preferentially expressed in T- but not B-cell-like cells in jawless vertebrates, such as lampreys [54].

While the functions of GATA-3 in T cell lineage commitment are not fully understood, GATA3 is suggested to mediate Notch signaling that is important for both promoting T cell development and inhibiting B cell development (reviewed in [55]). Fetal liver precursors cultured with stromal cells expressing Delta-like-1, a Notch ligand, develop into T cells with enhanced GATA3 mRNA expression [56, 57], and this up-regulation was reversed upon Notch ligand removal [57]. TCF1 was reported to control GATA3 expression downstream of Notch signaling during early T cell development. Notch activation induced TCF1 expression, which in turn increased transcription of GATA3 and T cell development independent of Notch signal [58]. While TCF1 promotes GATA3 expression by binding to the regulatory sites upstream of GATA3 exon 1b in Th2 cells [18], whether GATA3 is a direct transcriptional target of the TCF1 during early T cell development remains to be addressed. Activation of Notch signaling by introducing ICN into fetal liver precursors is not sufficient to promote T cell development when GATA3 is deficient. Nonetheless, ICN expression in fetal liver precursors effectively inhibited B cell development when GATA3 was absent [59]. Likewise, while GATA3-deficient DN2 thymocytes could develop into B cells in the absence of Notch signaling, Notch activation prevented B cell development of GATA3-deficient DN2 thymocytes [53]. Taken together, these findings suggest that

GATA3 is specifically required for Notch-driven T cell development but dispensable for Notch-mediated inhibition of B cell development.

The function of GATA3 in development extends beyond T cell lineage commitment in the thymus. In a mouse model engineered to delete GATA3 in double negative (DN) thymocytes (GATA3^{fl/fl}-LckCre mice), development of both CD4⁺ and CD8⁺ T cells is abrogated, and this defect was shown to relate to a requirement for GATA3 in TCR β chain-selection during the DN3-DN4 transition [60]. In contrast, when GATA3 is deleted at the double positive (DP) stage of thymocyte development (GATA3^{fl/fl}-CD4Cre mice), only CD4⁺ T cell development is abrogated in [22, 60–62], indicating a specific requirement for GATA3 in co-receptor expression during the DP to SP transition in developing T cells.

One of the mechanisms by which GATA3 controls co-receptor expression is by promoting the expression of ThPOK [62], a transcription factor that mediates the up-regulation of CD4 expression and the down-regulation of CD8 expression during DP to SP transition (reviewed in [63]). GATA3 binds to the ThPOK promoter suggesting that ThPOK acts downstream of GATA3 to control CD4SP thymocyte development. Transgenic expression of ThPOK in DP thymocytes, however, failed to restore the development of CD4⁺ T cells in the absence of GATA3 [62]. Two mutually non-exclusive possibilities may account for these findings: In a first scenario, GATA3 and ThPOK would function inter-dependently in a feedforward loop during CD4SP thymocyte development. Alternatively, GATA3 may also control processes relevant to expression of CD4 and CD8 in a ThPOK independent manner. Evidence exists to support the latter: Enhanced GATA3 expression increased the numbers of TCR^{hi}CD24^{lo} thymocytes with a CD4SP-like phenotype in a GATA3 transgenic model [64]. GATA3 was also found to repress the expression of Runx3, a factor that suppresses CD4 expression and promotes CD8 expression (reviewed in [65]), independent of ThPOK [64].

Like conventional CD4⁺ and CD8⁺ T cells, regulatory T (Treg) cells, a CD4⁺ T cell subset that is critical to suppress immune response and maintain self-tolerance (reviewed in [66, 67]), also develop in the thymus. Treg cells are characterized by expression of the X-linked transcription factor Foxp3 (reviewed in [68]). In a mouse model where GFP is knocked into endogenous *Foxp3* locus to mark Foxp3-expressing cells, Treg cells are detected in DN, DP and SP (mostly CD4SP) thymocytes [69, 70]. Like conventional CD4⁺ T cells, Foxp3⁺ Treg cells also express GATA3 [71]. To investigate the role of GATA3 in thymic development of Treg cells, several research groups deleted GATA3 specifically in Treg cells by crossing GATA3^{fl/fl} mice with mice bearing a Cre-knocked-in *Foxp3* allele [72, 73] or with mice carrying a Foxp3-Cre transgene in a bacterial artificial chromosome (BAC) [74]. The thymic generation of Treg cells in these mice is normal [72–74], indicating that unlike conventional CD4⁺ T cells, thymic development of Treg cells does not require GATA3.

Invariant NKT (iNKT) cells consist of subset of T cells expressing a semi-invariant TCR α chain (V α 14-J α 18 in the mouse and V α 24-J α 18 in humans). iNKT cells recognize and react to lipid antigens presented in the context of CD1d [75]. iNKT cells develop from DP thymocytes [76, 77], but undergo thymic selection restricted by CD1d instead of MHC [78, 79]. GATA3 is expressed in iNKT cells, and GATA3 deficiency leads to failed development of iNKT cells in a mouse model carrying a GATA3 conditional allele and Cre under the

control of the CD4 promoter (GATA3^{fl/fl}-CD4Cre) [6]. The mechanisms by which GATA3 impacts the development of iNKT cells are not clear. The transcription factors Myc and Myb may be involved, as Myc deletion and Myb deletion in CD4 cells (a Myc^{fl/fl}-CD4Cre mouse model [80] and a Myb^{fl/fl}-CD4Cre mouse model [81], respectively) results in defects in iNKT cell development similar to those observed in GATA3^{fl/fl}-CD4Cre mice. The finding that GATA3 controls Myc expression in T cells [22] implies that the GATA3-Myc signaling axis is critical in controlling iNKT cell development, a notion that warrants further investigation.

GATA3 functions in the maintenance of T cells in the periphery

In addition to controlling T cell development, GATA3 contributes to the maintenance of mature T cells in the periphery. GATA3 is expressed by mature CD8⁺ T cells in the periphery under steady state conditions, and recent studies point to a requirement for GATA3 in peripheral maintenance of these cells. CD8⁺ T cells isolated from GATA3^{fl/fl}-CD4Cre mice failed to be maintained in syngeneic hosts after adoptive transfer. In addition, CD8⁺ T cells isolated from GATA3^{fl/fl}-ERCre mice failed to be maintained in syngeneic recipients after GATA3 deletion *in vivo*; in this mouse model GATA3 is flanked by *flox* sites and Cre is expressed under the estrogen promoter (ER), which is pharmacologically induced by tamoxifen treatment. These GATA3-deficient CD8⁺ T cells were shown to express lower levels of surface IL7R α , and have associated defects in IL7-driven T cell survival [22]. GATA3 has been shown to bind to the promoter/enhancer region of *Il7r* gene in CD8⁺ T cells [22, 47], and because IL7 signal is essential for the maintenance of T cells in the periphery [82], the aforementioned findings suggest that one of the mechanisms by which GATA3 regulates CD8⁺ T cell maintenance is by regulating IL7 signaling.

GATA3 may also control the peripheral maintenance of naïve CD4⁺ T cells under steady state conditions because GATA3 expression is readily detectable in mature CD4⁺ T cells in the periphery. Failed CD4 T cell development in GATA3^{fl/fl}-CD4Cre mice however precludes the study of GATA3 function in mature CD4⁺ T cells. Nonetheless, this hypothesis may be addressed using the GATA3^{fl/fl}-ERCre model, which would allow deletion of GATA3 in mature CD4⁺ T cells in an inducible fashion [22].

The peripheral maintenance of Treg cells has different requirements from that of other CD4⁺ T cell subsets, in that Treg cells rely more on IL2 than on IL7 signaling [83–85]. GATA3 is required for the peripheral maintenance of these cells under homeostasis as well as inflammatory conditions [72–74]. IL2R α (CD25) expression on Treg cells is reduced in the absence of GATA3 [74]. Because GATA3 deletion leads to reduced Foxp3 expression in Treg cells [72–74], and Foxp3 is critical for IL2R α expression in Treg cells [86–88], a mechanism underlying the requirement of GATA3 for maintenance of Treg cells in the periphery may involve GATA3 regulation of IL2R α expression via Foxp3 up-regulation. GATA3 controls Foxp3 expression by binding to and promoting the activity of the conserved non-coding DNA sequence (CNS) 2 [72–74], a *cis*-acting element in *Foxp3* locus which is associated with sustained high levels of expression of Foxp3 [89]. GATA3 does not bind to the Foxp3 promoter, CNS1 - a site required for TGF- β -promoted Foxp3 expression, or CNS3 - a site required for Foxp3 induction during thymic development within *Foxp3* locus [89], although putative GATA3 binding sites are found in these regions. The DNA

binding pattern on *Foxp3* locus agrees with the finding that GATA3 is dispensable for the generation of Treg cells in the thymus but is required for optimal *Foxp3* expression and the peripheral maintenance of Treg cells. It should be noted that the critical function of GATA3 in Treg cells may not be mediated through *Foxp3* exclusively; GATA3 also regulates T cell function by controlling TCR- and cytokine-signaling that are important for Treg cell function.

Distinct aspects of T cell activation and proliferation require GATA3

As discussed above, GATA3 expression is exquisitely regulated by TCR and cytokine stimulation [19–22, 72, 90], which suggests that GATA3 regulates T cell function in response to these stimuli. Indeed, GATA3 deficient CD8⁺ T cells fail to grow in size (biomass increase) or to proliferate after TCR stimulation or stimulation with cytokines IL2, IL4 and IL15 [22, 91]. A similar requirement for GATA3 in CD4⁺ T cell proliferation is suggested by a study in which antisense DNA-mediated silencing of GATA3 led to reduced proliferation of a CD4⁺ T cell line [15]. In addition, GATA3-deficient Th2 cells proliferate poorly compared to wild-type cells [92]. The mechanisms by which GATA3 functions in T cell activation and proliferation are not fully understood; signaling immediately downstream of TCR ligation, such as the activation of NFκB, JNK, p38 and NFAT, is independent of GATA3 [22, 91]. Whereas the mechanisms by which GATA3 impacts cell size are not clear, the up-regulation of *Myc*, a critical regulator for the metabolism, cell size increase and proliferation of activated T cells [74, 93], was abolished in the absence of GATA3 [22]. It is plausible that the GATA3-*Myc* signaling axis is central for T cell metabolism and cell size increase, a notion that needs further investigation. This axis may also mediate GATA3's effects on T cell proliferation because GATA3 deletion led to failed up-regulation of *Myc* in proliferating T cells upon TCR activation; GATA3 was shown to bind directly to the *Myc* locus, and ectopic expression of *Myc* restored the proliferation of GATA3-deficient T cells to a similar extent as wild-type T cells [22]. *Myc* expression did not, however, fully restore the proliferation of GATA3-deficient T cells, which implies that additional GATA3-dependent pathways are involved. Indeed, the down-regulation of KLF2 [94] and TOB1 [95], transcription factors that suppress T cell proliferation following TCR activation, was impaired in GATA3-deficient T cells [22]. In addition, GATA3 regulates the proliferation of Th2 cells by associating with *Ruvbl2* [92] to suppress the expression of a cell cycle inhibitor *Cdkn2c* (p18) [92, 96]. Whether and how KLF2, TOB1 and other factors interact with GATA3 to control T cell proliferation are questions yet to be addressed.

Cell-type specific roles for GATA3 in T cell effector function

While GATA3 appears to be essential for the proliferation of both CD4⁺ and CD8⁺ T cells, its role in T cell effector function is cell-type specific. GATA3 deletion has minimal effects on IFN-γ production by CD8⁺ T cells *in vitro* when stimulated through TCR and with cytokines IL2, IL4 and IL15 [22]. In addition, GATA3-deficient CD8⁺ T cells expresses normal levels of FasL, Granzyme B and perforin when activated *in vitro* via TCR stimulation and with IL2. Accordingly, GATA3 deletion has minimal effects on the ability of CD8⁺ T cells to kill MHC-mismatched target cells *in vitro* [91]. Thus, GATA3 appears dispensable for effector function of CD8⁺ T cells. Whether GATA3 controls the effector

function of CD8⁺ T cells *in vivo* is difficult to address since few GATA3-deficient CD8⁺ T cells could be recovered *in vivo* during LCMV infection, acute GvHD response [22] or anti-tumor response [91]. Due to this same reason, it is difficult to study the role of GATA3 in the generation and function of CD8⁺ memory T cells. Nonetheless, in GATA3^{fl/fl}-CD4Cre mice and in bone marrow chimeric mice containing GATA3^{fl/fl}-CD4Cre T cells, the populations of atypical central memory (CD62L^{hi}CD44^{hi}) and effector memory (CD62L^{lo}CD44^{hi}) CD8⁺ T cells appeared decreased and increased respectively upon GATA3 deletion during homeostasis [22], which suggests a role for GATA3 in memory CD8⁺ T cells.

CD4⁺ T cells acquire distinct effector functions in the periphery by differentiating into different Th subsets that are defined by their cytokine production (Figure 1). These subsets include Th1, Th2, Th17 and Th9 cells (reviewed in [97]). GATA3 function in Th1 and Th2 cell differentiation has been studied extensively, as discussed above, and is relatively well understood. The involvement of GATA3 in the differentiation and function of other Th cell subsets is less clear. The role of GATA3 in Th17 cell differentiation has not been specifically addressed experimentally. GATA3-deficient Treg cells, however, produce elevated levels of IL17 under inflammatory conditions *in vivo* [72–74]. In addition, compared to wild-type Treg cells, GATA3-deficient Treg cells are prone to produce IL17 when activated in the presence of IL6 *in vitro* [74]. Therefore, GATA3 appears to suppress a Th17 differentiation program in Treg cells. It is unclear whether GATA3 does so directly through suppressing a *bona fide* Th17 differentiation program or indirectly through enhancing Foxp3 expression in Treg cells; indeed, both mechanisms could be at work. The observation that GATA3 binds to *Il17* and *Rorc* loci suggests that GATA3 may directly regulate the Th17 differentiation program independent of Foxp3 expression [47, 72].

Differentiation of the recently identified Th9 cells is promoted by IL4 and TGF- β [98, 99]. In line with IL4 being essential for the generation of Th9 cells, STAT6 was found to be critical for Th9 differentiation [98, 99]. IL4-STAT6 signaling induces the transcription factor IRF4 [100], which in turn promotes Th9 differentiation [101]. Compared to Th2 cells, Th9 cells express much less GATA3 mRNA but similar amounts of GATA3 protein [99, 100]. Both mRNA and protein expression of GATA3 in Th9 cells depends on STAT6 [100]. These findings suggest a potential role for GATA3 in controlling Th9 differentiation. Indeed, GATA3 deletion results in impaired Th9 differentiation *in vitro* [98]. It should be noted that GATA3 expression level during Th9 differentiation is exquisitely regulated; moderate overexpression of GATA3 in developing Th9 cells leads to Th2 differentiation [100].

GATA3 in innate lymphocytes

While much effort has been focused on the investigation of GATA3 function in cells of adaptive immunity, emerging evidence has revealed critical role for GATA3 in controlling the function of innate cells. Innate lymphoid cells (ILCs) are effectors of innate immunity that control lymphoid tissue remodeling. ILCs share common features including a lymphoid morphology, lack of recombination activating gene (RAG)-dependent rearranged antigen receptors, and lack of phenotypical markers for myeloid and dendritic cells. ILCs can be

classified into three groups based on cytokine production. Group 1 ILCs include NK cells, which produce IFN- γ ; Group 2 ILCs produce Th2-cytokines including IL5 and IL14; Group 3 ILCs produce Th17-cytokines IL17 and IL22 (Reviewed in [102]). GATA3 is expressed by certain subsets of ILCs and recent studies have shown an impact of GATA3 in the differentiation and function of these cells.

NK cells are a prototypical Group 1 ILC population that mediates early responses against viruses and cancer cells. While expressed in NK cell precursors, GATA3 was initially shown to be largely dispensable for peripheral NK cell development in the SCID mice received GATA3-deficient embryonic HSCs [5]. The same research group later found that GATA3 is highly expressed by IL7R α + thymic NK cells whose development requires IL7 signaling. By analyzing SCID mice received GATA3-deficient embryonic HSCs, they found that GATA3 is required for the development of IL7R α + thymic NK cells [103]. Thus, the GATA3-IL7R α signaling axis seems operative in both IL7R α + thymic NK cells and, as discussed above, CD8+ T cells [22]. In terms of NK cell function, GATA3-deficient NK cells display an immature phenotype based on cell-surface marker expression, fail to traffic to the liver and produce less IFN- γ , failing to mount an effective response against *Listeria monocytogenes* infection [5]. The molecular mechanisms underlying GATA3 control of NK cell function are undefined.

ILC2 cells reside at mucosal surfaces, and have been shown to play an important role in intestinal immunity against Helminth infection (e.g., *Nippostrongylus brasiliensis*) [104] and to regulate tissue repair after acute influenza virus infection [105]. Aberrant function of ILC2 may contribute to inflammation and autoimmunity (e.g., allergic airway inflammation) (reviewed in [106]). ILC2 express cell surface markers including IL7R α , IL2R α , IL33R, IL25R, Thy1, and Kit. Importantly, GATA3 is highly expressed in ILC2 [7, 8, 107], and what upstream signals promote GATA3 expression in ILC2 is a question of interest. Notch is expressed at high levels in ILC2, and is important for ILC2 development [108–110]. TCF1 has been shown to be critical for Notch-driven ILC2 generation, and to promote GATA3 expression in ILC2 cells [110, 111], suggesting a Notch-TCF1-GATA3 axis in ILC2. As mentioned above, TCF1 regulates GATA3 expression during early thymocyte development [58] as well as Th2 differentiation [18]. Thus, TCF1 control of GATA3 expression is a shared feature of T cell development, Th2 differentiation and ILC2 generation. Current evidence suggests that Notch-TCF1 signaling controls GATA3 during T cell development and ILC2 generation, whereas Wnt-TCF1 signaling controls GATA3 during Th2 differentiation. Nonetheless, a Wnt-independent, Notch-TCF1-dependent pathway may also contribute to GATA3 expression during Th2 differentiation since the inhibition of Wnt signaling results in reduced but not abolished GATA3 expression during Th2 differentiation [23]. Further investigation is needed to address whether the Notch-TCF1-GATA3 signaling axis controls all three processes of T cell development, CD4 differentiation and ILC2 function.

Expression of GATA3 is vital for ILC2 generation. Ectopic expression of GATA3 promoted surface marker expression and function of human ILC2 [7]. A partial silencing of GATA3 by shRNA in human ILC2 impaired their function [7]. In addition, deletion of GATA3 in developing ILC2 prevented ILC2 generation in mice [8]. It is, however, unclear how

GATA3 regulates the generation of ILC2. GATA3 controls ILC2 function likely through multiple mechanisms. A study of GATA3-binding sites in early thymocyte progenitors identified ROR- α as a GATA3 target [112]. Another study demonstrated that GATA3 is essential for the induction of ROR- α in fetal liver cells in the presence of Notch signal [113]. Because ROR- α is critical for ILC2 homeostasis [109, 114], one way by which GATA3 controls early ILC2 development may be through regulating ROR- α expression. Another mechanism through which GATA3 regulates ILC2 function may be through IL7R α . IL7R α is highly expressed by ILC2 and is important for ILC2 generation [115]. GATA3 was found to control IL7R α expression by binding to *Il7r* locus [22, 47]. How ROR- α , IL7R α and other factors mediate GATA3 controlled ILC2 function remains to be addressed. In addition to IL7R α + NK and ILC2 cells, hematopoietic cell-derived intestinal ILC3 cells require GATA3 because GATA3 deletion leads to defective generation and function of these cells [116]. Therefore, GATA3 regulates the function of ILC1, ILC2 and ILC3 cells, which suggests that GATA3 controls a common progenitor for these ILCs, a question to be addressed. With the recent increased interest in understanding regulation of ILC function, more mechanistic insights into the regulation and the function of GATA3 in ILCs are expected to be revealed in the near future.

Concluding Remarks

The immune function of GATA3 is multifaceted and extends beyond controlling Th2 differentiation. GATA3 is important for T cell development, homeostasis, activation, proliferation and effector functions. In addition, GATA3 controls ILC function. With the development of more sophisticated experimental tools, e.g. lineage-specific knockout mouse models, it is expected that additional functions of GATA3 in immune regulation will be unveiled in the future. More importantly, to fully appreciate the versatile functions of GATA3, we need to understand, on a molecular level, how GATA3 integrates diverse upstream signals into specific genetic programs depending on cellular- and micro-environmental contexts.

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References

1. Ko LJ, Engel JD. DNA-binding specificities of the GATA transcription factor family. *Mol Cell Biol.* 1993; 13:4011–4022. [PubMed: 8321208]
2. Merika M, Orkin SH. DNA-binding specificity of GATA family transcription factors. *Mol Cell Biol.* 1993; 13:3999–4010. [PubMed: 8321207]
3. Ku CJ, et al. GATA-3 regulates hematopoietic stem cell maintenance and cell cycle entry. *Blood.* 2012
4. Oosterwegel M, et al. Expression of GATA-3 during lymphocyte differentiation and mouse embryogenesis. *Developmental immunology.* 1992; 3:1–11. [PubMed: 1343100]
5. Samson SI, et al. GATA-3 promotes maturation, IFN-gamma production, and liver-specific homing of NK cells. *Immunity.* 2003; 19:701–711. [PubMed: 14614857]

6. Kim PJ, et al. GATA-3 regulates the development and function of invariant NKT cells. *J Immunol.* 2006; 177:6650–6659. [PubMed: 17082577]
7. 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]
8. 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]
9. Rothenberg EV. GATA-3 locks the door to the B-cell option. *Blood.* 2013; 121:1673–1674. [PubMed: 23471221]
10. Szabo SJ, et al. Molecular mechanisms regulating Th1 immune responses. *Annu Rev Immunol.* 2003; 21:713–758. [PubMed: 12500979]
11. Weaver CT, et al. IL-17 family cytokines and the expanding diversity of effector T cell lineages. *Annu Rev Immunol.* 2007; 25:821–852. [PubMed: 17201677]
12. Dong C. TH17 cells in development: an updated view of their molecular identity and genetic programming. *Nat Rev Immunol.* 2008; 8:337–348. [PubMed: 18408735]
13. Kaplan MH. Th9 cells: differentiation and disease. *Immunol Rev.* 2013; 252:104–115. [PubMed: 23405898]
14. Zhang N, Bevan MJ. CD8(+) T cells: foot soldiers of the immune system. *Immunity.* 2011; 35:161–168. [PubMed: 21867926]
15. 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]
16. Ouyang W, et al. Stat6-independent GATA-3 autoactivation directs IL-4-independent Th2 development and commitment. *Immunity.* 2000; 12:27–37. [PubMed: 10661403]
17. Yamane H, et al. Independent roles for IL-2 and GATA-3 in stimulating naive CD4+ T cells to generate a Th2-inducing cytokine environment. *J Exp Med.* 2005; 202:793–804. [PubMed: 16172258]
18. Yu Q, et al. T cell factor 1 initiates the T helper type 2 fate by inducing the transcription factor GATA-3 and repressing interferon-gamma. *Nat Immunol.* 2009; 10:992–999. [PubMed: 19648923]
19. Scheinman EJ, Avni O. Transcriptional regulation of GATA3 in T helper cells by the integrated activities of transcription factors downstream of the interleukin-4 receptor and T cell receptor. *J Biol Chem.* 2009; 284:3037–3048. [PubMed: 19056736]
20. Cook KD, Miller J. TCR-dependent translational control of GATA-3 enhances Th2 differentiation. *J Immunol.* 2010; 185:3209–3216. [PubMed: 20696860]
21. Guo L, et al. IL-1 family members and STAT activators induce cytokine production by Th2, Th17, and Th1 cells. *Proc Natl Acad Sci U S A.* 2009; 106:13463–13468. [PubMed: 19666510]
22. Wang Y, et al. GATA-3 controls the maintenance and proliferation of T cells downstream of TCR and cytokine signaling. *Nat Immunol.* 2013; 14:714–722. [PubMed: 23708251]
23. Notani D, et al. Global regulator SATB1 recruits beta-catenin and regulates T(H)2 differentiation in Wnt-dependent manner. *PLoS Biol.* 2010; 8:e1000296. [PubMed: 20126258]
24. Amsen D, et al. Direct regulation of Gata3 expression determines the T helper differentiation potential of Notch. *Immunity.* 2007; 27:89–99. [PubMed: 17658279]
25. 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]
26. Fang TC, et al. Notch directly regulates Gata3 expression during T helper 2 cell differentiation. *Immunity.* 2007; 27:100–110. [PubMed: 17658278]
27. Chen CH, et al. 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]
28. Maneechotesuwan K, et al. Regulation of Th2 cytokine genes by p38 MAPK-mediated phosphorylation of GATA-3. *J Immunol.* 2007; 178:2491–2498. [PubMed: 17277157]
29. Frelin C, et al. GATA-3 regulates the self-renewal of long-term hematopoietic stem cells. *Nat Immunol.* 2013; 14:1037–1044. [PubMed: 23974957]

30. Furusawa J, et al. Critical role of p38 and GATA3 in natural helper cell function. *J Immunol.* 2013; 191:1818–1826. [PubMed: 23851685]
31. Yamagata T, et al. Acetylation of GATA-3 affects T-cell survival and homing to secondary lymphoid organs. *Embo J.* 2000; 19:4676–4687. [PubMed: 10970860]
32. 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]
33. Takemoto N, et al. Cutting edge: the differential involvement of the N-finger of GATA-3 in chromatin remodeling and transactivation during Th2 development. *J Immunol.* 2002; 169:4103–4107. [PubMed: 12370337]
34. Bailis W, et al. Notch simultaneously orchestrates multiple helper T cell programs independently of cytokine signals. *Immunity.* 2013; 39:148–159. [PubMed: 23890069]
35. Maekawa Y, et al. Delta1-Notch3 interactions bias the functional differentiation of activated CD4+ T cells. *Immunity.* 2003; 19:549–559. [PubMed: 14563319]
36. Minter LM, et al. Inhibitors of gamma-secretase block in vivo and in vitro T helper type 1 polarization by preventing Notch upregulation of Tbx21. *Nat Immunol.* 2005; 6:680–688. [PubMed: 15991363]
37. Lee HJ, et al. GATA-3 induces T helper cell type 2 (Th2) cytokine expression and chromatin remodeling in committed Th1 cells. *J Exp Med.* 2000; 192:105–115. [PubMed: 10880531]
38. Zhou M, et al. Friend of GATA-1 represses GATA-3-dependent activity in CD4+ T cells. *J Exp Med.* 2001; 194:1461–1471. [PubMed: 11714753]
39. Ranganath S, Murphy KM. Structure and specificity of GATA proteins in Th2 development. *Mol Cell Biol.* 2001; 21:2716–2725. [PubMed: 11283251]
40. Yagi R, et al. An updated view on transcription factor GATA3-mediated regulation of Th1 and Th2 cell differentiation. *Int Immunol.* 2011; 23:415–420. [PubMed: 21632975]
41. 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]
42. Usui T, et al. 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]
43. Kaplan MH, et al. Impaired IL-12 responses and enhanced development of Th2 cells in Stat4-deficient mice. *Nature.* 1996; 382:174–177. [PubMed: 8700209]
44. 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]
45. Yagi R, et al. The transcription factor GATA3 actively represses RUNX3 protein-regulated production of interferon-gamma. *Immunity.* 2010; 32:507–517. [PubMed: 20399120]
46. Djuretic IM, et al. Transcription factors T-bet and Runx3 cooperate to activate Ifng and silence Il4 in T helper type 1 cells. *Nat Immunol.* 2007; 8:145–153. [PubMed: 17195845]
47. 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]
48. Ho IC, et al. Human GATA-3: a lineage-restricted transcription factor that regulates the expression of the T cell receptor alpha gene. *Embo J.* 1991; 10:1187–1192. [PubMed: 1827068]
49. Pandolfi PP, et al. Targeted disruption of the GATA3 gene causes severe abnormalities in the nervous system and in fetal liver haematopoiesis. *Nat Genet.* 1995; 11:40–44. [PubMed: 7550312]
50. Hosoya T, et al. GATA-3 is required for early T lineage progenitor development. *J Exp Med.* 2009; 206:2987–3000. [PubMed: 19934022]
51. Busslinger M. Transcriptional control of early B cell development. *Annu Rev Immunol.* 2004; 22:55–79. [PubMed: 15032574]
52. Banerjee A, et al. Transcriptional repression of Gata3 is essential for early B cell commitment. *Immunity.* 2013; 38:930–942. [PubMed: 23684985]
53. Garcia-Ojeda ME, et al. GATA-3 promotes T-cell specification by repressing B-cell potential in pro-T cells in mice. *Blood.* 2013; 121:1749–1759. [PubMed: 23287858]
54. Guo P, et al. Dual nature of the adaptive immune system in lampreys. *Nature.* 2009; 459:796–801. [PubMed: 19474790]

55. Radtke F, et al. Regulation of innate and adaptive immunity by Notch. *Nat Rev Immunol.* 2013; 13:427–437. [PubMed: 23665520]
56. Schmitt TM, et al. Induction of T cell development and establishment of T cell competence from embryonic stem cells differentiated in vitro. *Nat Immunol.* 2004; 5:410–417. [PubMed: 15034575]
57. Taghon TN, et al. Delayed, asynchronous, and reversible T-lineage specification induced by Notch/Delta signaling. *Genes Dev.* 2005; 19:965–978. [PubMed: 15833919]
58. Weber BN, et al. A critical role for TCF-1 in T-lineage specification and differentiation. *Nature.* 2011; 476:63–68. [PubMed: 21814277]
59. Hozumi K, et al. Notch signaling is necessary for GATA3 function in the initiation of T cell development. *Eur J Immunol.* 2008; 38:977–985. [PubMed: 18383037]
60. Pai SY, et al. Critical roles for transcription factor GATA-3 in thymocyte development. *Immunity.* 2003; 19:863–875. [PubMed: 14670303]
61. Zhu J, et al. Conditional deletion of Gata3 shows its essential function in T(H)1-T(H)2 responses. *Nat Immunol.* 2004; 5:1157–1165. [PubMed: 15475959]
62. Wang L, et al. Distinct functions for the transcription factors GATA-3 and ThPOK during intrathymic differentiation of CD4(+) T cells. *Nat Immunol.* 2008; 9:1122–1130. [PubMed: 18776904]
63. He X, et al. The role of ThPOK in control of CD4/CD8 lineage commitment. *Annu Rev Immunol.* 2010; 28:295–320. [PubMed: 20307210]
64. Xiong Y, et al. Thpok-independent repression of Runx3 by Gata3 during CD4+ T-cell differentiation in the thymus. *Eur J Immunol.* 2013; 43:918–928. [PubMed: 23310955]
65. Collins A, et al. RUNX proteins in transcription factor networks that regulate T-cell lineage choice. *Nat Rev Immunol.* 2009; 9:106–115. [PubMed: 19165227]
66. Sakaguchi S, et al. Regulatory T cells and immune tolerance. *Cell.* 2008; 133:775–787. [PubMed: 18510923]
67. Shevach EM. CD4+ CD25+ suppressor T cells: more questions than answers. *Nat Rev Immunol.* 2002; 2:389–400. [PubMed: 12093005]
68. Zheng Y, Rudensky AY. Foxp3 in control of the regulatory T cell lineage. *Nat Immunol.* 2007; 8:457–462. [PubMed: 17440451]
69. Fontenot JD, et al. Regulatory T cell lineage specification by the forkhead transcription factor foxp3. *Immunity.* 2005; 22:329–341. [PubMed: 15780990]
70. Wan YY, Flavell RA. Identifying Foxp3-expressing suppressor T cells with a bicistronic reporter. *Proc Natl Acad Sci U S A.* 2005; 102:5126–5131. [PubMed: 15795373]
71. Wang Y, et al. An intrinsic mechanism predisposes Foxp3-expressing regulatory T cells to Th2 conversion in vivo. *J Immunol.* 2010; 185:5983–5992. [PubMed: 20944002]
72. Wohlfert EA, et al. GATA3 controls Foxp3+ regulatory T cell fate during inflammation in mice. *J Clin Invest.* 2011
73. Rudra D, et al. Transcription factor Foxp3 and its protein partners form a complex regulatory network. *Nat Immunol.* 2012
74. Wang Y, et al. An Essential Role of the Transcription Factor GATA-3 for the Function of Regulatory T Cells. *Immunity.* 2011; 35:337–348. [PubMed: 21924928]
75. Kawano T, et al. CD1d-restricted and TCR-mediated activation of valpha14 NKT cells by glycosylceramides. *Science.* 1997; 278:1626–1629. [PubMed: 9374463]
76. Gapin L, et al. NKT cells derive from double-positive thymocytes that are positively selected by CD1d. *Nat Immunol.* 2001; 2:971–978. [PubMed: 11550008]
77. Benlagha K, et al. Characterization of the early stages of thymic NKT cell development. *J Exp Med.* 2005; 202:485–492. [PubMed: 16087715]
78. Bendelac A. Positive selection of mouse NK1+ T cells by CD1-expressing cortical thymocytes. *J Exp Med.* 1995; 182:2091–2096. [PubMed: 7500054]
79. Wei DG, et al. Expansion and long-range differentiation of the NKT cell lineage in mice expressing CD1d exclusively on cortical thymocytes. *J Exp Med.* 2005; 202:239–248. [PubMed: 16027237]

80. Dose M, et al. Intrathymic proliferation wave essential for V α 14⁺ natural killer T cell development depends on c-Myc. *Proc Natl Acad Sci U S A*. 2009; 106:8641–8646. [PubMed: 19423665]
81. Hu T, et al. The transcription factor c-Myb primes CD4⁺CD8⁺ immature thymocytes for selection into the iNKT lineage. *Nat Immunol*. 2010; 11:435–441. [PubMed: 20383148]
82. von Freeden-Jeffry U, et al. Lymphopenia in interleukin (IL)-7 gene-deleted mice identifies IL-7 as a nonredundant cytokine. *J Exp Med*. 1995; 181:1519–1526. [PubMed: 7699333]
83. Fontenot JD, et al. A function for interleukin 2 in Foxp3-expressing regulatory T cells. *Nat Immunol*. 2005; 6:1142–1151. [PubMed: 16227984]
84. Ma A, et al. Diverse functions of IL-2, IL-15, and IL-7 in lymphoid homeostasis. *Annu Rev Immunol*. 2006; 24:657–679. [PubMed: 16551262]
85. Mazzucchelli R, et al. Development of regulatory T cells requires IL-7R α stimulation by IL-7 or TSLP. *Blood*. 2008; 112:3283–3292. [PubMed: 18664628]
86. Fontenot JD, et al. Foxp3 programs the development and function of CD4⁺CD25⁺ regulatory T cells. *Nat Immunol*. 2003; 4:330–336. [PubMed: 12612578]
87. Gavin MA, et al. Foxp3-dependent programme of regulatory T-cell differentiation. *Nature*. 2007; 445:771–775. [PubMed: 17220874]
88. Wan YY, Flavell RA. Regulatory T-cell functions are subverted and converted owing to attenuated Foxp3 expression. *Nature*. 2007; 445:766–770. [PubMed: 17220876]
89. Zheng Y, et al. Role of conserved non-coding DNA elements in the Foxp3 gene in regulatory T-cell fate. *Nature*. 2010; 463:808–812. [PubMed: 20072126]
90. Hernandez-Hoyos G, et al. GATA-3 expression is controlled by TCR signals and regulates CD4/CD8 differentiation. *Immunity*. 2003; 19:83–94. [PubMed: 12871641]
91. Tai TS, et al. GATA-3 Regulates the Homeostasis and Activation of CD8⁺ T Cells. *J Immunol*. 2013; 190:428–437. [PubMed: 23225883]
92. Hosokawa H, et al. Gata3/Ruvbl2 complex regulates T helper 2 cell proliferation via repression of Cdkn2c expression. *Proc Natl Acad Sci U S A*. 2013; 110:18626–18631. [PubMed: 24167278]
93. Nie Z, et al. c-Myc Is a Universal Amplifier of Expressed Genes in Lymphocytes and Embryonic Stem Cells. *Cell*. 2012; 151:68–79. [PubMed: 23021216]
94. Buckley AF, et al. Transcription factor LKLF is sufficient to program T cell quiescence via a c-Myc-dependent pathway. *Nat Immunol*. 2001; 2:698–704. [PubMed: 11477405]
95. Tzachanis D, et al. Tob is a negative regulator of activation that is expressed in anergic and quiescent T cells. *Nat Immunol*. 2001; 2:1174–1182. [PubMed: 11694881]
96. Pei XH, et al. CDK inhibitor p18(INK4c) is a downstream target of GATA3 and restrains mammary luminal progenitor cell proliferation and tumorigenesis. *Cancer Cell*. 2009; 15:389–401. [PubMed: 19411068]
97. Wan YY, Flavell RA. How diverse--CD4 effector T cells and their functions. *J Mol Cell Biol*. 2009; 1:20–36. [PubMed: 19482777]
98. Dardalhon V, et al. IL-4 inhibits TGF- β -induced Foxp3⁺ T cells and together with TGF- β , generates IL-9⁺ IL-10⁺ Foxp3(–) effector T cells. *Nat Immunol*. 2008; 9:1347–1355. [PubMed: 18997793]
99. Veldhoen M, et al. Transforming growth factor- β 'reprograms' the differentiation of T helper 2 cells and promotes an interleukin 9-producing subset. *Nat Immunol*. 2008; 9:1341–1346. [PubMed: 18931678]
100. Goswami R, et al. STAT6-dependent regulation of Th9 development. *J Immunol*. 2012; 188:968–975. [PubMed: 22180613]
101. 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]
102. Spits H, Cupedo T. Innate lymphoid cells: emerging insights in development, lineage relationships, and function. *Annu Rev Immunol*. 2012; 30:647–675. [PubMed: 22224763]
103. Vosshenrich CA, et al. A thymic pathway of mouse natural killer cell development characterized by expression of GATA-3 and CD127. *Nat Immunol*. 2006; 7:1217–1224. [PubMed: 17013389]

104. 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. *J Exp Med*. 2006; 203:1105–1116. [PubMed: 16606668]
105. Monticelli LA, et al. Innate lymphoid cells promote lung-tissue homeostasis after infection with influenza virus. *Nat Immunol*. 2011; 12:1045–1054. [PubMed: 21946417]
106. Scanlon ST, McKenzie AN. Type 2 innate lymphoid cells: new players in asthma and allergy. *Curr Opin Immunol*. 2012; 24:707–712. [PubMed: 22985480]
107. Liang HE, et al. Divergent expression patterns of IL-4 and IL-13 define unique functions in allergic immunity. *Nat Immunol*. 2012; 13:58–66. [PubMed: 22138715]
108. Neill DR, et al. Nuocytes represent a new innate effector leukocyte that mediates type-2 immunity. *Nature*. 2010; 464:1367–1370. [PubMed: 20200518]
109. Wong SH, et al. Transcription factor RORalpha is critical for nuocyte development. *Nat Immunol*. 2012; 13:229–236. [PubMed: 22267218]
110. Yang Q, et al. T cell factor 1 is required for group 2 innate lymphoid cell generation. *Immunity*. 2013; 38:694–704. [PubMed: 23601684]
111. Mielke LA, et al. TCF-1 controls ILC2 and NKp46+RORgammat+ innate lymphocyte differentiation and protection in intestinal inflammation. *J Immunol*. 2013; 191:4383–4391. [PubMed: 24038093]
112. Zhang JA, et al. Dynamic transformations of genome-wide epigenetic marking and transcriptional control establish T cell identity. *Cell*. 2012; 149:467–482. [PubMed: 22500808]
113. 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. *Proc Natl Acad Sci U S A*. 2013; 110:10240–10245. [PubMed: 23733962]
114. 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]
115. 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]
116. Serafini N, et al. Gata3 drives development of RORgammat+ group 3 innate lymphoid cells. *J Exp Med*. 2014

Highlights

1. GATA3 integrates diverse upstream signals to control gene expression.
2. GATA3 controls the development and function of cells of adaptive immunity.
3. GATA3 regulates the generation and function of innate lymphoid cells.
4. GATA3 function is both cell-type specific and common for multiple cell types.

Figure 1. Th cell subsets and differentiation

Based on the signature cytokines produced, CD4⁺ helper T (Th) cells are classified as Th1, Th2, Th17 and Th9 cells. The differentiation of Th cells is induced by different combinations of cytokines and controlled by different transcription factors as depicted.

Figure 2. GATA3 integrates diverse upstream signals to control target gene expression and cellular functions

(A) GATA3 is regulated by multiple upstream signals. Signaling cascades activated through IL4 receptor (IL4R), IL2 receptor (IL2R), T cell receptor (TCR), Notch and Frizzled (receptor for Wnt) led to the activation of GATA3. p38 phosphorylates GATA3 to promote its nuclear translocation. GATA3 also activates its own expression through a feedback loop. Depending on the cellular and environmental contexts, one or more of these depicted signaling pathways control the functions of GATA3 in specific cell types. (B) GATA3 controls gene expression and cellular functions. As a transcription factor, GATA3 regulates target gene expression to control diverse cellular and biological processes. GATA3 activates TCR α and ThPOK expression during thymocyte development. It promotes Th2 cytokine expression to regulate Th1/Th2 cell differentiation, enhances Foxp3 expression to stabilize Treg cell function, and controls ROR- α expression for ILC2 generation. GATA3 is also required for the expression of Myc and IL7R α that are critical for the metabolism, proliferation and homeostasis of immune cells. GATA3 achieves these diverse functions either by directly activating target genes or by associating with co-factors to regulate gene expression and the epigenetic modification.

Figure 3. GATA3 controls the generation and function of immune cells

During immune cell development and function, GATA3 plays positive (green plus symbol) or negative (red minus symbol) roles in different cell types. GATA3 is central for the development of multiple immune cell types: It controls the generation of early T cell progenitors, thymocytes, CD4⁺ T and iNKT (invariant natural killer T) cells. In contrast, GATA3 suppresses B cell development. GATA3 is also required for the development of NK (natural killer) and ILC2 (type-2 innate lymphoid cell) cells. In addition, GATA3 is critical for the function of various sub-types of mature T cells: While GATA3 promotes T cell homeostasis, activation, proliferation and the differentiation of Th2 and Th9 cells, it antagonizes Th1 and Th17 differentiation. Moreover, GATA3 is required for Treg (regulatory T) cell homeostasis, suppressive activity and functional stability.