Regulation of the humoral type 2 immune response against allergens and helminths

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The type 2 immune response is associated with helminth infections and allergic inflammation where antibody production of the IgG1 and IgE isotypes can elicit protective or proinflammatory functions. Studies over the past few years revealed important new insights regarding the regulatory mechanisms orchestrating the humoral type 2 immune response. This includes investigations on B-cell extrinsic signals, such as IL-4 and IL-21, derived from different T-helper cell subsets or discovery of new follicular helper T cells with regulatory or IgE-promoting activities. In addition, studies on B-cell intrinsic factors required for germinal center formation and class switch recombination, including the transcription factors STAT3, STAT6, and BCL-6, led to a better understanding of these processes in type 2 immune responses. Here, we review the current understanding of mechanisms controlling humoral type 2 immunity in vivo including the generation of IgE-producing plasma cells and the memory IgE response.

Keywords: Allergy · Germinal center · Helminths · IgE · STAT6

Introduction

Type 2 immune responses are frequently elicited by infections with larger parasites, such as helminths, allergens, and venom toxins. Hallmarks of type 2 immunity include an increase in numbers of IL-4, IL-5, and IL-13-producing effector cells such as Th2 cells, type 2 innate lymphoid cells (ILC2s), NKT cells, eosinophils, basophils, and mast cells, differentiation of alternatively activated macrophages, goblet cell hyperplasia, smooth muscle cell activity, and elevated serum levels of IgE and IgG1. The cytokines IL-4 and IL-13 play a key role in type 2 immune responses and bind to heterodimeric receptors composed of either the IL-4Ra chain together with the common gamma (γc) chain (type I IL-4 receptor), binds only IL-4 or the IL-4Ra1 chain together with the IL-13Ra1 chain (type II IL-4 receptor, binds IL-4 and IL-13) [1, 2]. IL-13 further binds to IL-13Ra2, which can be expressed as transmembrane or soluble IL-13 receptor. The type I receptor is mainly expressed on hematopoietic cells, while the type II receptor is expressed on nonhematopoietic cells but also on B cells [3]. Both receptors activate the transcription factor STAT6, which plays an important role for the humoral type 2 immune response by promoting GC formation and differentiation of IgE-producing plasma cells (PCs). IgE-sensitized mast cells and basophils can contribute to protective immunity against helminths but also elicit allergic inflammation in response to allergens.

In this review, we discuss recent findings regarding the cellular and molecular mechanisms that control the humoral type 2 immune response at different levels, including the pre-GC phase, formation of GCs, role of Th2, and T-follicular helper (Tfh) cells and the generation of memory B cells and IgE-producing PCs.

Initiation of the GC response

The GC is a microenvironment generated during an immune response and localized within the B-cell zone in secondary lymphoid organs. GCs consist mainly of activated B cells, follicular dendritic cells (FDC) on the surface of which B cells see their antigens and Tfh cells that provide important differentiation and
survival signals to GC B cells. A GC is initiated after naïve B cells encounter their cognate antigen, which happens either outside of the B-cell follicle, in the subcapsular sinus of lymph nodes, in the submarginal sinus of the spleen, or directly in the B-cell follicle. B cells then upregulate the chemokine receptor CCR7 that guides them to the border between B-cell follicle and T-cell zone where they present internalized antigens to T-helper cells (Th) and receive further activating signals, mainly by CD40-CD40L interaction and cytokines. Such activated B cells downregulate expression of the chemokine receptor EBI-2 (GPR183) and migrate into the B-cell follicle to establish the GC where they can undergo Ig class switch recombination (CSR) and acquire somatic mutations in Ig genes which leads to affinity maturation by competitive selection within the GC structure [4, 5] (Fig. 1).

CSR and acquisition of somatic mutations are both dependent on the enzyme Activation-induced cytidine deaminase (AID) which shows highest expression in GC B cells [6]. CSR inside GCs has been reported to happen after the onset of somatic mutations [7]. However, a recent study in mice revealed that CSR mainly occurs before B cells enter the GC [8]. Others have shown that affinity maturation and CSR can also take place in a GC-independent manner under certain conditions [9, 10]. For example, mice lacking Treg showed elevated levels of IgE against allergens and this IgE production occurred independently of GCs [11]. Furthermore, patients with CD40L-deficiency were found to contain IgE+ CD27- B cells with few SHM suggesting a GC-independent origin [12].

Role of T cells and cytokines for GC formation and IgE production in mice during type 2 immunity

Tfh cells are characterized by their localization in GCs and expression of BCL-6, PD-1, CXCR5, CD40L, ICOS, and IL-21. They can originate from Th1, Th2, or Th17 cells and keep expression of the respective cytokines [13]. Hence, in type 2 immune responses Tfh cells can express both IL-21 and IL-4, and both cytokines play an important role for efficient GC formation. Mechanistically, they inhibit the degradation of the key transcriptional repressor BCL-6 which is required for the GC B-cell differentiation program [14]. In addition, IL-4-induced STAT6 activation promotes transcription of BCL-6 [14]. Although IL-4 and IL-21 can act on many cell types, several studies have shown that efficient GC formation requires direct recognition of these cytokines by B cells [15–17]. Kinetic studies further revealed that IL-21 expression precedes IL-4 expression in Tfh, before their migration into the B-cell follicle, indicating that IL-21 in contrast to IL-4 has a dominant role at the pre-GC B-cell stage [18]. However, others have shown that
an early wave of NKT cell-derived IL-4 also plays an important role for GC seeding by B cells during viral infections [19]. Besides their common activities, such as promoting B-cell proliferation and survival, IL-21 and IL-4 also appear to have opposite functions regarding IgE CSR and generation of IgE-producing PCs. IL-21 promotes B-cell proliferation and plasma blast differentiation by binding to the IL-21 receptor on B cells which is associated mainly with STAT3 for signal transduction and regulation of gene expression. IL-21 further inhibits IgE production while promoting the IgG1 response in a STAT3-dependent manner [20, 21]. Consistent with these findings, IL-21 and IL-21R-deficient mice develop spontaneous hyper-IgE phenotypes [22]. In humans, a dominant negative mutation of STAT3 results in hyper-IgE syndrome [23]. This indicates that Tfh-derived IL-21 increases the threshold for IgE CSR in GCs.

Tfh cells were further found to be the major source of IL-4 in reactive lymph nodes in type 2 immune responses against Leishmania major or different helminth infections [24–26]. However, this does not necessarily prove that they are the critical source of IL-4 for GC formation and IgE CSR. In fact, IL-4/IL-13 expression by CXCR5-deficient T cells that could not enter GCs was sufficient for a normal GC and IgE response [17]. This indicates that IL-4 from Th2 or pre-Tfh cells outside GCs plays a dominant role. Furthermore, it was shown that the majority of B cells in reactive lymph nodes of helminth-infected mice contain phosphorylated STAT6 indicating that IL-4 can diffuse through an entire LN [27]. Others have found that deletion of IL-4 in Tfh cells results in a diminished Th2 and IgE response to helminth infection [28]. It was further reported that IL-4-deficient mice develop impaired GCs and a poor IgE/IgG1 response in a mouse model of allergic lung inflammation demonstrating the general requirement of Tfh cells but not necessarily IL-4 production by them [29]. Studies with selective and inducible deletion of IL-4 in Tfh cells would be helpful to resolve this issue.

A recent study reported the identification of a small subset of Tfh cells in mouse and man that expresses the transcription factor GATA3 and high levels of IL-13 in addition to IL-4 and IL-5 [30]. These Tfh13 cells were only found after repeated challenge of mice with various allergens but not after primary infection with the helminth Nippostrongylus brasiliensis. Interestingly, Tfh13 cells promoted the differentiation of PCs producing high-affinity IgE antibodies that caused anaphylactic response upon antigen challenge [30]. In vitro stimulation of B cells with anti-CD40, IL-4, and IL-13 resulted in more IgE+ PCs as compared to cultures without IL-13. Further investigations are required to understand how IL-13 signaling in B cells or PCs promotes the generation of high-affinity anaphylactic IgE antibodies and whether this process is also relevant in the human immune system. A better understanding of this pathway could then lead to development of new therapeutic interventions to prevent formation of PCs producing anaphylactic IgE antibodies.

Several efforts are being made to inhibit Tfh functions in allergic conditions of humans. In a recent clinical study, it was observed that circulating CXCR5+ T cells which show a phenotype similar to Tfh cells in GCs were increased in number in patients with allergic asthma [31]. Those cells provide help to B cells like conventional Tfh cells and cause an increase of serum IgE. Another study observed increased Tfh cell numbers with more IL-4 secretion in tonsils of children sensitized to house dust mite allergens [32]. This marks Tfh cells as promising candidate for therapy with the ICOS/ICOS-L pathway being one potential target. In fact, treatment of mice with anti-ICOS-L antibodies ameliorated established allergy in the house dust mite model [33]. Another study in humanized mice demonstrated that NK cells expressing a chimeric antigen receptor against human PD-1 successfully removed Tfh cells without affecting other T-cell populations showing a promising future for treatment against Tfh-mediated diseases [34].

### Regulation of the GC and IgE responses by follicular regulatory T (Tfr) cells

Tregs are an important part of the immune system and are tasked with inhibition of self-reactive T and B cells [35, 36]. However, they also have further functions beyond protection against autoimmunity. Tregs are also present in the GC and constitute a distinct subset named follicular regulatory T (Tfr) cells. The function of these cells is to maintain and control the overall GC reaction. Deleting Tfr cells leads to increased GC size and GC B-cell proliferation but also causes impaired affinity maturation and increased autoimmunity [37, 38]. Tfr cells can either directly confer inhibitory signals to B cells or modulate the GC response indirectly via interfering with interactions between Tfh cells and B cells [36, 39]. By these mechanisms Tfr cells can generally fine tune the GC response (Fig. 1).

Tfr cells also play a critical role for controlling IgE responses. It was shown in mice that expression of the transcriptional repressor Blimp-1 in Tfr cells is required for prevention of spontaneous autoantibody production including self-reactive IgE [35]. Blimp-1 promoted the Tfr phenotype by maintaining expression of Foxp3, activating the STAT5 signaling pathway, and controlling CXCR5/CCR7 expression for homing of Tfr cells into the GC. Tfr cells also regulate the immune response to allergens as shown in a mouse model of house dust mite allergy where Tfr cells modulated the activity of Tfh cells [38]. In addition, Tfr cells can directly inhibit B cells via IL-10 signaling in a model of food allergy [40].

### B-cell intrinsic STAT6-dependent regulation of the IgE and GC response in mice

Antigen-activated B cells are required to integrate extrinsic signals from T cells and other sources to alter their gene expression profile and differentiate into GC B cells, undergo CSR and further develop into memory B cells and PCs. The transcription factor STAT6 plays a key role for these processes in type 2 immune responses.

STAT6 promotes CSR to IgE and IgG1 by direct binding to DNA elements in the germline β and γ promoters [41–43].
This process is enhanced by the poly AD-ribosyl polymerase (PARP-14) by release of inhibitory histone deacetylases (HDACs) from STAT6 binding sites [44, 45]. In addition, STAT6 induces expression of the transcription factor NFIL3 which is also required for CSR to IgE [46]. B cells can switch from IgM to IgE expression either directly or sequentially with an intermediate step of IgG1 expression. Sequential IgE CSR is required for production of affinity-matured IgE and for the memory IgE response [47, 48]. IgE+ GC B cells are very rare and this might be explained by IgE BCR-mediated signals that promote GC exit and differentiation to short-lived PCs independently of antigen binding [49, 50]. The IgE BCR was further shown to inhibit the formation of memory B cells and long-lived PCs [51].

In addition to promoting CSR to IgE and IgG1, B cell-intrinsic STAT6 is required for GC formation in type 2 immune responses to helminths or allergens but not for GC formation in response to viral infections [17]. STAT6 controls expression of well over 100 genes in B cells and the function of most STAT6-regulated genes in the context of GC formation and the B-cell fate during type 2 immune responses remains to be explored [52]. STAT6 promotes expression of MHC-II and CD86 on B cells, which could contribute to better interaction with antigen-specific T cells [42]. STAT6 further acts together with NF-kB to induce expression of AID [53]. Caspase-6 is another STAT6-regulated gene which appears to be required for CSR to IgG1 and PCs differentiation [54]. Activated STAT6 also promotes the upregulation of the low-affinity IgE receptor CD23 on the surface of B cells and this may lead to uptake of IgE-bound antigens that can then be presented to CD4 T cells [42, 55, 56]. Functional characterization of further STAT6-regulated proteins will help to better understand the GC response in type 2 immunity.

In addition to STAT6, GC formation and IgG1 CSR is also promoted by IL-21-induced activation of STAT3 in B cells [20, 21]. However, STAT3 inhibits IgE CSR in a B cell-intrinsic manner and this effect can be overcome by strong CD40 signaling [21, 57] (Fig. 2). How exactly STAT3 inhibits IgE CSR and promotes IgG1 CSR remains to be investigated. Another transcription factor that inhibits IgE CSR is BCL-6 which shares several DNA recognition sites in the genome with STAT6 including binding sites in the germline ε promoter [58–60]. On the other hand, BCL-6 is required for GC formation [60]. The high expression level of BCL-6 in GC B cells could therefore explain the relatively low frequency of IgE+ GC B cells in various settings of type 2 immune responses.

**PCs development and memory B-cell responses in type 2 immunity**

PCs are the major population of antibody-producing cells. They are generated during each step of the immune response, and therefore, differ in their phenotype, half-life, affinity, and type of secreted antibody. Early PCs are derived from B cells that do not enter GCs or exit GCs at an early phase of the immune response and therefore acquired only few somatic mutations. They can provide first initial protection against pathogens and help to capture and therefore provide antigen for the GC reaction itself [5, 61]. In contrast, PCs produced during or at the end of the GC reaction are characterized by secretion of affinity matured and Ig class-switched antibodies.

Differentiation of B cells into PCs is strictly regulated by Thh and Tfr cells. PC differentiation is promoted by interaction between ICOS and CD40L expressed on Thh cells and ICOS-L and CD40 expressed on GC B cells [61]. A high BCR affinity would further improve this interaction thus favoring high affinity B cells to develop into PCs that leave the GC. Tfr cells on the other hand prevent the generation and GC exit of PCs that produce unspecific or autoreactive antibodies [38, 40, 62].

The half-life of IgE+ PCs is about 60 days in mice and four times shorter than the half-life of IgG1+ PCs [63]. However, chronic allergen exposure can cause accumulation of IgE+ PCs in the BM of both mice and humans [64]. The same IgE clonotypes can be detected in the peripheral blood of birch pollen-allergic individuals in consecutive pollen seasons but not in off-season indicating that memory IgE persists long term at the clonal level [65]. IgE CSR may also occur within mucosal tissues as clonally related IgE+ and other isotypes have been recently identified in stomach and duodenum of human subjects affected by peanut allergy [66].

IgE-producing PCs express IgE BCRs on the cell surface (mIgE). This mIgE contains a so-called immunoglobulin tail tyrosine (ITT) motif in the cytoplasmic tail and a Tyr≥Phe mutation within this motif or deletion of the mIgE tail results in impaired memory IgE responses to helminths or allergen [67]. The ITT motif promotes surface expression of mIgE on PCs and due to the autonomous signaling capacity of mIgE may thereby promote...
In another study, a humanized anti-IgE antibody was described that neutralized and thereby prevents further IgE production [69, 70]. In another study, an antibody against the M1 prime epitope of mIgE targets IgE producing PCs. Indeed, an antibody against the M1 prime epitope of mIgE effectively neutralized the memory IgE response indicating that either IgG1 or that IgE memory B cells receive extrinsic survival signals through the IgG1 Fc receptor. This indicates that the mIgE might be a good target to specifically delete IgE-producing PCs. This indicates that IgE memory persists either in the form of long-lived PCs that can be directly reactivated or in the form of IgG1+ memory B cells that immediately give rise to sequentially switched IgE+ PCs upon antigen encounter (Fig. 3). The memory IgE response seems indeed to be conserved at the level of IgG1+ memory B cells, at least in mice [48, 63, 68]. The second step of the sequential switch to IgE during the memory phase requires again IL-4 from T cells [48, 63]. Exchange of the extracellular part of IgG1 with IgE sequences during the memory phase requires again IL-4 from T cells [48, 63, 68]. The second step of the sequential switch to IgE during the memory phase requires again IL-4 from T cells [48, 63, 68]. The recent discovery of IL-13 producing Tfh cells required for production of anaphylactic IgE in response to allergens puts a new layer of complexity on the regulation of IgE production. Tfr cells are required for control of the GC response in general but also the IgE response in a house dust mite model. How these cells control Tfr cells and B cells inside GCs and whether they also act on emerging PCs requires further investigations.

The discovery of new regulatory mechanisms that control GC formation and IgE production during type 2 immune response could help to develop novel therapeutic strategies for treatment of allergic diseases.

Figure 3. Progressive differentiation of IgE-producing plasma cells (PCs). PCs can be generated during early type 2 immune responses by direct IgM to IgE switching resulting in production of low-affinity IgE which does not elicit allergic reaction. Later immune responses generated sequentially switched, GC-dependent and high affinity IgE-producing PC. The cytoplasmic tail of the IgE BCR promotes the accumulation of IgE+ PCs.

Conclusions

The humoral type 2 immune responses leading to GC formation and production of protective or anaphylactic IgE antibodies against pathogens and allergens are controlled by several B-cell extrinsic and intrinsic mechanisms. The T cell-derived cytokines IL-4 and IL-21 play a critical role for development of productive GCs but they appear to have opposite functions regarding the IgE response where IL-4 promotes and IL-21 inhibits CSR to IgE. The regulation of gene expression and IgE CSR by STAT6, STAT3, and BCL-6 in primary B cells remains an important field of research to identify further mechanisms that control the humoral type 2 immune response in vivo.

B cells that directly switch from IgM to IgE do not undergo affinity maturation in GCs but rather differentiate to short-lived PCs and this process is driven by autonomous signaling of the mIgE. There is now good evidence that the memory IgE response is conserved at the level of IgG1+ memory B cells that sequentially switch to IgE and then differentiate to IgE+ PCs upon rechallenge. IgE+ PCs also express mIgE and the cytoplasmic tail of mIgE promotes their expansion by mechanisms that are not yet understood. The recent discovery of IL-13 producing Tfh cells required for production of anaphylactic IgE in response to allergens puts a new layer of complexity on the regulation of IgE production. Tfr cells are required for control of the GC response in general but also the IgE response in a house dust mite model. How these cells control Tfr cells and B cells inside GCs and whether they also act on emerging PCs requires further investigations.

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References

1. Nelms, K., Keegan, A. D., Zamorano, J., Ryan, J. J. and Paul, W. E., The IL-4 receptor: signaling mechanisms and biologic functions. Annu. Rev. Immunol. 1999. 17: 701–738.
2. de Vries, J. E., The role of IL-13 and its receptor in allergy and inflammatory responses. J. Allergy. Clin. Immunol. 1998. 102: 165–169.
3. Punnonen, J., Aversa, G., Cocks, B. G., McKenzie, A. N., Menon, S., Zurawski, G., de Waal Malefyt, R. et al., Interleukin 13 induces interleukin 4-independent IgG4 and IgE synthesis and CD23 expression by human B cells. Proc. Natl. Acad. Sci. USA 1993. 90: 3730–3734.
4. Biram, A., Davidzohn, N. and Shulman, Z., T cell interactions with B cells during germinal center formation, a three-step model. Immunol. Rev. 2019. 288: 37–48.
5. Haberman, A. M., Gonzalez, D. G., Wong, P., Zhang, T. T. and Kerfoot, S. M., Germinal center B cell initiation, GC maturation, and the coevolution of its stromal cell niches. Immunol. Rev. 2019. 288: 10–27.
6. Muramatsu, M., Kinoshita, K., Fagarasan, S., Yamada, S., Shinkai, Y. and Honjo, T., Class switch recombination and hypermutation require
initiation of germinal center B cells and subsequent self-renewal transi-
in the germinal center. Immunol. 2019. 51: 337–350.

Miyaochi, K., Sugimoto-Ishige, A., Harada, Y., Adachi, Y., Usami, Y., Kaji, T., Inoue, K. et al., Protective neutralizing influenza antibody response in the absence of T follicular helper cells. Nat. Immunol. 2016. 17: 1447–1458.

Takemori, T., Kaji, T., Takahashi, Y., Shimoda, M. and Rajewsky, K., Generation of memory B cells inside and outside germinal centers. Eur. J. Immunol. 2014. 44: 1258–1264.

Tai, Y., Takano, A., Haga, K., Koshida, K. and Harada, Y., Spontaneous anti-body production caused by regulatory T cell deficiency occurs through a germinal center-independent pathway. Biochem. Biophys. Res. Commun. 2020. 527: 909–914.

Berkowska, M. A., Heeringa, J. J., Hajdarbegovic, E., van der Burg, M., Thio, H. B., van Hagen, P. M., Boon, L. et al., Human IgE+B cells are derived from T cell-dependent and T cell-independent pathways. J. Allergy Clin. Immunol. 2014. 134: 688–697.

Crotty, S., Follicular helper CD4 T cells (TFH). Ann. Rev. Immunol. 2011. 29: 621–663.

Chevrier, S., Kratina, T., Emslie, D., Tarlinton, D. M. and Corcoran, L. M., IL4 and IL21 cooperate to induce the high Bcl6 protein level required for germinal center formation. Immunol. Cell Biol. 2017. 95: 925–932.

Linterman, M. A., Beaton, L., Yu, D., Ramiscal, R. R., Srivastava, M., Hogan, J. J., Verma, N. K. et al., IL-21 acts directly on B cells to regulate Bcl-6 expression and germinal center responses. J. Exp. Med. 2010. 207: 353–363.

Zotos, D., Coquet, J. M., Zhang, Y., Light, A., D’Costa, K., Kallies, A., Corcoran, L. M. et al., IL-21 regulates germinal center B cell differentiation and proliferation through a B cell-intrinsic mechanism. J. Exp. Med. 2010. 207: 365–378.

Turqueti-Neves, A., Otte, M., Costa, O. P., Höpken, U. E., Lipp, M., Buch, T. and Voehringer, D., B-cell-intrinsic STAT6 signaling controls germinal center formation. Eur. J. Immunol. 2014. 44: 2130–2138.

Gonzalez, D. G., Cote, C. M., Patel, J. R., Smith, C. B., Zhang, Y., Nickerson, K. M., Zhang, T. et al., Nonredundant roles of IL-21 and IL-4 in the phased initiation of germinal center B cells and subsequent self-renewal transitions. J. Immunol. 2018. 201: 3569–3579.

Gaya, M., Barral, P., Burbage, M., Aggarwal, S., Montaner, B., Warren Navia, A., Ail, M. et al., Initiation of antiviral B cell immunity relies on innate signals from spatially positioned NK cells. Cell 2018. 172: 517–533.

Suto, A., Nakajima, H., Hirose, K., Suzuki, K., Kagami, S., Seto, Y., Hoshimoto, A. et al., Interleukin 21 prevents antigen-induced IgE production by inhibiting germ line (C elegans) transcription of IL-4-stimulated B cells. Blood 2002. 100: 4565–4573.

Yang, Z., Wu, C.-A. M., Targ, S. and Allen, C. D. C., IL-21 is a broad negative regulator of IgE class switch recombination in mouse and human B cells. J. Exp. Med. 2020. 217: e20190472.

Ozaki, K., Spolski, R., Feng, C. G., Qi, C. F., Cheng, J., Sher, A., Morse, H. C., 3rd et al., A critical role for IL-21 in regulating immunoglobulin production. Science 2002. 298: 1630–1634.

Minegishi, Y., Saito, M., Tsuchiya, S., Tsuge, I., Takada, H., Hara, T., Kawamura, N. et al., Dominant-negative mutations in the DNA-binding domain of STAT3 cause hyper-IgE syndrome. Nature 2007. 448: 1058–1062.

Zaretsky A., G., Taylor, J. J., King, I. L., Marshall, F. A., Mohrs, M. and Pearce, E. J., T follicular helper cells differentiate from Th2 cells in response to helminth antigens. J. Exp. Med. 2009. 206: 991–999.

Reinhardt, R. L., Liang, H. E. and Locksley, R. M., Cytokine-secreting follicular T cells shape the antibody repertoire. Nat. Immunol. 2009. 10: 385–393.

King, I. L. and Mohrs, M., IL-4-producing CD4+ T cells in reactive lymph nodes during helminth infection are T follicular helper cells. J. Exp. Med. 2009. 206: 1001–1007.

Perona-Wright, G., Mohrs, K. and Mohrs, M., Sustained signaling by canonical helper T cell cytokines throughout the reactive lymph node. Nat. Immunol. 2010. 11: 520–526.

Meli, A. P., Fontés, G., Leung Soo, C. and King, I. L., T follicular helper cell-derived IL-4 is required for IgE production during intestinal helminth infection. J. Immunol. 2017. 199: 244–252.

Kobayashi, T., Iijima, K., Dent, A. L. and Kita, H., Follicular helper T cells mediate IgE antibody response to airborne allergens. J. Allergy Clin. Immunol. 2017. 139: 300–313.

Gowthaman, U., Chen, J. S., Zhang, B., Flynn, W. F., Lu, Y., Song, W., Joseph, J. et al., Identification of a T follicular helper cell subset that drives anaphylactic IgE. Science 2019. 365: eaaw6433.

Gong, F., Zhu, H.-Y., Zhu, J., Dong, Q.-J., Huang, X. and Jiang, D.-J., Circulating CXCR5+CD4+ T cells participate in the IgE accumulation in allergic asthma. Immunol. Lett. 2018. 197: 9–14.

Foster, W. S., Grime, C. J., Tan, H.-L., Williams, G. S., Robinson, M. J., Carlesso, G., Saglani, S. et al., Enhanced frequency and function of follicular T cells in the tonsils of house dust mite-sensitized children. Allergy 2020. 75: 1240–1244.

Uwadiase, F. I., Pyle, C. J., Walker, S. A., Lloyd, C. M. and Harker, J. A., Targeting the ICOS/ICOS-L pathway in a mouse model of established allergic asthma disrupts T follicular helper cell responses and ameliorates disease. Allergy 2019. 74: 650–662.

Reighard, S. D., Cranert, S. A., Rangel, K. M., Ali, A., Gyurova, I. E., de la Cruz-Lynch, A. T., Tuazon, J. A. et al., Therapeutic targeting of follicular T cells with chimeric antigen receptor-expressing natural killer cells. Cell Rep. Med. 2020. 1: 100003.

Shen, E., Rabe, H., Luo, L., Wang, L., Wang, Q., Yin, J., Yang, X. et al., Control of germinal center localization and lineage stability of follicular regulatory T cells by the Bmp1 transcription factor. Cell Rep. 2019. 29: 1848–1861.

Wing, J. B., Lim, E. L. and Sakaguchi, S., Control of foreign Ag-specific Ab responses by Treg and Tfr. Immuno. Rev. 2020. 296: 104–119.

Chung, Y., Tanaka, S., Chu, F., Nuriev, R. I., Martinez, G. J., Rawal, S., Wang, Y.-H. et al., Follicular regulatory T cells expressing Foxp3 and Bcl-6 suppress germinal center reactions. Nat. Med. 2011. 17: 983–988.

Clement, R. L., Daccauche, J., Mohammed, M. T., Diallo, A., Blazar, B. R., Kuchroo, V. K., Lovitch, S. B. et al., Follicular regulatory T cells control humoral and allergic immunity by restraining early B cell responses. Nat. Immunol. 2019. 20: 1360–1371.

Xie, M. M. and Dent, A. L., Unexpected help: follicular regulatory T cells in the germinal center. Front. Immunol. 2018. 9:1536.

Xie, M. M., Chen, Q., Liu, H., Yang, K., Koh, B., Wu, H., Maleki, S. J. et al., T follicular regulatory cells and IL-10 promote food antigen-specific IgE. J. Clin. Invest. 2020. 130: 3820–3832.

Shimoda, K., van Deursen, J., Sangster, M. Y., Sarawar, S. R., Carson, R. T., Tripp, R. A., Chu, C. et al., Lack of IL-4-induced Th2 response and IgE class switching in mice with disrupted Stat6 gene. Nature 1996. 380: 630–633.

Takeda, K., Tanaka, T., Shi, W., Matsumoto, M., Minami, M., Kashiwamura, S., Nakanishi, K. et al., Essential role of Stat6 in IL-4 signalling. Nature 1996. 380: 627–630.
43 Linehan, L. A., Warren, W. D., Thompson, F. A., Grusby, M. J., and Berton, M. T., STAT6 is required for IL-4-induced germline Ig gene transcription and switch recombination. J. Immunol. 1998. 161: 302–310.

44 Mehrrota, P., Riley, J. P., Patel, R., Li, F., Voss, L. E. and Goenka, S., PARP-14 functions as a transcriptional switch for Stat6-dependent gene activation. J. Biol. Chem. 2011. 286: 1767–1776.

45 Cho, S. H., Raybuck, A., Wei, M., Erickson, J., Nam, K. T., Cox, R. G., Troenhagen, A. et al., B cell-intrinsic and -extrinsic regulation of antibody responses by PARP14, an intracellular (ADP-Ribosyl)transferase. J. Immunol. 2013. 191: 3169–3178.

46 Rothman, P. B. The transcriptional regulator NFIIL3 controls IgE production. Trans. Am. Cliniat. Assoc. 2010. 121: 156–171.

47 Xiong, H., Dolpady, J., Wab, M., Curotte de Lafaille, M. A. and Lafaille, J. J., Sequential class switching is required for the generation of high affinity IgE antibodies. J. Exp. Med. 2012. 209: 353–364.

48 Turqueti-Nieves, A., Otte, M., Schwartz, C., Schmitt, M. E., Lindner, C., Pabst, O., Yu, P. et al., The extracellular domains of IgG1 and IgV cell-derived IL-4/IL-13 are critical for the polyclonal memory IgE response in vivo. Proc. Biol. Sci. 2015. 13: e1002290.

49 Yang, Z., Sullivan, Brandon M. and Allen Christopher, D. C., Fluorescent in vivo detection reveals that IgE+ B Cells are restrained by an intrinsic cell fate predisposition. Immunity 2012. 36: 857–867.

50 Yang, Z., Robinson, M. J., Chen, X., Smith, G. A., Taunton, J., Liu, W. and Allen, C. D., Regulation of B cell fate by chronic activity of the IgE B cell receptor. Elife. 2016. 5: e22138.

51 Haniu, K., Fukao, S., Kodama, T., Hasegawa, H. and Kitamura, D., Autonomous membrane IgE signaling prevents IgE-memory formation. Nat. Immunol. 2016. 17: 1109–1117.

52 Mokada-Gopal, L., Boeser, A., Lehmann, C. H. K., Drepper, F., Dudziak, D., Warscheid, B. and Voehringer, D., Identification of novel Stat6-regulated proteins in mouse B cells by comparative transcriptome and proteome analysis. J. Immunol. 2017. 198: 3737–3745.

53 Dedeoglu, F., Horwitz, B., Chaudhuri, J., Alt, F. W. and Geha, R. S., Induction of activation-induced cytidine deaminase gene expression by IL-4 and CD40 ligation is dependent on Stat6 and NF-kappaB. Int. Immunol. 2004. 16: 395–404.

54 Watanabe, C., Shu, G. L., Zheng, T. S., Flavell, R. A. and Clark, E. A., Caspase 6 regulates B cell activation and differentiation into plasma cells. J. Immunol. 2008. 181: 6810–6819.

55 Gould, H. J. and Sutton, B. J., IgE in allergy and asthma today. Nat. Rev. Immunol. 2008. 8: 205–217.

56 Haase, P., Mokada-Gopal, L., Radtke, D. and Voehringer, D., Modulation of the humoral immune response by constitutively active Stat6 expression in murine B cells. Eur. J. Immunol. 2020. 50: 558–567.

57 Dascani, P., Ding, C., Kong, X., Tieri, D., Hu, X., Zhang, H.-g., Kitamura, D. et al., Transcription factor Stat3 serves as a negative regulator controlling IgE class switching in mice. Immuno Horizons 2018. 2: 349–362.

58 Harris, M. B., Chang, C.-C., Berton, M. T., Danial, N. N., Zhang, J., Kuehner, D., Ye, B. H. et al., Transcriptional repression of Stat6-dependent interleukin-4-induced genes by Bcl-6: specific regulation of Iκ transcription and immunoglobulin E switching. Mol. Cell. Biol. 1999. 19: 7264–7275.

59 Audzevich, T., Pearce, G., Breucha, M., Gündüz, G. and Jessberger, R., Control of the Stat6−Bcl6 antagonism by SWAP-70 determines IgE production. J. Immunol. 2013. 190: 4946–4955.

60 Dent, A. L., Shaffer, A. L., Yu, X., Allman, D. and Staudt, L. M., Control of inflammation, cytokine expression, and germinal center formation by Bcl-6. Science 1997. 276: 589–592.

61 Ise, W. and Kurosaki, T., Plasma cell differentiation during the germinal center reaction. Immunol. Rev. 2019. 288: 64–74.

62 Bott, D., Fuller, M. J., Marquez-Lago, T. T., Bachus, H., Bradley, J. E., Weinmann, A. S., Zajac, A. J. et al., Dynamic regulation of T follicular regulatory cell responses by interleukin 2 during influenza infection. Nat. Immunol. 2017. 18: 1249–1260.

63 Jiménez-Saiz, R., Chu, D. K., Mandur, T. S., Walker, T. D., Gordon, M. E., Chaudhary, R., Koenig, J. et al., Lifelong memory responses perpetuate humoral TH2 immunity and anaphylaxis in food allergy. J. Allergy Clin. Immunol. 2017. 140: 1604–1615.

64 Asrat, S., Kaur, N., Lui, X., Ben, L.-H., Kajimura, D., Murphy, A. J., Sleeman, M. A. et al., Chronic allergen exposure drives accumulation of long-lived IgE plasma cells in the bone marrow, giving rise to serological memory. Sci. Immunol. 2020. 5: eaav4602.

65 Otte, M., Mahler, V., Kerpes, A., Pabst, O. and Voehringer, D., Persistence of the IgE repertoire in birch pollen allergy. J. Allergy Clin. Immunol. 2016. 137: 1884–1887.

66 Hoh, R. A., Joshi, S. A., Lee, J. Y., Martin, B. A., Varma, S., Kwok, S., Nielsen, S. C. A. et al., Origins and clonal convergence of gastrointestinal IgE(+) B cells in human peanut allergy. Sci. Immunol. 2020. 5: eaay4209.

67 Schmitt, M. E. R., Lutz, J., Haase, P., Bös, M. R., Wienands, J., Engels, N. and Voehringer, D., The B-cell antigen receptor of IgE-switched plasma cells regulates memory IgE responses. J. Allergy Clin. Immunol. 2020. 146: 642–651.

68 He, J.-S., Subramanian, S., Narang, V., Srinivasan, K., Saunders, S. P., Carbajo, D., Wen-Shan, T. et al., IgG1 memory B cells keep the memory of IgE responses. Nat. Commun. 2017. 8: 641.

69 Gauvreau, G. M., Harris, J. M., Boulet, L.-P., Scheerens, H., Fitzgerald, J. M., Putnam, W. S., Cockcroft, D. W. et al., Targeting membrane-expressed IgE B cell receptor with an antibody to the M1 prime epitope reduces IgE production. Sci. Transl. Med. 2014. 6: 243ra285–243ra285.

70 Brightbill, H. D., Jeet, S., Lin, Z., Yan, D., Zhou, M., Tan, M., Nguyen, A. et al., Antibodies specific for a segment of human membrane IgE deplete IgE-producing B cells in humanized mice. J. Clin. Invest. 2010. 120: 2218–2229.

71 Nyborg, A. C., Zacco, A., Ettinger, R., Jack Borrok, M., Zhu, J., Martin, T., Woods, R. et al., Development of an antibody that neutralizes soluble IgE and eliminates IgE expressing B cells. Cell Mol. Immunol. 2016. 13: 391–400.

Abbreviations: AID: activation-induced cytidine deaminase · CSR: class switch recombination · ITT: immunoglobulin tail tyrosine · ILC2: innate lymphoid cells · PCs: plasma cells · Tfh: T-follicular helper · Tfr: follicular regulatory T cells

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