

B-lymphocyte lineage cells and the respiratory system

Atsushi Kato, PhD,^a Kathryn E. Hulse, PhD,^a Bruce K. Tan, MD,^b and Robert P. Schleimer, PhD^{a,b} *Chicago, Ill*

INFORMATION FOR CATEGORY 1 CME CREDIT

Credit can now be obtained, free for a limited time, by reading the review articles in this issue. Please note the following instructions.

Method of Physician Participation in Learning Process: The core material for these activities can be read in this issue of the Journal or online at the JACI Web site: www.jacionline.org. The accompanying tests may only be submitted online at www.jacionline.org. Fax or other copies will not be accepted.

Date of Original Release: April 2013. Credit may be obtained for these courses until March 31, 2014.

Copyright Statement: Copyright © 2013-2014. All rights reserved.

Overall Purpose/Goal: To provide excellent reviews on key aspects of allergic disease to those who research, treat, or manage allergic disease.

Target Audience: Physicians and researchers within the field of allergic disease.

Accreditation/Provider Statements and Credit Designation: The American Academy of Allergy, Asthma & Immunology (AAAAI) is accredited by the Accreditation Council for Continuing Medical Education (ACCME) to provide continuing medical education for physicians. The AAAAI designates this journal-based CME activity for a maximum of 1 *AMA PRA Category 1 Credit*[™]. Physicians should claim only the

credit commensurate with the extent of their participation in the activity.

List of Design Committee Members: Atsushi Kato, PhD, Kathryn E. Hulse, PhD, Bruce K. Tan, MD, and Robert P. Schleimer, PhD

Activity Objectives

1. To understand the development and differentiation of B lymphocytes.
2. To understand the transcription factors, mediators, and signaling pathways involved in B-lymphocyte maturation.
3. To describe the role of B lymphocytes and immunoglobulins in diseases of the respiratory tract.

Recognition of Commercial Support: This CME activity has not received external commercial support.

Disclosure of Significant Relationships with Relevant Commercial

Companies/Organizations: A. Kato and K. E. Hulse have received research support from the National Institutes of Health (NIH). B. K. Tan has received research support from the NIH and the Triological Society, American College of Surgeons. R. P. Schleimer has received research support from the NIH; has received consultancy fees from Intersect ENT, GlaxoSmithKline, Allakos, and Aurasense; and has stock/stock options in Allakos.

Adaptive humoral immune responses in the airways are mediated by B cells and plasma cells that express highly evolved and specific receptors and produce immunoglobulins of most isotypes. In some cases, such as autoimmune diseases or inflammatory diseases caused by excessive exposure to foreign antigens, these same immune cells can cause disease by virtue of overly vigorous responses. This review discusses the generation, differentiation, signaling, activation, and recruitment pathways of B cells and plasma cells, with special emphasis on unique characteristics of subsets of these cells functioning within the respiratory system. The primary sensitization events that generate B cells responsible for effector responses throughout the airways usually occur in the upper airways, tonsils, and adenoid structures that make up the Waldeyer ring. On secondary exposure to antigen in the airways, antigen-processing dendritic cells migrate into secondary lymphoid

organs, such as lymph nodes, that drain the upper and lower airways, and further B-cell expansion takes place at those sites. Antigen exposure in the upper or lower airways can also drive expansion of B-lineage cells in the airway mucosal tissue and lead to the formation of inducible lymphoid follicles or aggregates that can mediate local immunity or disease. (*J Allergy Clin Immunol* 2013;131:933-57.)

Key words: B cells, plasma cells, plasmablasts, respiratory diseases

Discuss this article on the JACI Journal Club blog: www.jacionline.blogspot.com.

Along with the skin and gastrointestinal tract, the respiratory tract is a large surface that interacts extensively with the environment outside the body. The exposed area of the respiratory tract is huge; in human subjects it has a surface area of 500 m², which is roughly the size of a tennis court.¹ Because large volumes of air are moved through the respiratory system rapidly and constantly, there is considerable exposure to airborne organisms that might cause pathology. The nose performs a filtering role, and many bacteria, viruses, and fungi are deposited there. Innate immune responses include passive mechanisms, such as formation of a mucus blanket, mucociliary removal, and swallowing of particles and constitutive expression of host defense molecules by airway epithelium and submucosal glands. Also at play are active innate responses, such as receptor-mediated activation of

From the Departments of ^aMedicine and ^bOtolaryngology–Head and Neck Surgery, Northwestern University Feinberg School of Medicine.

Supported in part by National Institutes of Health grants K23DC012067, R01 HL078860, R01 AI072570, and R37 HL068546 and by a grant from the Ernest S. Bazley Trust.

Received for publication December 26, 2012; revised February 20, 2013; accepted for publication February 21, 2013.

Corresponding author: Robert P. Schleimer, PhD, Allergy-Immunology Division, Department of Medicine, Northwestern University Feinberg School of Medicine, Rm M-318, 240 E Huron, Chicago, IL 60611. E-mail: rpschleimer@northwestern.edu.

0091-6749/\$36.00

© 2013 American Academy of Allergy, Asthma & Immunology

<http://dx.doi.org/10.1016/j.jaci.2013.02.023>

Abbreviations used

AID:	Activation-induced cytidine deaminase
APRIL:	A proliferation-inducing ligand
BAFF:	B cell–activating factor of the TNF family
BAL:	Bronchoalveolar lavage
BCMA:	B-cell maturation antigen
BCR:	B-cell receptor
BLNK:	B-cell linker protein
BTK:	Bruton tyrosine kinase
CD40L:	CD40 ligand
COPD:	Chronic obstructive pulmonary disease
CRS:	Chronic rhinosinusitis
CRSwNP:	Chronic rhinosinusitis with nasal polyps
CSR:	Class-switch recombination
CTD:	Autoimmune connective tissue diseases
CVID:	Common variable immune deficiency
DC:	Dendritic cell
EBI2:	EBV-induced molecule 2
ERK:	Extracellular signal-regulated kinase
GC:	Germinal center
GOLD:	Global Initiative for Chronic Obstructive Lung Disease
HEV:	High endothelial venule
HP:	Hypersensitivity pneumonitis
iBALT:	Inducible bronchus-associated lymphoid tissue
IC:	Immune complex
ILF:	Isolated lymphoid follicle
iNOS:	Inducible nitric oxide synthase
IPF:	Idiopathic pulmonary fibrosis
IRF:	Interferon regulatory factor
LN:	Lymph node
MAPK:	Mitogen-activated protein kinase
MyD88:	Myeloid differentiation primary response gene–88
MZ:	Marginal zone
NALT:	Nasopharynx-associated lymphoid tissue
NF-κB:	Nuclear factor κB
NIK:	Nuclear factor κB–induced kinase
OB:	Obliterative bronchiolitis
OVA:	Ovalbumin
PI3K:	Phosphoinositide 3-kinase
pIgR:	Polymeric immunoglobulin receptor
PIP ₃ :	Phosphatidylinositol-3,4,5-triphosphate
PKC:	Protein kinase C
PLCγ2:	Phospholipase Cγ2
PP:	Peyer patch
RA:	Rheumatoid arthritis
SHM:	Somatic hypermutation
sIgA:	Secretory IgA
SLE:	Systemic lupus erythematosus
SLO:	Secondary lymphoid organ
STAT:	Signal transducer and activator of transcription
SYK:	Spleen tyrosine kinase
TACI:	Transmembrane activator and CAML interactor
T _{FH} :	Follicular helper cells
TLR:	Toll-like receptor

release of host defense molecules by epithelial cells, alveolar macrophages, and other cells; activation of glandular secretion; recruitment of phagocytic cells to the airways; and exudation of vascular fluids. These responses provide a robust defense against all but the most aggressive of potential pathogens. Frequently, innate immune responses do not deter microorganisms, and adaptive immune responses must be marshaled to maintain the integrity of airway function and survival of the host. Adaptive

immune responses in the airways are mediated by B and T cells that express highly evolved and specific receptors. In some cases, such as autoimmune diseases or inflammatory diseases caused by excessive exposure to self-antigens or foreign antigens, these same immune cells can cause disease by virtue of overly vigorous responses.

The purpose of this review is to discuss the cells of the B-cell lineage and their role in disease and immunity in the respiratory system. We discuss the generation, differentiation, signaling, activation, and recruitment pathways of B cells and plasma cells, with special emphasis on unique characteristics of subsets of these cells functioning within the respiratory system. There are some important differences between human subjects and mice, the most studied species, with respect to the organization of B cell–containing tissues and responses (Table I). Nonetheless, much of the best information on the molecular and cellular pathways used by B cells has been derived in the mouse. Likewise, there is considerable information on the natural history of B-lineage cells in the gastrointestinal tract, probably because the vast majority of total bodily antibody production occurs in the gut. Although our greatest interest is in the role of B cells and plasma cells in human airways disease, we have frequently incorporated findings and interpretations that arise from study of the mouse, the gastrointestinal tract, or both, although we have tried not to burden the review by qualifying all such references.

Another important point to make is that B cells have been associated with immunoglobulin responses since their discovery and distinction from T cells in the middle of the 20th century. Although immunoglobulin production remains the most recognized, most studied, and probably most important function of B cells, we would be remiss if we did not point out that impressive recent studies have demonstrated many important roles of B cells that are independent of immunoglobulin production, such as roles in antigen presentation and as regulatory cells akin to regulatory T cells. Very little is known about either of these functions in the respiratory system. We have mentioned these activities where information is available but have focused primarily on immunoglobulin responses. Finally, although there have been a few very valuable reviews of B cells and the respiratory system published, it has become abundantly clear that the study of B-cell biology in the respiratory system, especially in human subjects, is an exceedingly important subject that yeams for significantly more investigation.^{2–4}

OVERVIEW OF THE ADAPTIVE IMMUNE RESPONSE AND IMMUNOGLOBULIN PRODUCTION

B-cell lineages

The majority of B cells develop from lymphoid progenitor cells in the specialized microenvironment of the bone marrow. Early stages of this process are dependent on contact between the developing B-cell precursors and bone marrow stromal cells but do not require antigen. These developmental steps revolve around functional rearrangement of the B-cell receptor (BCR), are tightly regulated by positive and negative selection steps, and result in the formation of immature B cells that are licensed to traffic to peripheral lymphoid tissues. A detailed discussion of this process is beyond the scope of this review but has recently been elegantly presented elsewhere.⁵ An overview of the important structural organization of immune tissues and events that occur in the selection and expansion of B-lineage cells in the airways is provided

TABLE I. Comparative differences between murine and human airway anatomy and B-cell biology

Mouse	Human subject
Molecular	
Express only IgA	Express IgA ₁ and IgA ₂
IgD only found coexpressed with IgM on immature B cells	Monomeric IgD found on memory B cells
Do not express FcαR	Express FcαR
Express pIgR in liver, gut, and lung	Do not express pIgR in liver
Naive B cells express TLR4 and respond to LPS	Naive B cells do not express TLR4 or respond to LPS
BTK knockout: partial B-cell defect	BTK knockout: virtual B-cell defect
Cellular	
Have distinct B1 and B2 cells	B1-like B cells express CD43
CD27 not unique to memory B cells	CD27 a marker for memory B cells
Anatomic	
Rudimentary paranasal sinuses connected with nasal passage	Have multiple enclosed paranasal sinuses separated from nasal passage by relatively small ostia
Diffuse olfactory mucosa on extensively branched ethmoturbinals with limited respiratory mucosa	Focal olfactory mucosa on rudimentary ethmoturbinals comprised predominantly of respiratory mucosa
Have well-developed constitutive NALT and variable constitutive BALT	Morphologically different but constitutive Waldeyer ring in the upper airway but lack of BALT in the absence of disease
No bronchial circulation	Bronchial circulation for potential movement of B-cell lineages

in Figs 1 to 3. Once in lymphoid tissues in the periphery, immature B cells rapidly pass through 2 transitional stages, mainly in the spleen, before committing to either follicular or marginal zone (MZ) B-cell fates. The majority of immature B cells will become follicular IgM⁺IgD⁺ mature naive cells that recirculate through peripheral lymphoid tissues until they encounter their cognate antigen,⁶ after which they can further differentiate into long-lived memory B cells or antibody-secreting plasma cells (discussed below). The remaining immature B cells become MZ B cells and localize primarily in the marginal sinus of the spleen, which positions them to rapidly initiate immune responses against blood-borne pathogens, especially encapsulated bacteria.^{6,7} The decision to commit to one of these 2 fates is dependent on the integration of signals from the BCR, Notch2, and B cell-activating factor of the TNF family (BAFF).⁶ Relatively strong BCR signaling in the presence of BAFF will funnel developing B cells toward a follicular fate, whereas weak BCR signaling in combination with BAFF and Notch2 signals will favor the development of MZ B cells.⁶ However, it is important to note that although human MZ B cells share some features with their murine counterparts, they also have distinct characteristics, including the ability to recirculate through peripheral lymphoid organs and undergo somatic mutations. On the basis of these observations, it is thought that human MZ B cells represent a subpopulation of IgM⁺ memory cells rather than a distinct naive B-cell lineage.⁷

In mice a separate subset of B cells, the B1 B cells, develop in the fetal liver from a precursor cell that is most likely distinct from the one that gives rise to conventional B cells.⁸ These self-renewing cells can be divided into 2 subsets, B1a and B1b cells, based on their expression of CD5 and are part of a family of innate-like B cells that includes MZ B cells.⁹ B1 B cells express high levels of CXCR5 and traffic to the peritoneal and pleural cavities in response to CXCL13, where they represent the majority of the B-cell subpopulations at these sites.^{9,10} These cells can also be found at mucosal sites, including the gut and respiratory tract, as well as in lymph nodes (LNs) and the spleen.⁹ B1 B cells can secrete large amounts of natural IgM and IgA and produce the majority of the natural antibodies that are important for early pathogen

recognition and maintenance of tissue homeostasis.⁹ Natural antibodies are those that are primarily encoded in the germline, are not generated by somatic hypermutation (SHM), and have antigen specificity for a number of naturally occurring epitopes on the surface of microorganisms, including phosphorylcholine, lysophosphatidylcholine, phosphatidylcholine, and LPS.¹¹ These natural antibodies also often have self-reactivity and can contribute to autoimmune responses, as is the case for rheumatoid factor, which contributes to the formation of immune complexes (ICs) and pathogenesis in patients with rheumatoid arthritis (RA).¹² Intriguingly, although BCR signaling is critical for B1 B-cell development, it does not provide sufficient signal strength to activate most B1 B cells. However, they can be readily activated by other signals, including IL-5, IL-10, and Toll-like receptor (TLR) agonists, which trigger their migration to LNs and mucosal sites, where they differentiate into IgM- or IgA-secreting cells.⁹ Exposure to TLR agonists, such as LPS, can mobilize these cells from the peritoneum to peripheral tissues. It is not known whether the population of B1 B cells residing in the pleural cavity includes cells set to selectively migrate to the lung, but this question is worthy of study. However, it has been demonstrated that natural IgM secreted by B1 B cells in the draining LNs of the respiratory tract accounts for the majority of the protective antiviral IgM responses generated during an influenza infection,¹³ indicating that B1 B cells can home to the airways during viral infections.

Despite the accumulating evidence of the importance of B1 B cells in mice, an equivalent subset in human subjects has not been definitively characterized, in part because the classical marker of murine B1 B cells, CD5, is expressed on a variety of human B cells.¹⁴ Recently, however, intriguing new work identified 2 subsets of B1-like B cells in human subjects that express CD43 and have distinct surface phenotypes but share key functional characteristics with murine B1 B cells.¹⁵ The majority of these human B1 B cells do not express CD11b and spontaneously secrete IgM, whereas a smaller fraction of human B1 B cells express CD11b, have potent T cell–modulatory properties, and are increased in patients with lupus.^{14,16,17} Whether a similar population of cells plays a significant role in early antibody production

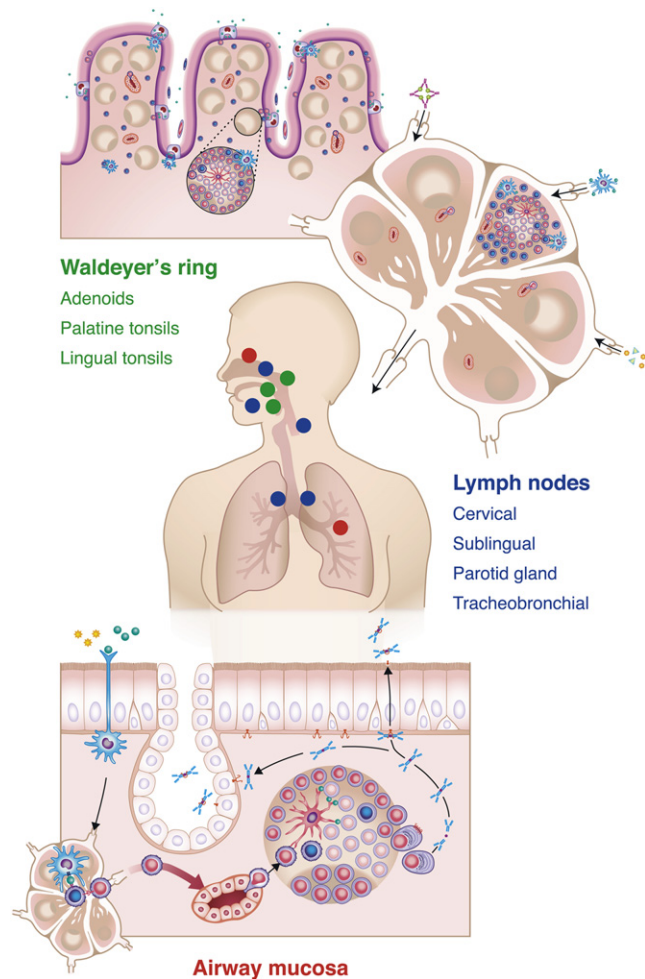


FIG 1. Overview of the organization of tissues involved in the induction, elicitation, and effector responses of B cells and plasma cells in the airways of human subjects. Shown in *green text* are the components of the Waldeyer ring, including the adenoid(s) and palatine and lingual tonsils (shown in *green dots* on the transverse model of a human subject). The *blue text* and *blue dots* in the model indicate LNs that are centrally involved in sensitization and elicitation of airways responses. These include cervical, sublingual, and parotid nodes, as well as tracheobronchial nodes. The *figure at the bottom* displays an effector response in the airways (for details, see Fig 3, A), which could occur either in the lower or upper airways and sinuses, as indicated by the *red dots* in the transverse section of the model. An important point is that the primary sensitization events that generate B cells responsible for effector responses throughout the airways often occur in the upper airways. For further discussion, please see the main text of the review.

and pathogen clearance in the respiratory system or elsewhere in human subjects remains to be determined.

B-cell activation and differentiation in secondary lymphoid organs

It is generally thought that mature naive B cells circulate mainly through secondary lymphoid organs (SLOs) in the periphery until they are activated by an encounter with their cognate antigen, although new evidence has emerged suggesting that these cells can also traffic through nonlymphoid tissues.¹⁸ SLOs are immune induction sites that are developmentally pre-programmed and include LNs, the spleen, and a variety of

mucosal-associated lymphoid tissues (Figs 1-3).¹⁹ Peripheral SLOs share many common organizational features, including follicular B-cell zones with defined germinal centers (GCs; where SHM is believed to primarily occur) that help to ensure optimal cell-cell interactions that favor efficient B-cell activation and production of high-affinity antigen-specific antibodies while providing controls to limit survival of autoreactive clones.¹⁹ Before antigen encounter, naive mature B cells express high levels of the chemokine receptor CXCR5 that allow them to respond to CXCL13 produced by follicular stromal cells and dendritic cells (DCs) and home to the B cell-rich follicles of SLOs.^{20,21} However, the T_H cells that are critical for full B-cell activation and production of high-affinity antibodies are located in a distinct area adjacent to the follicles because of their expression of the chemokine receptor CCR7. Cognate antigen encounter is the first step toward bringing B cells and T_H cells together for the induction of an optimal antibody response. On entry into a follicle, B cells can be exposed to their cognate antigen through a variety of different cell types. BCR signaling through antigen induces upregulation of CCR7 on follicular B cells, and the chemokine CCL21, which is produced by fibroblastic reticular cells and high endothelial venules (HEVs) in the T-cell zone, stimulates them to traffic to the border of the T- and B-cell zones and interact with cognate T_H cells.²¹ Importantly, proper localization of B cells to this border region requires integration of signals from both CCR7 and CXCR5.²⁰ The interaction between T and B cells at this border is highly dynamic (Fig 2, B), as visualized by means of intravital 2-photon imaging of the process, showing active movement of both T and B cell participants.²² B-cell antigen encounter not only stimulates their traffic toward CCL21 in this area but also provides the signals for B cells to upregulate their own antigen presentation and costimulation capacities. As such, although T cells provide help to B cells through CD40–CD40 ligand (CD40L) interaction and cytokine production, B cells provide peptide–MHC class II complexes in the context of costimulatory molecules to T cells, which facilitates their survival and cytokine production.²³ CD11c^{hi} DCs are also found in the border region between the B- and T-cell zones and can contribute to B-cell survival through presentation of antigen and production of BAFF and a proliferation-inducing ligand (APRIL).^{24,25} These initial antigen-specific interactions also play an important role in determining the specific functional and developmental fate to which the B cells will commit.

After the initial interaction with T_H cells, B cells can follow one of 3 alternative fates by differentiating into either extrafollicular plasma cells, GC B cells, or early memory B cells that have the capacity to recirculate.^{23,26} Of these cell types, extrafollicular plasma cells and early memory B cells are formed independently of the GC reaction and therefore likely do not undergo SHM and are similar to the B cells activated during T cell-independent responses. The cells destined to become GC B cells re-enter the follicle to participate in the GC reaction. Although the factors that favor the development of extrafollicular plasma cells or GC B cells are fairly well understood, those that favor the development of early memory B cells are less clearly defined.²⁷ BCR antigen recognition strength and differential expression of chemoattractant receptors are key factors in determining the fate of particular B cells.^{23,28} Sensibly, cells with BCRs that are strongly activated by antigen, either through relatively high affinity or density, will become extrafollicular plasma cells, whereas those that are less strongly activated will enter the GC reaction.²⁸ Those cells

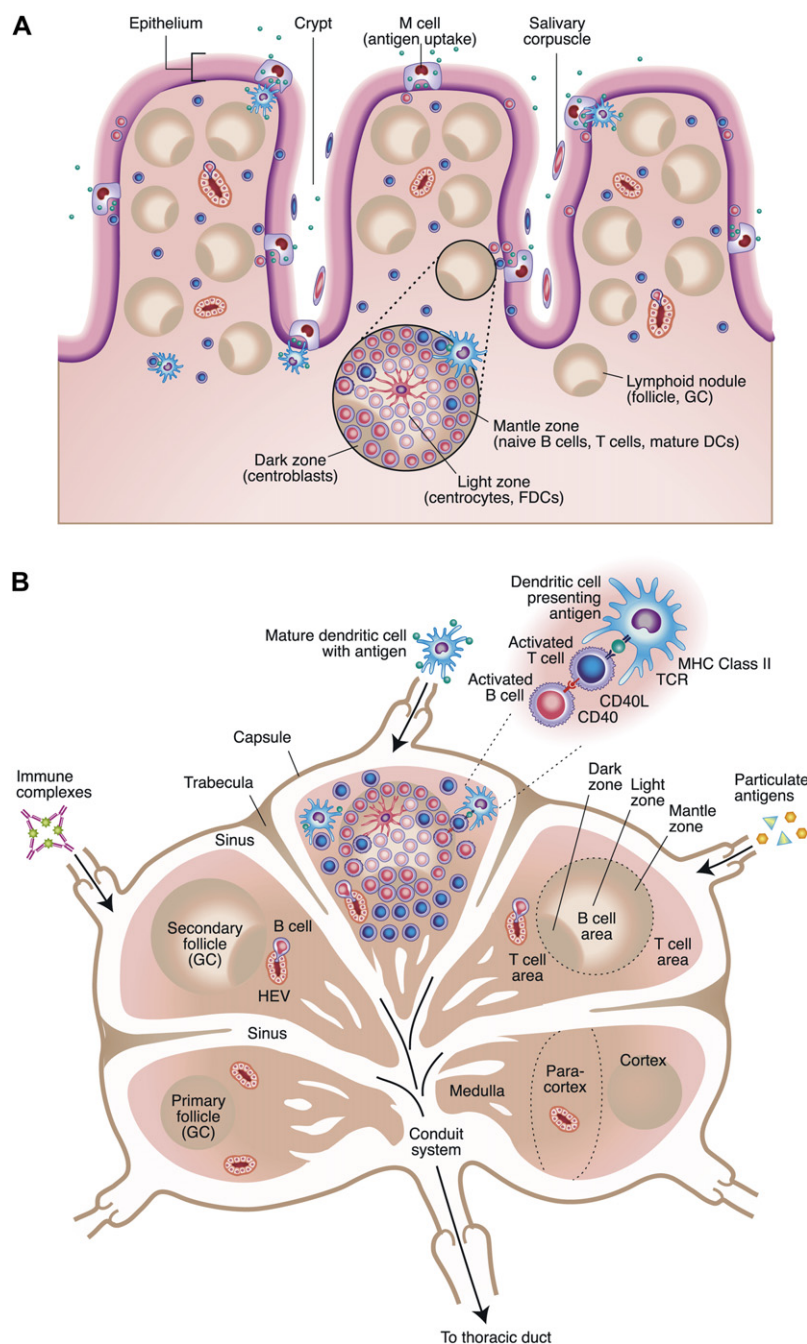


FIG 2. Detailed overview of primary inductive lymphoid tissues contained in the tonsils (**A**, top) and LNs (**B**, bottom). Tonsils and adenoid tissue contain M cells that mediate antigen uptake into the tissue that is rich with lymphoid follicles in which the primary expansion of naïve B cells occurs, followed by the subsequent generation of memory B cells that populate other lymphoid tissues, especially LNs. Dark and light zones containing centroblasts and centrocytes are shown, and the mantle zone, in which DCs, B cells, and T cells collaborate for B-cell activation, are shown in the *magnified inset*. A similar expansion of memory (and naïve) B cells can occur with secondary exposure in the LNs that are draining the airways (Fig 2, B). Detailed discussion of the cellular dynamics in these primary and secondary expansions of antigen-specific B cells is found within the text. *FDC*, Follicular dendritic cells; *TCR*, T-cell receptor.

destined to become extrafollicular plasma cells will also maintain expression of EBV-induced protein 2 (EBI2) and upregulate CXCR4, whereas those destined to become GC B cells will not express EBI2 and upregulate CXCR5.²⁹⁻³¹ Recently, a novel ligand for EBI2 was identified as 7 α ,25-dihydroxycholesterol and

was shown to be critical for the migration of EBI2⁺ cells to extrafollicular sites.^{32,33} Differential expression of specific transcription factors also regulates this fate decision. Whereas high expression of Blimp-1 and interferon regulatory factor (IRF) 4 are required for plasma cell development, high expression of

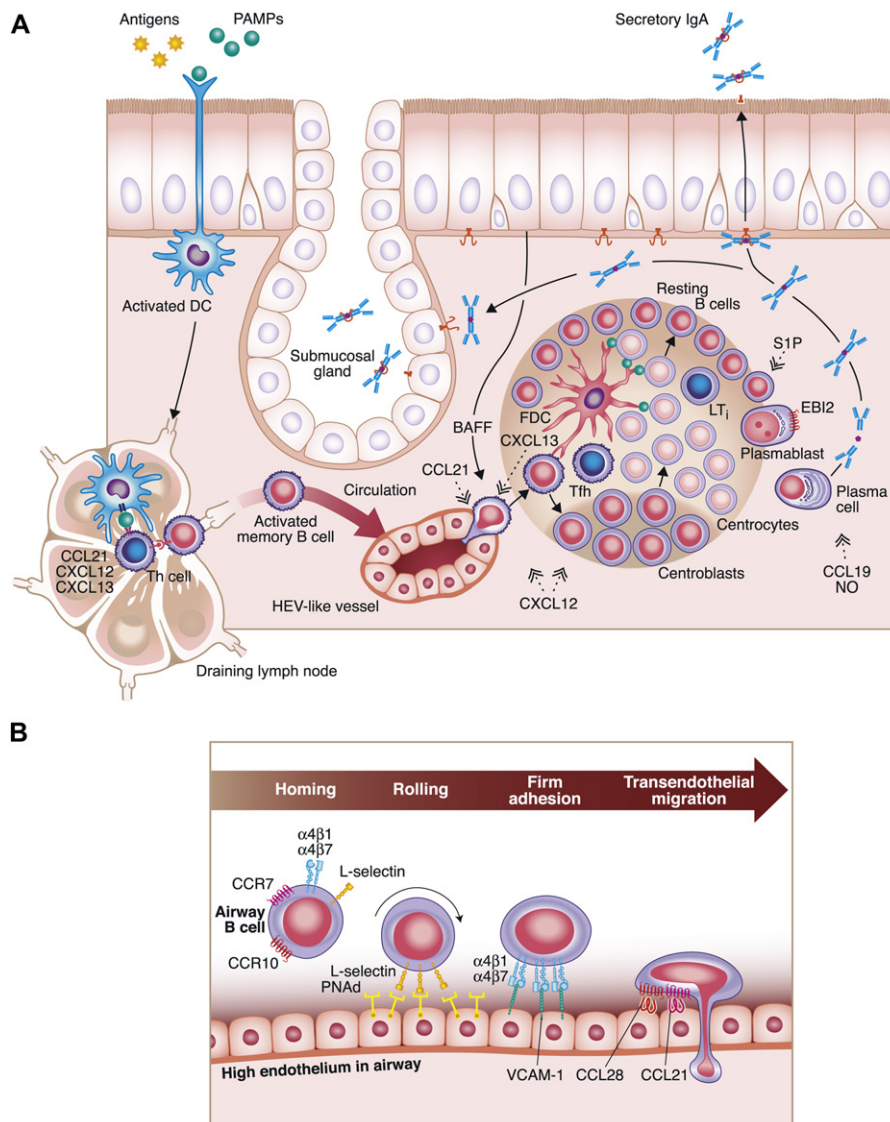


FIG 3. Representative effector response within airway tissue (**A**, top) in which antigen exposure leads to local recruitment of memory (and perhaps naive) B cells (**B**, bottom) and subsequent local expansion of B cells and plasma cells within the airway tissue. The chemokines that drive the recruitment of B cells from the circulation include CCL21, and those involved in localization within quasiorganized inducible lymphoid tissue include CXCL12 and CXCL13, as well as CCL19 and CCL21. The expansion of B cells within the tissue bears a resemblance to the initial induction and expansion of B cells discussed in Fig 1 and in the text. Recruitment of circulating B cells occurs partly through transendothelial migration across the endothelium that resembles HEVs found in LNs. Some of the molecular participants in the rolling, firm adhesion, and transendothelial migration of B cells into the tissue are described in Fig 3, **B**. *FDC*, Follicular dendritic cells.

Bcl-6 is required for GC differentiation, and Blimp-1 and Bcl-6 repress each other (see below).²³ Extrafollicular plasma cells are short lived (3-5 days), but they provide an important source of relatively high-affinity, germline-encoded antigen-specific IgM or IgG antibodies early in the immune response.²⁸ However, in situations of persistent antigen exposure, such as during chronic infections, or with self-antigens, extrafollicular plasma cells have been shown to persist for much longer.³⁴ These cells downregulate CXCR5 while upregulating CXCR4 and maintaining expression of EBI2, a G protein-coupled receptor that binds to ligands derived from cholesterol hydroxylation, allowing them to traffic to splenic bridging channels or LN medullary cords, where they will receive survival signals, such as BAFF and APRIL, from

CD11c^{hi} DCs.^{5,23,32,33} Conversely, GC B cells maintain expression of CXCR5, do not express EBI2, and traffic back to the follicle in response to CXCL13.

The GC reaction is characterized by large amounts of rapid clonal expansion and is generally polarized into 2 distinct zones. The dark zone is proximal to the T-cell zone and contains proliferating B cells, called centroblasts, whereas the light zone is distal to the T-cell zone and contains noncycling B cells (centrocytes).³⁵ This compartmentalization is shown in Figs 2, **B**, and 3, **A**, and is maintained by differential expression and localization of CXCL12 and CXCL13. Centroblasts express higher levels of CXCR4 and therefore traffic to the CXCL12-rich areas of the dark zone, whereas centrocytes express CXCR5 but low levels

of CXCR4 and traffic to the CXCL13-rich areas of the light zone.³⁶ Classically, the GC reaction is thought to proceed in a cyclical fashion wherein centroblasts downregulate surface BCR and undergo SHM while proliferating in the dark zone. They then exit the cell cycle, re-express their newly mutated BCR, and migrate to the light zone as centrocytes. Here, B cells with mutated BCRs that have the highest affinity have a competitive advantage in the presence of antigen and are positively selected by interactions with follicular DCs and T follicular helper (T_{FH}) cells. Positively selected cells can then exit the GC reaction to become long-lived memory B cells or plasma cells or re-enter the cell cycle and migrate back to the dark zone for another round of proliferation, SHM, and selection.³⁷ An alternative view to the above has been suggested in which B-cell trafficking within the GC is predominantly unidirectional, rather than cyclical, because it was found that B-cell re-entry to the dark zone from the light zone was a rare event.³⁸ Although recent studies have demonstrated that S1P and its receptor, S1P2, play a role in restricting localization of B cells in the bone marrow and within the GC by blocking the action of some of the chemokines that otherwise cause them to relocate,^{33,35} the signals that determine whether a particular cell will remain in the GC, leave as a memory B cell, or leave as a plasma cell are unclear.³⁷ However, accumulating evidence suggests that GC output might be regulated temporally so that memory B cells emerge early and plasma cells emerge late during the GC reaction.³⁴ In addition, intriguing new studies have demonstrated that during proliferation, GC B cells asymmetrically divide, resulting in preferential expression of key molecules, such as Bcl-6 and IL-21 receptor, in one daughter cell.³⁹ Although it is still unclear what the functional consequences might be of this asymmetric division, it is tempting to speculate that this might provide cues that could influence the fate of respective daughter cells. Once they leave the GC, memory B cells recirculate through SLOs, are poised to rapidly produce high-affinity antibodies on antigen re-exposure, and can be found months after initial immunization events occur.^{36,37} In contrast, GC-derived plasma cells typically home to the bone marrow or spleen, where they can continually secrete high-affinity antibodies into the systemic circulation for decades.³⁷ T_{FH} cells are critically important to several aspects of GC formation and maintenance, positive selection of high-affinity B-cell clones, and regulation of GC B-cell commitment to either memory B-cell or plasma cell fates.^{36,37,40,41} Although it has been demonstrated that T_{FH} cells can potentially be derived from other T_H lineages, including forkhead box protein 3–positive cells in the Peyer patches (PPs),⁴² it has recently become evident that T_{FH} cells likely represent a separate and distinct lineage of T_H cells.⁴⁰ T_{FH} cells require Bcl-6 expression for their differentiation, specialize in providing help to developing B cells, and require B-cell interactions for their survival.⁴⁰ An excellent comprehensive review of this newly identified T_H cell subtype and its role in the GC reaction has recently been published elsewhere.⁴⁰

B-cell homing and activation in the airway

Classically, it was thought that primary immune responses could only be generated in SLOs, as described above.⁴³ However, studies in lymphotoxin α -deficient mice, which lack LNs and PPs and have a disorganized spleen, have demonstrated that antigen-specific B and T cells can be primed in nonlymphoid tissues.^{44,45} In fact, many studies have revealed the presence of organized

ectopic or tertiary lymphoid-like structures in tissues affected by chronic inflammation, including the airways.⁴⁶ Unlike the well-organized and defined SLOs, ectopic lymphoid structures are relatively less well organized, can develop in a wide variety of tissues and sites, and arise quickly and spontaneously in response to infection, chronic inflammation, or both.^{19,46} Interestingly, however, animal models have demonstrated that the factors required to generate ectopic lymphoid structures are similar to those required for the development of SLOs and include lymphotoxin signaling and localized expression of CXCL12 and CXCL13, among others.⁴⁶

Some of the best-described ectopic lymphoid structures are the so-called isolated lymphoid follicles (ILFs) in the gut. These structures require microbial stimulation for their formation and have been shown to contain large numbers of naive lymphocytes.⁴⁷ This suggests that these non-LN sites where memory B cells can readily be reactivated can also facilitate naive B-cell activation and maturation.⁴⁷ Although ectopic lymphoid structures do generally contain all the necessary cellular and biochemical components to facilitate B-cell activation and production of high-affinity antibodies, as outlined above, and probably evolved as a local mechanism to control infection, their apparent lack of organization has prompted speculation that they might also facilitate abnormal activation of T cells, B cells, or both that could lead to autoimmunity or amplification of chronic inflammation.⁴⁶ Formation of ILFs is seen in the lungs and the upper airways in many normal airways immune responses, as well as in patients with many pathologic conditions. Their presence and potential roles in airways disease are discussed further below.

Class-switch recombination (CSR) is the process by which the heavy chain composition of a specific antibody gene is altered in a single B cell expressing that antibody and plays a critical role in dictating the overall specificity and affinity of the resulting antibodies. This process can occur in a T cell–dependent manner through CD40–CD40L interactions or in a T cell–independent manner through BAFF and APRIL (which are members of the TNF family).⁴⁸ Similar to lymphocyte activation, CSR was thought to only occur in SLOs and to yield circulating memory B cells or effector B cells that express isotypes, such as IgG and IgM and, of particular importance in the airways, IgE and IgA. However, significant data have accumulated to demonstrate that CSR can also occur in the airways. Studies by several groups of investigators have demonstrated this in allergic inflammation, showing that biopsy tissue from the lungs or the nasal cavity of patients with asthma or hay fever express the molecular fingerprint of ongoing CSR and are able to express IgE antibodies *ex vivo*.^{49,50}

Others have also implicated DC-derived inducible nitric oxide synthase (iNOS) as a centrally important enzyme in IgA CSR.⁵¹ iNOS regulates expression of the TGF- β receptor, as well as production of APRIL and BAFF, which all play important roles in IgA class switching.⁵¹ Because very large quantities of NO are produced in the respiratory system, especially in the sinuses and in the vicinity of nasopharynx-associated lymphoid tissue (NALT), this might be particularly important in the nasal cavity.

One of the primary functions of B cells in the airways is the production of immunoglobulins, both within the parenchyma and for export to the mucosal surface of the airway. In particular, airway B cells, like those in the gut, produce polymeric forms of

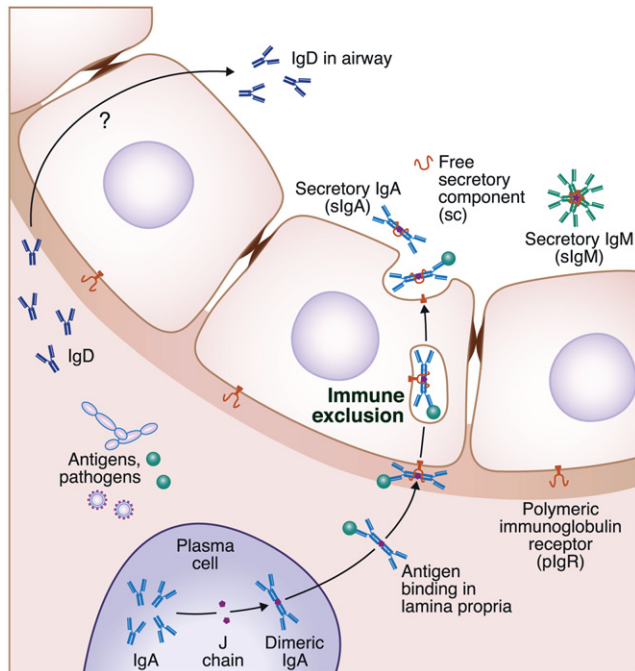


FIG 4. The principle of immune exclusion. Local expression of immunoglobulins of the IgA and IgM isotype can be leveraged for the removal of antigens, allergens, and pathogens through the pIgR. The pIgR binds to multimeric forms of IgA and IgM containing the J chain and moves them across the epithelium through a vesicular transport mechanism that is robust enough to transport whole organisms bound to the immunoglobulin.

IgA and IgM that are structurally maintained by the joining of J chain. Early studies determined that these polymeric immunoglobulins, when detected within the lumen of the airways or gut, also contained a secretory component. This secretory component is added by the action of the polymeric immunoglobulin receptor (pIgR), which binds polymeric IgA or IgM at the basal surface of the mucosal or glandular epithelium and transports the immunoglobulin across the epithelial cell (Fig 4).⁵²⁻⁵⁵ In human subjects lacking IgA (selective IgA deficiency, see below), there is a relatively modest compromise in respiratory immunity because IgM production and possibly IgD production are thought to compensate in the absence of IgA.⁵⁶ However, the absence of pIgR causes a profound increase in respiratory tract infections because both of these secretory antibodies are lacking in the mucosal surface in such patients.^{57,58} The combined action of secretory IgA (sIgA) and secretory IgM constitutes a process referred to as “immune exclusion,” in which antigens and even entire organisms can be carried from the lamina propria across the epithelium and expelled into the lumen of the airways (or gut; Fig 4).

Early studies in human subjects showed that after the oral ingestion of a bacterial vaccine, specific IgA-positive cells appeared in the blood and could be associated with the production of sIgA in saliva, tears, and other sites, leading to the proposal that there exists a common mucosal immune system, one in which B-cell responses in the major mucosal sites would be unified or shared.^{59,60} However, the preponderance of the evidence suggests that B-cell homing is very specific within the airways of human subjects based on a number of studies using tonsillectomy of children as an opportunity to study this question. These studies are elegantly reviewed by Brandtzaeg et al and others.^{4,61,62} Studies compared intratonsillar injection

of a vaccine with peroral administration, intranasal administration, and parenteral injection and found that peroral and parenteral administration yielded little or no antibody-secreting cells in the tonsils, whereas intratonsillar injection not only led to very large local responses but also these responses were primarily restricted to the injected tonsil.⁶³ The tonsils have been referred to as functional analogs of the intestinal PP for the “aerodigestive” tract (ie, the combined proximal locations in which the respiratory and digestive systems are unified) insofar as tonsils are a major site of induction from which B cells that exert effector responses elsewhere in the respiratory system emanate. An early study by Ogra⁶⁴ showed that tonsillectomy is associated with a profound and long-lasting blunting of sIgA production in the nasopharynx. It is now well established that the specific location of antigen encounter and B-cell activation can influence the type of immune response that is initiated, as well as the destination to which the activated cells will preferentially home. As such, the context of the initial activation of a B cell can have lasting implications on the type and location of the resulting immune response. Mucosal surfaces, including the intestines and airways, are continually exposed to foreign microbes and particles and are therefore likely places for B cells to encounter antigens and become activated.

Activation of B cells and their subsequent maturation and homing preferences have been extensively studied in the gut and, to a lesser extent, in the airways. The gut mucosa contains a variety of immune induction sites, including secondary (PPs and draining LNs) and ectopic (ILFs) lymphoid organs. Analogous structures can be found in the airways and include SLOs (NALT and draining LNs) and ectopic lymphoid structures (inducible bronchus-associated lymphoid tissue [iBALT], Fig 1).⁶² Extensive studies in the gut have shown that B-cell activation in PPs results in the formation of IgA-secreting plasma cells that preferentially home back to the gastrointestinal tract.⁵² Furthermore, it has been clearly demonstrated that DCs within the PPs influence both isotype switching and homing preferences through production of retinoic acid, which enhances IgA switching (induced by TGF- β , BAFF, and APRIL) and expression of $\alpha_4\beta_7$ integrin and CCR9.⁵² $\alpha_4\beta_7$ integrin binds to mucosal vascular addressin cell adhesion molecule 1 (MAdCAM-1), which is expressed mainly on intestinal endothelial cells and facilitates critical steps of lymphocyte rolling and entry into the tissue, whereas the ligand for CCR9 (ie, CCL25) is expressed by intestinal epithelial cells and further supports intestinal homing.⁵² Similarly, in the airway it has been shown that B cells activated in the NALT become plasma cells that preferentially switch to IgA and home back to the airway, as well as other mucosal sites.⁶² Although a specific airway-homing molecule, similar to $\alpha_4\beta_7$ integrin in the gut, has not been definitively identified, in some cases homing to the airways might be dependent on $\alpha_4\beta_1$ integrin and CCR10, the ligands of which (ie, vascular cell adhesion molecule 1 [VCAM-1] and CCL28) are expressed in the respiratory tract (Fig 3, B).⁶² Supporting this, a unique IgD⁺IgM⁻ mature B-cell subset that is almost exclusively formed in the NALT has been shown to express CCR10 and home specifically to the airway mucosa but not the gut.^{48,56} Moreover, Cerutti et al⁴⁸ have demonstrated that IgD from these cells binds to mast cells and basophils and recognizes antigens from common respiratory pathogens, suggesting an important role for these unique B cells in immune responses within the airways.⁶⁵ Furthermore, based on the critical role of intestinal DCs in influencing class switching and homing properties, it is reasonable to expect that airway DCs would have a similar

function. It seems unlikely that retinoic acid would be responsible for induction of site-specific homing molecules in both the intestines and airway, but there are many factors associated with IgA class-switching (TGF- β , IL-4, IL-6, IL-10, and iNOS) that could play an analogous role in the airway. As mentioned above, naive B cells are known to continually traffic through SLOs in search of antigen, but there is mounting evidence that naive cells can traffic through nonlymphoid tissues and become activated within the ectopic lymphoid structures there.^{18,47,66} As in ILFs, it has been shown that iBALT can support activation of naive B cells and production of long-lived antibody responses in models of allergic airway inflammation, as well as during influenza infection.⁶⁷⁻⁷⁰

It seems likely that B cells are activated in a variety of locations, including draining LNs, and within the affected mucosal tissue itself during any given immune response. Because B cells activated at distinct sites will have unique properties and homing abilities, this ensures maximal protection against regional re-exposure to a similar antigen. Activation within the mucosa will generate mainly IgA-secreting plasma cells that home back to the tissue and provide long-lasting local immunity, whereas activation in the LNs will generate mainly IgG-secreting plasma cells that home preferentially to the bone marrow and provide systemic protection.⁷¹ In addition, regardless of the location of initial activation, long-lived memory B cells will also be generated that have similar homing patterns to naive cells and can be rapidly activated on antigen re-encounter.

Proper resolution of any immune response is critical to prevent chronic inflammation and tissue damage. When these control measures fail, chronic inflammatory and autoimmune diseases can develop. These diseases are associated with increased production of proinflammatory mediators that perpetuate the cycle of inflammation. As mentioned above, formation of ectopic lymphoid structures has been associated with a variety of inflammatory diseases. In line with this, overexpression of proinflammatory cytokines in the lungs (IL-6 and the IL-6 receptor, TNF, or IL-5) has also been shown to induce formation of iBALT in the absence of antigen.⁷²⁻⁷⁴ Although these studies did not specifically address B-cell activation within iBALT, they did note a large increase in gene transcription of several immunoglobulin genes in response to TNF overexpression,⁷³ as well as the presence of B cells in the follicle-like structures.⁷² These studies highlight the point that factors involved in chronic inflammation can favor induction or perhaps maintenance of ectopic lymphoid structures in the airway, which might prolong or exacerbate inflammation. Moreover, although BAFF and APRIL play critical roles in B-cell survival and CSR during normal immune responses, their presence in chronically inflamed tissues might promote the survival of local plasma cells.⁵² Depending on the disease, these plasma cells could exacerbate inflammation by producing antibodies that activate effector cells, such as mast cells or eosinophils, or provide an uncontrolled source of autoantibodies. Studies aimed at the disruption of chronic local B-cell activation or accumulation in the airway (see below) could provide valuable new targets for the development of improved therapeutic agents.

SIGNALING IN B CELLS AND PLASMA CELLS

IgA is the most abundant immunoglobulin of the healthy respiratory tract and is thought to be the most important immunoglobulin for lung defense. In addition, other immunoglobulins, IgM, IgD, IgG, and IgE, also contribute to the health and disease

of the lung. B cells and plasma cells are the sole producers of immunoglobulins, representing a major component of adaptive immunity. The different isotypes of immunoglobulins have very different functional characteristics, such as the ability to activate complement, resistance to protease degradation, and half-life within tissues and the blood. Antibodies also bind to specific receptors expressed on immune cells, including macrophages, neutrophils, eosinophils, basophils, and mast cells. There are 6 IgG receptors, 1 IgA receptor, and 2 IgE receptors in human subjects that bind to and are activated (or inactivated) by immunoglobulins. Various IgG receptors can trigger phagocytosis and antigen presentation or can inhibit the function of several different cell types. Among the 6 known IgG receptors, Fc γ RIIB is an inhibitory receptor and is the only Fc γ R that is expressed on B cells and plasma cells. The cross-linking of Fc γ RIIB and BCR through ICs induces the termination of antibody production on B cells. The cross-linking of Fc γ RIIB in the absence of BCR can induce apoptosis of B cells and plasma cells.⁷⁵ Receptors for IgE can activate mast cells and basophils to degranulate and cause profound local or systemic anaphylactic inflammation. IgA receptors can play a role in immune exclusion and tolerance mechanisms, as well as activation of inflammatory cells, such as neutrophils and eosinophils. As a consequence of this diversity, differentiation and isotype switching in B cells is exceedingly important and highly regulated. As discussed above, T cell- and protein antigen-dependent activation in GCs leads to formation and expansion of B cells expressing high-affinity antibodies. In addition, B-cell expansion and functions are controlled by T cell-independent activation from DCs or cytokines, inducing generation of low-affinity antibodies, especially IgM and IgA, often against nonprotein antigens, including polysaccharides and glycolipids. In this section we discuss the molecular pathways of differentiation and activation of B cells and plasma cells, as well as the potential therapeutic targets of each pathway relevant to diseases of the respiratory system.

Survival and differentiation of B cells/plasma cells

Activation, differentiation, maintenance of survival, and regulation of programmed cell death in B-cell lineage cells involves a number of cell-surface molecules and transcription factors (Table II). The network of transcription factors involved in the differentiation of B cells and plasma cells has been elucidated through the generation of gene knockout and transgenic mice and microarray analyses in isolated cells. Pax5 and Blimp-1 are well-recognized key transcription factors controlling the differentiation of B cells and plasma cells, respectively.^{23,76,77} Pax5 is a critical factor for B-cell development and maintenance that activates genes associated with B-cell function, including activation-induced cytidine deaminase (AID) and BCR signaling molecules, such as CD19, CD79a, and BLNK.⁷⁷ AID is an essential component of the machinery that mediates CSR, a process in which the diverse variable (V) regions of the immunoglobulin molecule are transferred from one set of constant (C) regions to another. At the same time, Pax5 represses genes associated with plasma cell development and function, including immunoglobulin genes (IgH, IgL, and the J chain) and the transcription factor Xbp1, which induces formation of the secretory apparatus necessary for the production of large amounts of antibody.⁷⁷ In contrast, Blimp-1 is required for plasma cell differentiation and for maintenance of long-lived plasma cells.^{76,77} Blimp-1 controls genes involved in immunoglobulin secretion, including IgH, IgL, the J chain, and Xbp1. Blimp-1

TABLE II. Molecules involved in B-lymphocyte lineage cell responses

Trigger	Receptor	Signaling pathway	Transcriptional factor	B-cell function
CD40L (TNFSF5)	CD40 (TNFRSF5)	Classical NF- κ B	NF- κ B1 (/AID)	Survival, maturation, proliferation, CSR
		Alternative NF- κ B	NF- κ B2	Survival, maturation, proliferation
		JNK/p38	JUN/ATF2/MEF2C?	Survival, proliferation
		?	Pax5/IRF4	CSR, GC formation
Antigen	BCR	Classical NF- κ B	NF- κ B1 (/AID)	Survival, maturation, proliferation, CSR
		ERK	Elk1/CREB	Survival, proliferation
		JNK	JUN/ATF2	Survival, proliferation
		p38	MEF2C	Proliferation
		PI3K	AKT	Survival, proliferation
		Calcineurin	NFAT	Maturation, proliferation
BAFF (TNFSF13B)	BAFFR (TNFRSF13C)	Alternative NF- κ B	NF- κ B2	Survival, maturation, proliferation
		Classical NF- κ B (weak)	NF- κ B1 (/AID)	Survival, maturation, proliferation, CSR
		PI3K	AKT	Survival, proliferation
BAFF (TNFSF13B), APRIL (TNFSF13)	TACI (TNFRSF13B)	MyD88/Classical NF- κ B	NF- κ B1 (/AID)	Plasma cell differentiation, CSR to IgA
		Negative regulation of alternative NF- κ B?	Inhibits NF- κ B2?	Negative regulation to B cells
BAFF (lo), APRIL (hi)	BCMA (TNFRSF17)	Classical NF- κ B	NF- κ B1	Later stage of B-cell differentiation, survival of plasmablasts and plasma cells
Pathogens, (ssRNA/CpG-DNA)	TLR7/9 (endosomal TLR)	MyD88/Classical NF- κ B	NF- κ B1, AP-1, IRF7	Survival, maturation, proliferation, CSR
IL-4	IL-4R α / γ c	Jak1/3	STAT6	CSR to IgG ₄ and IgE
IL-4, IL-13	IL-4R α /IL-13R α 1	Jak1, Tyk2	STAT6/STAT3	CSR to IgG ₄ and IgE
IL-6	IL-6R α /glycoprotein 130	Jak1/2, Tyk2	STAT3	Proliferation
IL-7	IL-7R α / γ c	JAK1/3	STAT5/Pax5	Development, survival, differentiation
IL-9	IL-9R α / γ c	JAK1/3	STAT3/STAT5	Enhance IL-4-dependent CSR
IL-10	IL-10R1/IL-10R2	Jak1, Tyk2	STAT3	CSR to IgG ₁ , IgG ₃ , IgG ₄ , and IgA
IL-21	IL-21R α / γ c	JAK1/3	STAT3/IRF4	Induction of Blimp-1, differentiation to plasma cells, CSR to IgG ₁ , IgG ₃ , and IgA
IL-24	IL-20R1/IL-20R2 and IL-22R1/IL-20R2	Jak1, Tyk2	STAT1/STAT3	Inhibit plasma cell differentiation
TGF- β	TGFB β 1/TGFB β 2		SMAD/RUNX	CSR to IgA
Type 1 interferon	IFNAR1/IFNAR2	Jak1, Tyk2	STAT1/STAT2	Expand short-lived plasma cells
Chemokines, oxysterols	7TMRs (CCRs, CXCRs, EBI2)			B-cell and plasma cell migration

AP-1, Activator protein 1; ATF2, activating transcription factor 2; CREB, cAMP response element-binding protein; JNK, c-Jun N-terminal kinase; MEF2C, myocyte-specific enhancer factor 2C; NFAT, nuclear factor of activated T cells.

also induces the suppression of mature B-cell genes, including Pax5, SpiB, CTIIA, AID, and Bcl-6.^{76,77} On balance, the reciprocal inhibitory effects of Pax5 and Blimp-1 suggest that these 2 factors are central to the molecular events that occur when the B-cell and plasma cell lineages diverge. IRF4 is induced during the plasma cell differentiation stage, and it controls induction of Blimp-1.^{23,77} Therefore it seems likely that IRF4 also plays a critical role in the initiation of plasma cell differentiation. Although it has been reported that Pax5 is induced by IL-7 through the activation of signal transducer and activator of transcription (STAT) 5 and Blimp-1 is induced by IL-21 through the activation of STAT3 and IRF4, it is still not quite clear how activation of Pax5 in B cells and Blimp-1 in plasma cells is controlled.^{78,79}

ACTIVATION: MOLECULAR PATHWAYS AND REGULATORS

The airway is the place first contacted by inhaled antigens. Airway epithelial cells provide a first line of defense against exposure to potentially harmful inhaled allergens, including particulate substances and microbial pathogens.⁸⁰ Local and infiltrating DCs in the airway have a vital role in the initiation of adaptive immune responses to inhaled foreign antigens, and airway

epithelial cells control the recruitment of DCs to epithelium by releasing chemokines, including CCL20.⁸⁰ Studies with tracer molecules, including fluorescein isothiocyanate (FITC)-dextran and FITC-ovalbumin (OVA) clearly show that lung DCs take up antigen in the epithelium and then migrate into the draining LNs in a CCR7-dependent manner to present antigen to T cells.⁸¹ B cells that are simultaneously activated through BCR and TLR downregulate CXCR5 and induce CCR7 and then migrate into the T-cell zone, as described above. In this area B cells are activated by T cells through the interaction of CD40-CD40L and the production of cytokines, resulting in the initiation of proliferation, differentiation, and early immunoglobulin CSR. B-cell activation and CSR also occur locally in airways through both T cell-dependent and T cell-independent mechanisms.² T cell-independent mechanisms are mainly triggered by the TNF family cytokines BAFF and APRIL.^{25,82} In this section we describe the molecular pathways of B-cell activation.

BCR

The BCR is a complex receptor consisting of membrane-bound IgM (or IgD), Ig α (CD79a), and Ig β (CD79b).⁸³ Because membrane-bound IgM has a short cytoplasmic tail, BCR-mediated

signals are transduced by CD79a and CD79b.^{83,84} Cross-linking of membrane-bound IgM induces the phosphorylation of immunoreceptor tyrosine-based activation motif (ITAM) domains on CD79a and CD79b by Src family kinases, including Lyn, and it leads to the recruitment and activation of spleen tyrosine kinase (Syk).⁸²⁻⁸⁴ Activation of Syk is a critical event in BCR signaling, leading to the formation of a plasma membrane-associated signaling complex that contains adaptor molecules (including B-cell linker protein [BLNK]) and multiple tyrosine kinases (including Lyn and Bruton tyrosine kinase [BTK]). Phosphorylation of BLNK induces the activation of Ras, phospholipase C γ 2 (PLC γ 2), and BTK, contributing to the activation of distinct signaling pathways. Activation of Ras leads ultimately to phosphorylation of extracellular signal-regulated kinase (ERK), which in turn activates the transcription factors Elk1 and cAMP response element-binding protein (CREB) that induce cell proliferation and inhibit expression and function of apoptotic proteins.^{84,85} Activation of PLC γ 2 induces the production of inositol-1,4,5-triphosphate, which is required for the subsequent release of cytosolic Ca²⁺, and diacylglycerol, which in turn is required for the activation of protein kinase C (PKC) β .^{84,86} Intracellular Ca²⁺ release and activation of PKC β induce phosphorylation of mitogen-activated protein kinases (MAPKs), ERK, c-Jun N-terminal kinase (JNK), and p38 and then lead to the activation of the transcription factors Elk1, CREB, monocyte-specific enhancer factor 2C (MEF2C), JUN, and activating transcription factor 2 (ATF2). Increased cytosolic Ca²⁺ also induces the activation of the transcription factor nuclear factor of activated T cells (NFAT) through the calmodulin/calcineurin pathway.^{82,86} The activation of PKC β also induces phosphorylation of I κ B kinase, leading to activation of the transcription factor nuclear factor κ B1 (NF- κ B1) through the induction of the classical NF- κ B pathway.⁸² NF- κ B1 induces the expression of antiapoptotic genes and AID (see below).

Complement activation is an essential process of the early response to infection. The complement cleavage product C3d enhances adaptive immunity through binding to complement receptor 2 (CD21), which is a part of the B-cell coreceptor complex.⁸⁷ Activation of the B-cell coreceptor complex, consisting of CD21, CD19, and CD81, enhances the BCR-dependent reaction.^{82,87,88} Phosphorylation of CD19 results in the recruitment of Lyn, which can efficiently amplify BCR signaling by enhancing the phosphorylation of CD79a/b.⁸⁹ Phosphorylated CD19 also activates phosphoinositide 3-kinase (PI3K), which in turn further induces the production of the lipid phosphatidylinositol-3,4,5-triphosphate (PIP₃).⁸⁸ PIP₃ recruits a number of BCR signaling components, including PLC γ 2 and BTK, to the plasma membrane and activates them. PIP₃ also recruits and activates the serine/threonine kinase AKT, which in turn activates the NF- κ B pathway and inhibits glycogen synthase kinase 3 (GSK-3), a negative regulator of MYC and D-type cyclins. AKT also inactivates the forkhead box class O1 transcription factor that targets cell-cycle arrest genes, such as cyclin G2 and Rb12.⁹⁰

BCR-related signaling is critical for early B-cell development, and mutations of BCR-related genes results in primary B-cell deficiencies. Mutations in BTK, CD79a, CD79b, IgLL1, and BLNK account for approximately 90% of patients with X-linked or autosomal recessive agammaglobulinemia, which is characterized by defects in early B-cell development, and 85% of these result from mutations of the BTK gene.⁹¹ Mutations in CD19 or CD81 result in hypogammaglobulinemia.⁹¹⁻⁹³ As discussed below, these B-cell deficiency diseases often result in frequent and

severe upper and lower respiratory tract infections because of the importance of an intact immunoglobulin response to immunity in the respiratory system.

BCR-related signaling molecules are one of the key groups of therapeutic targets for B cell-related diseases. Several small compound inhibitors for kinases, including Syk (R788), BTK (PCI-32765 and GDC-0834), PKC β (enzastaurin), Bcr-Abl/Lyn (bafetinib), p38 MAPK (pamapimod, BMS-582949, SB-681323, and SD0006), MEK/ERK (RO5068760, GSK1120212, AZD6244, and CH4987655), and Akt (MK2206, GSK2141795, PBI-05204 and SR13668), are currently in clinical trials for B cell-related diseases, including RA, systemic lupus erythematosus (SLE), B-cell lymphoma, and leukemia.⁹⁴⁻¹⁰⁴ Bortezomib is the first therapeutic proteasome inhibitor that suppresses NF- κ B signaling in the treatment of human cancer.¹⁰⁵ Although none of these inhibitors except SB-681323 are currently under study specifically for respiratory disease, they will likely become therapeutic options for B cell-related airway diseases, as discussed below (Table III).⁹⁴⁻¹¹⁵

CD40

T cell-mediated activation of B cells is generally controlled by CD40-CD40L interactions. CD40 is a member of the TNF family (TNFRSF5) and is constitutively expressed on B cells. The engagement of CD40 by CD40L, which is expressed on activated T_H cells, promotes the recruitment of TNF receptor-associated factor (TRAF) family molecules, leading to the activation of multiple signaling pathways, including both classical and alternative NF- κ B pathways, MAPKs, PI3K, and PLC γ pathways.^{23,116,117} One of the key gene products of the CD40 pathway is AID, a critical factor for SHM and immunoglobulin CSR.¹¹⁸ Knockout of either the CD40 or CD40L gene in mice results in profound defects in antibody production and CSR, and mutations of the CD40L gene in human subjects result in the disease called X-linked hyper-IgM syndrome, in which excess IgM is produced because of a defect in subsequent loss from the IgM⁺ cell population as CSR occurs.^{116,119} CD40 signaling in B cells also promotes GC formation and differentiation to memory B cells and long-lived plasma cells.¹¹⁷

Interference with the CD40-CD40L interaction is a potential target either to augment CD40 signaling for patients with immunodeficiency or to block signaling in patients with chronic inflammatory diseases.¹²⁰ Blockade of CD40 signaling with mAbs has been shown to prevent or improve inflammatory diseases in several animal models. Antibodies against CD40L are also known to induce tolerance in mouse airway graft models.¹²¹ Various mAbs against CD40 and CD40L are undergoing clinical trials to test their safety and potential clinical benefits in patients.¹²⁰

BAFF/APRIL

BAFF (also known as BLyS and TNFSF13B) and APRIL (also known as TNFSF13) play centrally important roles in B-cell function. Studies of deficient mice indicate that BAFF is an essential factor for B-cell maturation and survival and that APRIL is an important factor for CSR to IgA.^{25,82,122} BAFF and APRIL share 2 receptors: transmembrane activator and CAML interactor (TACI) and B-cell maturation antigen (BCMA). TACI binds to BAFF and APRIL with moderate affinity, whereas BCMA has high affinity for APRIL and low affinity for BAFF. In addition, BAFF also binds to the high affinity receptor BAFF-R. BAFF-R

TABLE III. Drugs targeting B-lymphocyte lineage cell-related molecules

Target	Drug	Diseases	References
BCR related			
Syk	Fostamatinib (R788)	RA (phase III), lymphoma (phase II)	95
BTK	PCI-32765	Lymphoma, leukemia (phase II/III)	94
	GDC-0834	RA (phase I)	100
PKC β	Enzastaurin	Non-Hodgkin lymphoma (phase III), cancer (phase II)	104
Bcr-Abl/Lyn	Bafetinib	Leukemia (phase II)	97
p38 MAPK	Pamapimod	RA (phase II)	95
	BMS-582949	RA, psoriasis (phase II)	95
	SB-681323	RA, ARDS, COPD (phase II)	95, 103
	SD0006	RA (phase I)	103
MEK/ERK	Trametinib (GSK1120212)	Cancer (phase II/III)	96
	Selumetinib (AZD6244)	Cancer, MM (phase II)	102
	RO5068760	Cancer (phase I)	98
	CH4987655	Cancer (phase I)	99
Akt	MK2206	Cancer (phase II)	101
	GSK2141795	Cancer (phase I)	101
	SR13668	Cancer (phase I)	101
	PBI-05204	Cancer (phase I)	101
NF- κ B	Bortezomib	Cancer (approved)	105
Cytokines			
BAFF	Belimumab	SLE (approved)	106
BAFF	Blisibimod (A-623)	SLE (phase II/III)	107
BAFF	Tabalumab (LY2127399)	SLE, RA, MM (phase III)	107
BAFF	Briobcept (BAFFR-Ig)	RA (phase I)	107
BAFF/APRIL	Atacicept (TACI-Ig)	RA, SLE (phase II/III), MS, RA (phase II)	95, 107
IL-6	Siltuximab	Cancer (phase II)	113
IL-6R	Tocilizumab	RA (market)	112
IL-9	MEDI-528	Asthma (phase II)	112
IL-13	Lebrikizumab	Asthma (phase III)	109, 111
	Tralokinumab	Asthma, ulcerative colitis (phase II)	109
IL-13/IL-4	Pitrakinra (mutated IL-4)	Asthma, atopic eczema (phase II)	110
IL-21	NNC114-0005 (fully human anti-IL-21 mAb)	RA (phase I)	114
IFN- α	Rontalizumab	SLE (phase II)	115
	MEDI-545	SLE (phase II)	115
Other			
CD20	Rituximab	RA, non-Hodgkin lymphoma and B-CLL (approved)	115
	Ofatumumab	SLE (phase III)	
		Chronic lymphocytic leukemia (approved)	115
	Ocrelizumab	RA (phase III), MS (phase II)	
		RA, SLE, MS (phase III)	115
	Veltuzumab	RA, lymphoma (phase II),	115
	TrU015	RA (phase II), SLE (phase I)	115
CD22	Epratuzumab	SLE (phase III)	115
IgE	Omalizumab	Asthma (approved)	108
	MEMP1972A (anti-M1 prime)	Asthma (phase II), AR (phase I)	109

ARDS, Acute respiratory distress syndrome; B-CLL, B cell chronic lymphocytic leukemia; MEK, MAPK/ERK kinase; MM, multiple myeloma.

is a potent regulator of mature B-cell survival and also controls T cell-independent immunoglobulin production. Loss-of-function mutations of BAFF-R result in impairment of B-cell development, T cell-independent responses, and immunoglobulin production, except for IgA responses in human subjects.^{93,123} Binding of BAFF to BAFF-R activates 2 major pathways: the alternative NF- κ B pathway and the PI3K/AKT pathway. Activation of BAFF-R induces the recruitment of the TRAF3/NF- κ B-induced kinase (NIK) complex and degrades TRAF3 in a TRAF2-dependent manner. Degradation of TRAF3 leads to the stabilization of free NIK and the activation of NF- κ B2, which in turn induces genes involved in B-cell survival. The PI3K/

AKT pathway promotes B-cell survival, as well as B-cell growth. In contrast to BAFF-R, the downstream signaling pathways of TACI and BCMA are poorly understood. TACI is an inhibitory receptor for the general B-cell population, although the mechanism of this negative effect remains to be elucidated. TACI interacts with several TRAFs, including TRAF3, which acts as a negative regulator of the alternative NF- κ B pathway by maintaining low levels of cellular NIK.¹²⁴ This might be a pathway of TACI-mediated negative regulation. TACI also interacts with myeloid differentiation primary response gene-88 (MyD88), leading to activation of the classical NF- κ B pathway to promote plasma cell differentiation and immunoglobulin CSR, especially to

IgA.¹²⁵ TACI mutations are significantly associated with common variable immunodeficiency (CVID), although they are likely to require additional genetic contributions because genetic mutation alone does not necessarily lead to a CVID phenotype.¹²⁶ BCMA strongly activates the classical NF- κ B pathway and promotes later stages of B-cell differentiation and survival of plasmablasts and long-lived plasma cells, although the details of the signaling pathways involved remain unclear.¹²⁷

The cross-talk between BCR and BAFF-R signaling pathways plays a key role in B-cell survival.^{82,84} The BCR is the main activator of the classical NF- κ B pathway, whereas BAFF-R primarily activates the alternative NF- κ B pathway. Because it is now known that both of these NF- κ B pathways are required for B-cell development and survival, the participation of both the BCR and BAFF-R is believed to be key. Consequently, disruptions or genetic abnormalities that affect either the BCR, the production of ligands for the BAFF-R, or the signaling of either receptor can have profound effects on B cells in homeostasis or disease.

BAFF and APRIL might be involved in the pathogenesis of airway inflammatory diseases. It has been reported that BAFF levels are increased in the setting of asthma, chronic obstructive pulmonary disease (COPD), and chronic rhinosinusitis with nasal polyps (CRSwNP).¹²⁸⁻¹³⁰ In a murine model of asthma, treatment with TACI-Ig, which blocks both BAFF- and APRIL-dependent signals, prevents airway inflammation and is more effective than treatment with anti-IgE in reducing airway hyperresponsiveness to inhaled antigen.¹³¹ Importantly, anti-BAFF (belimumab; Benlysta, GlaxoSmithKline, Research Triangle Park, NC) has been recently approved for the treatment of SLE, and TACI-Ig (atacept) is under clinical trials for B cell–related diseases, including RA, SLE, and B-cell lymphoma.^{95,106,107} These and other new drugs can become therapeutic options for BAFF or B cell–driven respiratory diseases in the future (Table III).

TLRs

The innate immune response is the first line of host defense and is responsible for immediate recognition and control of microbial invasion. One of the primary families of molecules responsible for innate immunity in human subjects are the TLRs, homologues of Toll in *Drosophila* that recognize pathogen-associated molecular patterns of microbial organisms.¹³² To date, 10 human TLRs have been identified. Among them, TLR1, TLR2, TLR6, TLR7, TLR9, and TLR10 are expressed on human B cells, and TLR1 through TLR9 are expressed on human plasma cells.^{133,134} Although LPS, which is a TLR4 ligand, is well known to induce immunoglobulin production in mouse B cells, TLR4 is absent on human B cells, except malignant B cells, suggesting that LPS does not directly activate human B cells. In addition, the expression of TLR2 in naive B cells is low. In general, bacterial cell-wall components that mainly activate cell-surface TLRs (TLR1/2, TLR2/6 and TLR4) do not strongly activate human B cells.¹³³ In contrast, endosomal TLRs, such as TLR7, a receptor for single-stranded RNA, and TLR9, a receptor for unmethylated CpG motifs containing DNA, are highly expressed in B cells.¹³²⁻¹³⁴ The activation of TLR7 and TLR9 by RNA and DNA initiates signaling through recruitment of the adaptor molecule MyD88 and the sequential activation of NF- κ B, MAPKs, activator protein 1, and IRF7, resulting in B-cell proliferation and immunoglobulin production.¹³²⁻¹³⁴ However, many vaccine adjuvants contain ligands for surface TLR, including monophosphoryl lipid A, suggesting

that TLR activation on other important cells, such as DCs, tissue structural cells, or T cells, can promote robust B-cell responses.^{132,133} Interestingly, a recent study showed that B cells respond rapidly during sepsis and play an important role in a rapid innate immune response through an interferon activation pathway downstream of TLR activation.¹³⁵

ICs that are formed from the multivalent binding of an autoantibody to an autoantigen are known to be pathogenic in patients with autoimmune diseases. DNA- or RNA-binding ICs are potent IFN- α inducers in plasmacytoid DCs through the activation of Fc γ RIIa and TLR7/9.¹³⁶ This is now recognized to be one of the important pathogenic mechanisms for autoimmune diseases, including SLE and systemic sclerosis.¹³⁶ In addition, DNA- or RNA-binding ICs also potentially activate autoreactive B cells through sequential engagement of BCR and TLR7/9 and play a key role in autoantibody responses in patients with SLE.¹³⁷⁻¹³⁹ SLE is considered a multisystem disease. The pulmonary system in patients with SLE is vulnerable to injury, in part mediated by ICs.¹⁴⁰ ICs are also known to initiate acute lung injury in patients with hypersensitivity pneumonitis (HP).¹⁴¹ Although mechanisms of IC-related inflammation in these diseases are well studied in regard to Fc γ R-related and complement-mediated reactions, BCR and TLR7/9 might also be involved in IC-mediated inflammation.

Cytokines

The expression of antibodies and cytokines is involved in homeostasis and disease in the lung. Although CSR is highly regulated by the CD40, BAFF/APRIL, and TLR pathways, cytokines control immunoglobulin isotype switching. IL-4 and IL-13 induce CSR to IgG₄ and IgE through activation of STAT6.¹⁴² IL-9 enhances IL-4–dependent IgE production in B cells through activation of STAT3 and STAT5, although IL-9 alone does not have this effect.¹⁴³ TGF- β induces CSR to IgA through activation of Smad and Runx.¹⁴² IL-10 is known to induce CSR to IgG₁, IgG₃, IgG₄, and IgA.¹⁴² IL-21 induces CSR to IgG₁, IgG₃, and IgA and also enhances IL-4–dependent CSR to IgE through activation of STAT3.^{144,145} Increases in IL-4 and IL-13 levels and subsequent induction of IgE from B cells in the lung are well-known factors in the initiation and exacerbation of bronchial asthma. An anti-IgE antibody, omalizumab, is approved for the therapeutic treatment of allergic asthma, emphasizing the importance of IgE-producing B cells in this disease.¹⁰⁸ However, selective inhibition of IL-4 for the treatment of asthma has thus far not been effective. Currently, targeting of IL-13 alone or the combination of IL-13 and IL-4 is being pursued by using a humanized anti-IL-13 antibody (lebrizumab), a human anti-IL-13 antibody (tralokinumab), and a mutated IL-4 (pitracinra).¹⁰⁹ Pitracinra blocks the effect of both IL-4 and IL-13. Pitracinra and lebrizumab showed promising effects in the treatment of asthma in clinical trials.^{110,111} IgE-producing B cells will be targeted in a phase II trial by anti-M1 prime, an antibody that binds to the unique extracellular domain of IgE found on the cell-surface form.¹⁰⁹ IL-9 is a classical T_H2 cytokine and now is known to be produced by a novel T-cell subtype called T_H9. In addition to its effects on B cells, IL-9 is known to be involved in the growth and differentiation of mast cells, playing a role in allergic airway inflammation and fibrosis.¹¹² An anti-IL-9 humanized antibody (MEDI-528) is in clinical trials for the treatment of asthma.¹¹²

Several cytokines promote B-cell function. IL-6 is an important factor in the proliferation of B cells and plasma cells through activation of STAT3.¹⁴⁶ Levels of IL-6 are increased in patients with several airway inflammatory diseases, including asthma, COPD, and CRSwNP.^{112,147} A humanized anti-IL-6 receptor antibody, tocilizumab, is approved for the therapeutic treatment of RA and has been proposed for clinical trials in patients with asthma and COPD.¹¹² An anti-IL-6 antibody (siltuximab) is in clinical trials for the treatment of cancer.¹¹³ Although IL-7 is absolutely required for human T-cell development, IL-7 also promotes B-cell development through the activation of STAT5 and Pax5.¹⁴⁸ IL-21 is an important regulator of the proliferation and survival of B cells.¹⁴⁹ In addition, IL-21 regulates differentiation of B cells to plasma cells. A fully human anti-IL-21 antibody is in clinical trials for RA.¹¹⁴ IL-17-producing T cells have been demonstrated to be potent activators of B-cell help, and approaches to blocking IL-17 might have value in the treatment of autoimmune diseases that involve B cells, such as SLE.^{150,151} IL-24 inhibits plasma cell differentiation, and strategies to emulate this effect might have some promise.¹⁵² Type I interferons promote expansion of short-lived plasma cells and are known to play a pathogenic role in SLE.¹⁵³ A humanized anti-IFN- α antibody (rontalizumab) and a fully human anti-IFN- α antibody (MEDI-545) are in clinical trials for SLE.¹¹⁵ Earlier in the pipeline are compounds being developed to inhibit the receptor EBI2, which appears to be nearly essential for early plasmablast development.

Although we do not discuss the details of the mechanisms, several B-cell depletion therapies have been approved or are in clinical trials. A chimeric anti-CD20 antibody, rituximab (Rituxan; Genentech, South San Francisco, Calif), is approved for the therapeutic treatment of RA, non-Hodgkin lymphoma, and chronic lymphocytic leukemia.^{115,154} Other humanized and fully human anti-CD20 antibodies are in clinical trials for RA and SLE. CD22 is an inhibitory coreceptor of the BCR that is exclusively expressed on B cells. It plays a key role in setting the threshold of BCR responses.¹¹⁵ A humanized anti-CD22 causes partial B-cell depletion and is currently in clinical trials for SLE.¹¹⁵ Importantly, the treatment of patients with Churg-Strauss syndrome with rituximab has resulted in a rapid and substantial reduction in disease activity.¹⁵⁵ Neutralization of BAFF by a soluble receptor (Atacicept) or specific antibody (Benlysta) might also have some promise in the treatment of asthma (see above). The progress in the field suggests that B-cell depletion or B cell-targeting therapies might become therapeutic options for respiratory diseases in the future (Table III).

ORGANIZATION AND STRUCTURE OF B LYMPHOCYTES IN HUMAN AIRWAY

The organization and density of B lymphocytes in the normal physiologic state exhibits significant variation across species and age. In healthy human subjects classic nasal-associated lymphoid tissue (NALT), consisting of lymphoid follicles associated with lymphoepithelium, is frequently found in infants but, unlike in mice, is not found in adults.¹⁵⁶ In the upper airways of human subjects, most mucosal-associated lymphoid aggregates are found in the Waldeyer ring, which consists of the adenoid, tubal, palatine, and lingual tonsils (Fig 1).¹⁵⁷ Although limited experimental evidence suggests that these sites are functional analogs of the

NALT found in mice and other rodents, rigorous comparisons between the functional immunologic characteristics of these structures are still lacking.¹⁵⁸ Interestingly, the structure and tight junctions used by the overlying lymphoepithelium are uniquely specialized in each site, with pseudostratified ciliated columnar epithelium found in the adenoid and stratified squamous epithelium predominating in tonsillar tissue.¹⁵⁹ Although not as clearly delineated as in the gut, epithelial cells with some immunohistologic and electron microscopic characteristics of M cells are reported in human tonsils and adenoid,^{159,160} although experimental evidence for their role as antigen-transporting cells in the nasopharynx has only been described in mice.¹⁶¹ Furthermore, it appears that the classic immunohistologic markers for M cells in the gut, namely vimentin and cytokeratins 8 and 18, are not specific markers for M cells in the upper airway, thus complicating efforts to study the antigen-transporting characteristics of these cells.¹⁶² Other lymphoid aggregates are also found in the human larynx epithelium (termed laryngeal-associated lymphoid tissue) and have been shown to persist into adulthood.¹⁶³

In the lower airway bronchial-associated lymphoid tissue (BALT) is found rarely in the lungs of human fetuses without the presence of intrauterine infection^{164,165} or healthy adults without a history of pulmonary disease.^{166,167} This situation is unlike that in rats, where BALT is found in germ-free pups as early as 4 days after birth along bifurcations of the upper bronchi. BALT in human subjects was frequently found to be homogeneously distributed in the lungs of children (36% to 44%) who died of sudden infant death syndrome or trauma, suggesting that development of BALT in human subjects results from strong antigenic stimulation.¹⁶⁸ It was also found in adults after infection in the setting of chronic inflammation caused by smoking¹⁶⁶ or occlusion of the airway by tumor.¹⁶⁹ Regardless of the differences in organization and density of NALT and BALT in human subjects, palatine tonsils, and possibly the adenoid, are thought to be highly efficient sites of antigen uptake and inductive sites for airway-specific humoral immune responses. A previous study compared intratonsillar vaccination and intranasal vaccination of patients who were about to undergo a tonsillectomy with peroral and parenteral vaccination with tetanus and cholera toxin. They demonstrated that the intratonsillar and intranasal routes were both efficient inducers of systemic specific antibodies but that mucosal immunity was conferred only to the upper airway, possibly because of preferential use of adhesion molecules and chemokines among upper airway mucosal B cells. Activation and induction of NALT and BALT are also prominently found in patients with inflammatory airway disease and are further discussed in the following sections.^{2,167}

IMMUNOGLOBULINS IN THE HUMAN AIRWAY AND HUMORAL IMMUNODEFICIENCIES

In human subjects IgA is the major immunoglobulin of the healthy upper and lower respiratory tract, but unlike the lower gastrointestinal tract, IgD and IgG represent approximately 25% of nasal immunoglobulins.¹⁷⁰ Human subjects, unlike mice, express 2 forms of IgA in airway secretions: the IgA₁ subclass is predominant at both sites, but nasal airway secretions typically have less IgA₂ than bronchial secretions.⁴⁸ In human subjects IgD comprises approximately 10% of nasally produced immunoglobulins, although secretion into the small intestine is minimal.¹⁷⁰ Although first discovered more than 50 years ago, IgD was

recognized until recently largely for its presence on naive B cells. However, recent findings demonstrate that secreted IgD is produced largely by IgD⁺IgM⁻ B cells with a plasmablast phenotype found in the human nasal mucosa and tonsils.⁶⁵ Interestingly, IgM-to-IgD class-switching occurs through a noncanonical mechanism and can occur both in a T-dependent or T-independent mechanism requiring AID. The secreted IgD antibodies are frequently polyreactive and recognize respiratory bacteria, such as *Moraxella catarrhalis* and *Haemophilus influenzae* but, surprisingly, have a high rate of autoreactivity.¹⁷¹ Furthermore, IgD has interesting properties that enable it to interface between the adaptive and innate immune responses as it binds to peripheral basophils and tonsillar mast cells through an as yet undescribed receptor and is capable of triggering degranulation on IgD cross-linking.⁶⁵

The isotypes, quantity, and transport of immunoglobulins into the airway each play a critical role in maintaining the equilibrium between immunity and inflammation. Patients with selective IgA deficiency do have marginally increased rates of infection but also have a relatively modest compromise in respiratory immunity, possibly because IgM, IgG, and IgD compensate in the absence of IgA.⁵⁶ In patients with CVID, affected subjects have mutations in one of several genes, including BAFF-R and TACI, and consequently have decreased levels of IgA and IgG but often normal serum levels of IgM.¹⁷² Despite an intact transepithelial transport of IgM by pIgR, affected subjects have recurrent pneumonia, otitis media, sinusitis, and septicemia, with particularly impaired immunity against encapsulated bacteria, such as *Haemophilus influenzae*, *Streptococcus pneumoniae*, and *Staphylococcus aureus*.^{173,174} Paradoxically, these patients also exhibit higher rates of autoimmune diseases, such as immune thrombocytic purpura and autoimmune hemolytic anemia. More profound demonstrations of the critical role immunoglobulins play in airway immunity can be found in the clinical manifestations of X-linked agammaglobulinemia, in which a nearly complete absence of immunoglobulins causes a profound increase in respiratory tract infections beginning in infants once the effects of maternally derived IgG fade.⁹¹

Conversely, conditions resulting in the overproduction of specific isotypes of immunoglobulins also have a significant effect on airway health. The X-linked hyper-IgM syndrome is caused by a defect in the CD40L gene that results in normal-to-increased levels of serum IgM and low-to-undetectable IgG, IgD, IgA, and IgE levels.¹⁷⁵ Affected subjects have increased susceptibility to upper respiratory tract infections and autoimmune manifestations in addition to opportunistic pulmonary infections by *Pneumocystis carinii*, which are likely secondary to the effects of CD40L deficiency on T-cell function. A different pattern of disease is found in patients with the hyper-IgE syndrome, which results from mutations in STAT3, with affected subjects having eczema, mucocutaneous candidiasis; recurrent staphylococcal abscesses of the skin, lungs, and viscera; and increased serum IgE concentrations.^{174,176} These immunodeficiencies likely culminate from the critical role of STAT3 signaling in the differentiation and generation of memory T and B cells.¹⁷⁷⁻¹⁷⁹ Finally, patients with the hyper-IgD syndromes have lifelong recurrent episodes of systemic inflammation and periodic attacks of aphthous ulcers, as well as pharyngitis in some subsets. Recent insights into the role of IgD in upper airway secretions demonstrated that patients with hyper-IgD syndromes have increased numbers of IgD-secreting B cells and increased numbers of “IgD-armed”

basophils, suggesting possible triggers for the periodic inflammatory episodes associated with hyper-IgD syndromes.⁶⁵

B LYMPHOCYTES IN PATIENTS WITH CHRONIC DISEASES OF THE LOWER AIRWAY

Although classically associated with antibody production, B lymphocytes serve additional roles as antigen-presenting cells and sources of both inflammatory and regulatory cytokines,¹⁸⁰ which is perhaps illustrative of the pleiotropic roles of B cells as effectors and regulators of the humoral immune response. B-cell responses and airway-produced antibodies are also associated with pathology in patients with a number of inflammatory diseases of the lower airway, such as asthma, HP, idiopathic fibrosing alveolitis, COPD, sarcoidosis, autoimmune diseases, and lung transplant rejection (Table IV).¹⁸¹⁻²⁰⁶

In mice sensitized by means of intratracheal OVA, ectopic GCs are found within the parenchyma of the inflamed lungs, and OVA-specific immunoglobulin-producing cells can be detected in the pulmonary tissue.⁶⁷ These features are observed along with the eosinophilia and epithelial basement membrane fibrosis classically found in asthma models.²⁰⁷ A recent study examined OVA-sensitized mice after aerosolized antigen challenge and found that pulmonary OVA exposure resulted in increased numbers of specific IgG- and IgE-producing pulmonary plasma cells.¹⁸¹ The plasma cells did not persist after cessation of antigen exposure. In human asthma reports on the prevalence of organized BALT and induction in lung tissue are inconsistent, although isolated clusters of B cells are frequently found in lung biopsy specimens of patients with severe asthma.^{2,208} It is unclear whether the majority of B cells found in the lungs of patients with asthma are sensitized within the secondary lymphatic organs, such as the bronchial LNs, and then traffic to the lungs or whether local activation, expansion, and class-switching occurs. Evidence for local CSR and production of IgE is inferred by the detection of ϵ -circle transcripts, mRNA encoding the heavy chain of IgE, and AID in asthmatic patients compared with healthy control subjects.¹⁸² In contrast to the IgE-mediated responses in patients with asthmatic disease, B-lymphocyte responses resulting from chronic exposure to organic antigens, such as avian antigens in pigeon fancier's disease, can trigger HP.²⁰⁹ In patients with HP, organized BALT containing B cell-predominant follicles surrounded by a parafollicular T-cell zone are frequently found in lung biopsy specimens.^{183,184} Bronchoalveolar lavage fluid from patients with HP also demonstrates increased numbers of plasma cells that are temporally related to antigen exposure.²¹⁰ Interestingly, although both patients with pigeon HP and asymptomatic pigeon breeders demonstrate increased airway levels of anti-avian IgG and IgA antibodies,¹⁸⁵ patients with HP simultaneously express increased levels of class-switched antibodies against self-IgG (or rheumatoid factor). The levels of serum rheumatoid factor are similar to those found in patients with RA¹⁸⁶ and might be locally produced in the lungs because titers were higher in bronchoalveolar lavage fluid than in serum.²¹¹ Together, these findings suggest that pulmonary challenge from inhaled antigen is capable of triggering a robust humoral response within the lung. However, the initiation of a pathogenic inflammatory response appears to depend on intrinsic factors of the antigen and yet unknown host susceptibility or secondary triggers. In addition, the persistence of the response, at least in patients with HP, appears to require the continued presence of antigen.

TABLE IV. Evidence for B-cell infiltrates and morbidity-associated specific antibodies in patients with select airway disease

Disease	Presence of B-cell clusters	Morbidity-associated antibody specificities	References
Lower airways			
Asthma	Inconsistent	Allergen-specific IgE, anti-collagen V	181, 182, 199, 200
COPD	Yes, in adventitia of small airways	Anti-pulmonary epithelial cell Anti-elastin Anti-nuclear antibody	187-190, 201
Chronic transplant rejection	Yes, peribronchiolar	Anti-HLA class I Anti-collagen V	194, 202, 203
HP	Yes, peribronchiolar	Anti-avian (pigeon fancier's disease) Rheumatoid factor	184-186
IPF	Yes, around alveoli	Anti-epithelial Anti-annexin	192, 193, 204
RA	Yes, parenchymal	Locally anti-CCP in BAL fluid	183
Sjogren disease	Yes, parenchymal	Anti-Ro/SSA, anti-La/SSB Anti-M3 muscarinic	183, 191, 205
Upper airways			
Allergic rhinitis	Rare	Allergen-specific IgE	195, 206
Nasal polyposis	Yes, in nasal polyps	Allergen-specific immunoglobulin, <i>Staphylococcus aureus</i> -specific IgE Local autoantibodies against dsDNA, BP180, and other antigens	196-198

Similar to findings in patients with HP, smokers appear to generate a dichotomous response to inhaled tobacco combustion products in which patients with COPD have a vigorous inflammatory response, which is in contrast to nonaffected at-risk smokers.¹⁸⁷ Unlike pigeon HP, in which humoral immune responses to the inhaled antigen are thought to be a dominant pathogenic event, COPD is thought to result from the proteolytic destruction of the lung extracellular matrix by products of neutrophils and macrophages.^{212,213} Although a pathogenic role for B cells in patients with COPD is not yet established, analysis of the inflammatory response demonstrates dramatic increases, particularly in the numbers of B cells and lymphoid follicles found in the adventitia of small airways.²¹⁴ The organization of these follicles has also been proposed to be mediated by the expression of CXCR3 by B and T cells, along with its cognate ligands interferon-inducible protein 10/CXCL10, monokine induced by gamma interferon (Mig)/CXCL9, and interferon-inducible T cell alpha chemoattractant (I-TAC)/CXCL11, which are expressed by epithelial cells, endothelial cells, and CD68⁺ macrophages surrounding these follicles.²¹⁵ Comparing lung biopsy specimens from late Global Initiative for Chronic Obstructive Lung Disease (GOLD) stage (stages 2-4) with those from early GOLD (stage 0-1) subjects, Hogg et al¹⁸⁷ showed 5- to 7-fold higher numbers of B cells and lymphoid follicles that correlated with the airway obstruction and pulmonary parenchymal destruction associated with this disease. Van der Strate et al²¹⁶ characterized the B cells found in follicles in patients with COPD and found that the majority of B cells were post-GC CD27⁺ and further used laser-capture microdissection to demonstrate evidence for an oligoclonal, antigen-specific response similar to their findings in mice chronically exposed to tobacco smoke. A subsequent study examined the bronchial biopsy specimens of 114 patients with COPD and found increased numbers of subepithelial CD20⁺ cells in patients with COPD compared with those of control subjects without COPD. There was also a correlation between more advanced GOLD stage disease and increased subepithelial CD20⁺ cell numbers.¹⁸⁸ In contrast to the study by Hogg et al,¹⁸⁷ this study did not find a follicular organization of these B

lymphocytes, although the caliber of airways examined and the severity of lung disease in these 2 studies were not directly comparable. Similarly, increased numbers of subepithelial and periglandular plasma cells were found in the lung biopsy specimens of smokers with chronic bronchitis compared with those from asymptomatic smokers. Interestingly, 69% of periglandular plasma cells expressed IL-4, and this was correlated with the number of PAS⁺ mucous glands, suggesting that gland-associated plasma cells might play a role in the mucin hypersecretion associated with chronic bronchitis.²¹⁷ Although the previously discussed evidence suggests an excess of B cells and plasma cells surrounding the airway, a recent study found that sIgA levels were actually decreased in the lungs of patients with COPD, particularly those with the most severe GOLD staging. In this study the decrease in sIgA levels correlated with increased airway remodeling and decreased pIgR expression, suggesting that defective sIgA transport might be a factor in the defective mucosal immunity observed in patients with COPD.²¹⁸ Further evidence for a pathogenic role for airway B cells is found in the discovery of increased levels of autoantibodies against elastin¹⁸⁹ and pulmonary epithelial cells¹⁹⁰ found in patients with COPD. ELISpot analysis of single cells isolated from the peripheral lung tissue of patients with COPD confirmed the local production of anti-elastin antibodies within lung tissue, and these findings were associated with a decrease in CD4⁺CD25⁺CD62L⁺ regulatory T-cell numbers in patients with emphysema.¹⁸⁹ These results are intriguing in light of the known pathogenic role for proteolytic enzymes from macrophages and neutrophils in patients with COPD.²¹² Together, these studies suggest that the proteolytic breakdown of the extracellular matrix secondary to cigarette smoke activation might generate self-epitopes for a pathogenic autoimmune humoral response. It is also likely in patients with COPD that repeated bacterial colonization of the lower airways drives a profound antibody response and formation of tertiary lymphoid tissue in this disease. COPD is not the only chronic airway disease in which this scenario appears to play out (see below).

The autoimmune connective tissue diseases (CTDs) are a heterogeneous group of systemic inflammatory disorders,

including SLE, RA, systemic sclerosis, and Sjögren syndrome. Pulmonary inflammation in patients with CTDs has been well described, although the role of B cells in the pathogenesis of this type of inflammation is still largely unexplored.²¹⁹ RA and Sjögren syndrome are frequently associated with the development of ectopic lymphoid tissue, which is thought to be a site at which high-affinity autoreactive B-cell clones are generated.²⁰⁸ Interstitial lung disease is the most common pulmonary manifestation in patients with RA, and lung biopsy specimens from these patients consistently demonstrated the presence of BALT.¹⁸³ Because BALT is not constitutively found in the normal human airway, Randall²⁰⁸ describes these as inducible forms of BALT (iBALT). Although iBALT exhibited the presence of HEVs and characteristics of B-cell follicles, including CXCL13 and BCL-6 expression (see above), the authors did not mention the presence of specialized lymphoepithelium or M cells in conjunction with iBALT. The presence of iBALT also correlated with anticyclic citrullinated peptide (anti-CCP) antibodies but not rheumatoid factor in bronchoalveolar lavage fluid. Similarly, other studies examining the interstitial lung disease of patients with systemic sclerosis and Sjögren syndrome have demonstrated increased BALT levels.^{220,221} Studies of the follicular structures associated with Sjögren syndrome demonstrate B cell–enriched follicles with increased expression of CXCL13 and CCL21. Immunohistochemistry with biotinylated recombinant Ro and La proteins also demonstrated increased numbers of autoantibodies reactive to these antigens being produced in these follicles.²²¹ Other studies have reported the presence of salivary IgA anti-M₃ muscarinic acetylcholine receptors in patients with Sjögren syndrome with sicca symptoms.¹⁹¹ These salivary antibodies might provide a novel pathway through which secretory glandular dysfunction is propagated in the upper and lower airways.

Idiopathic pulmonary fibrosis (IPF), also called cryptogenic fibrosing alveolitis, is a chronic diffuse interstitial lung disease of unknown cause characterized by enhanced collagen deposition in the lung parenchyma.¹⁹² Although the cause of this disease continues to be unknown, consistently implicated factors include inhaled irritants and toxins from cigarette smoke and viruses. DNA from EBV and herpesviruses is frequently detected in the lungs of patients with IPF, although its role in the pathogenesis, progression, or even exacerbation of this disease remains unclear.²²² Regardless of cause, loosely organized B-cell aggregates interacting with T cells and follicular DCs are one of the more consistent histologic aspects of this disease.^{193,223,224} However, it is still unclear whether these B cells are responsible for the generation of the numerous autoantibodies, both circulating and in bronchoalveolar lavage fluid, which target multiple pulmonary epithelial autoantigens in IPF.¹⁹² That a pathogenic link between these autoantibodies and the fibrosis observed in patients with IPF exists might be inferred by studies showing enhanced TGF- β and tenascin production by epithelial cells stimulated *in vitro* with isolated autoantibodies.²²⁵

Recent studies have implicated an increasing role for alloimmune stimulation in breaking self-tolerance, leading to autoimmunity to self-antigens. Obliterative bronchiolitis (OB), a chronic inflammatory and fibroproliferative condition of the small airways, is a frequent manifestation of chronic rejection after lung transplantation. Sato et al²²⁶ examined lung biopsy specimens of patients who had received a lung transplant and found lymphoid neogenesis in lungs affected by chronic rejection. The lymphoid clusters had T and B cells, along with peripheral lymph node

addressin (PNAd⁺) HEVs. The authors then expanded on these findings in an animal model of OB in which they transplanted tracheal isografts or allografts into lung tissue and demonstrated that both isografts and allografts triggered an intense lymphocytic infiltrate in the host lung 7 days after transplantation. By day 28, however, the lymphocytic aggregates in the lung had largely resolved in those receiving isografts but persisted in those receiving allografts. The grafted tracheal epithelium of isografts remained intact, whereas those of the allografts showed complete loss of epithelium and obliterative airway fibrosis that replicates some of the features of OB. Downstream effects of the humoral immune activation in lung transplantation appear to result in a loss of self-tolerance and the generation of autoantibodies. Indeed, the presence of anti-HLA class I and collagen V autoantibodies seems to correlate with the development of OB.¹⁹⁴

Reviewing the spectrum of B-cell inflammatory responses in patients with pulmonary disease reveals several intriguing similarities. In these diseases pulmonary exposure to an antigen, whether inhaled in patients with asthma and HP, possibly viral in patients with IPF, generated through proteolytic breakdown in patients with COPD, introduced in OB caused by chronic transplant rejection, or a self-antigen as seen in patients with CTDs, results in an exuberant B-cell response. The B cells present in these conditions apparently undergo local activation, with evidence for local class-switching in asthmatic patients or localized oligoclonal antigen-specific activation seen in patients with HP, COPD, or CTDs. These localized responses generate increased systemic levels of antigen-specific antibodies associated with the loss of tolerance to self-antigens and the generation of autoantibodies in the most severe manifestations of these diseases.

Despite these similarities, there remain several key aspects of the variable susceptibility to these pathogenic processes that remain unclear. For example, the responsible factors by which an inhaled antigen triggers asthma instead of HP and the underlying host factors or comorbid illness that cause certain pigeon fanciers and some smokers to have iBALT, even though unaffected breeders and unaffected smokers are exposed to similar antigenic challenge remain yet to be determined.

B LYMPHOCYTES IN PATIENTS WITH CHRONIC DISEASES OF THE UPPER AIRWAY

Although studies of B cell–mediated inflammation of the upper airways are rare compared with studies of the gut and even the lower airway, evidence suggests that B cells play a prominent role in both normal immune homeostasis and inflammatory diseases of the nose and paranasal sinuses. Studies in the 1970s by Brostoff, Gleich, and others demonstrated that IgA- and IgE-expressing B cells are found in nasal airways and that these cells produce IgE and IgA specific to known inhaled antigens.²²⁷⁻²²⁹ Several studies have shown that levels of aeroallergen-specific IgE are much higher in the airways than in the serum when normalized to total IgE or albumin.²²⁸⁻²³⁴ It is not uncommon for patients with allergic rhinitis or chronic rhinosinusitis (CRS) to manifest antigen-specific IgE in nasal secretions or nasal tissue with no apparent specific IgE in the serum for the same antigen, and documented cases exist in which nasal antigen challenge responses are elicited in subjects without skin test sensitivity.^{227,229,230,232} A survey study found that 19% of patients with rhinitis and polyposis had specific IgE in the nose but not the serum.²³⁵ More recent studies that used time-resolved fluorescence

immunosorbent assays to determine the relative proportions of total and specific immunoglobulins in the airways and circulation concluded that the majority of total-body aeroallergen-specific antibodies of the IgE and IgA isotypes are produced in the airways and that systemic sensitization largely reflects spillover of immunoglobulins from the mucosal site of their production into the circulation.²³⁶ However, early studies compared the efficacy of systemically administered live attenuated versus inactivated measles vaccines in generating both systemic and nasal-specific immune responses. They found that both types of vaccine efficiently generated a systemic IgG response and the attenuated virus almost universally generated nasal IgA antibodies; the inactivated vaccine only generated measurable nasal anti-measles antibodies in 3 of 7 cases.²³⁷ Together, these studies suggest the nasal antibody armamentarium does not passively reflect that of the systemic circulation. Possible explanations for superior local responses with local vaccination include SLO-independent localized B-cell class-switching within the nasal mucosa, such as the S δ -S μ switch circles found constitutively in nasal mucosa. Moreover, increased levels of ϵ -germline transcripts, ϵ -circle transcripts, and mRNA for AID are found in the nasal mucosa of symptomatic allergic patients.⁴⁹ Other possible explanations include the preferential use of homing molecules in mucosal B cells, such as CCR7, and lack of $\alpha_4\beta_7$, as discussed earlier. A separate study by Cameron et al²³⁸ showed that antigen exposure of the allergic nasal mucosa resulted in a significant increase in local IL-4 production and increased IgE production, as evidenced by expression of the I ϵ germline promoter site and ϵ -circle transcripts that was not seen in control mucosa from nonallergic subjects. These responses could be blocked by pretreatment with the glucocorticoid fluticasone propionate. Indeed, examination of nasal tissue from patients with allergic rhinitis demonstrates the presence of small B-cell clusters in addition to increased levels of B cells and IgE⁺ plasma cells, suggesting the presence of ectopic GCs where localized activation can occur.^{49,195} Recently, it has been shown that eosinophils in bone marrow are nearly indispensable for the generation of plasma cells in bone marrow, a finding that begs the question of whether highly eosinophilic airways diseases, such as hay fever, asthma, or CRS (see below), sustain a local plasma cell presence through a similar eosinophil-mediated process.²³⁹

Several laboratories, including our own, have had an interest in the study of B lymphocytes in the context of CRS with nasal polyps (CRSwNP). Patients with this disease manifest vigorous chronic inflammatory responses in the nose and paranasal sinuses that result in obstructive polyp formation and olfactory loss. Multiple causes, including triggering by *Staphylococcus* species superantigens, fungal antigens, and, more recently, an epithelial barrier defect, have been proposed as possible inciting mechanisms.^{240,241} Clinically, more than half of the patients with CRSwNP are multiply sensitized to inhaled allergens,^{242,243} and a large proportion (19%) of patients with CRS demonstrate the presence of local IgE against aeroallergens without evidence of circulating IgE against the same antigens.²³⁵ Patients with CRSwNP also have significantly higher rates of asthma,²⁴³ most clearly manifested in the most severe forms of CRSwNP, which include a triad with concurrent asthma and aspirin sensitivity. Published reports have demonstrated the occasional presence of GC-like follicles, high numbers of B cells and plasma cells, increased CXCL12 and CXCL13 levels, and increased IgA levels in nasal polyp tissue.^{2,196,244} An overview of the potential role of

B-lineage cells in patients with CRS is presented in Fig 5. One possible mechanism driving the formation of ectopic lymphoid aggregates is the presence of high levels of BAFF mRNA and protein in nasal polyp tissue compared with control tissue.¹²⁸ Furthermore, the expression of BAFF correlated with the expression of CD20 in nasal polyp tissue. It has also been demonstrated that BAFF expression is highly induced by the TLR3 ligand double-stranded RNA and is synergistically enhanced by TLR3- and IFN- β -dependent signaling in airway epithelial cells.^{129,245} In fact, levels of BAFF expression from airway epithelial cells are similar to those produced by DCs and other myeloid cells,^{245,246} suggesting that BAFF synthesis by epithelial cells might contribute to local accumulation, activation, CSR, and immunoglobulin synthesis by B cells in the airways. After activation and expansion by BAFF, another potential factor that might drive B cells to undergo plasma cell differentiation in sinonasal tissues is IL-6, levels of which are also found to be increased within nasal polyp tissue.^{147,247} Recent studies have demonstrated that the number of plasma cells in nasal polyps actually exceeds the number of B cells by an order of magnitude, and these plasma cells are frequently of the CD38⁺ plasma blast phenotype.²⁴⁸ Another study examined the immunoglobulin isotypes expressed by nasal polyp plasma cells and found the locally present plasma cells expressed predominantly IgA and IgG but also IgE.²⁴⁹ Whether this profound plasma cell response is mechanistically driven by BAFF and IL-6 has not been established, but patients with nasal polyps do demonstrate an exuberant nasal antibody response. Locally increased levels of IgA and IgE antibodies to antigens commonly encountered in the nose, such as *Staphylococcus aureus*, dust mite, and fungal elements,^{197,250} have been reported in nasal polyp tissue. Because BAFF is a potent stimulator of B-cell proliferation and class-switching in B cells²⁵¹ and because mice engineered to overexpress this cytokine manifest intense autoimmunity,²⁵² Tan et al¹⁹⁸ recently evaluated sinonasal tissue from patients with CRSwNP for the presence of autoantibodies. These studies revealed that increased levels of several class-switched autoantibodies, particularly anti-dsDNA, are present within nasal polyp tissue. Echoing the findings in the lower airway discussed earlier, the most highly increased levels of anti-dsDNA autoantibodies were observed in nasal polyps obtained from patients undergoing repeated nasal surgery for recurrent and obstructive nasal polyposis. In these studies the presence of increased specific antibody levels within nasal polyps was not paralleled by increases in levels of the same antibodies in the systemic circulation. These findings suggest that class-switching and antibody production in the nasal airway might be uniquely compartmentalized from systemic immunity. Indeed, studies of nasal biopsy specimens from patients with allergic rhinitis that were exposed to allergen *ex vivo* demonstrated local induction of IL-4 and IL-13, as well as increased levels of sterile transcripts of the IgE and IgG isotype, indicating local CSR to form IgG- and IgE-producing B cells. Together, these studies drive home the importance of localized, extralymphatic humoral responses found in the nasal airway.

CLINICAL IMPLICATIONS

The structural variability, antigen specificity, adaptable affinity, and functional characteristics of immunoglobulins make them superb effectors of immune responses both within tissues and at the body's surfaces, as attested to by the enormous clinical and commercial success of immunoglobulins in therapy. The airways make an almost irresistible surface for the growth of bacteria and

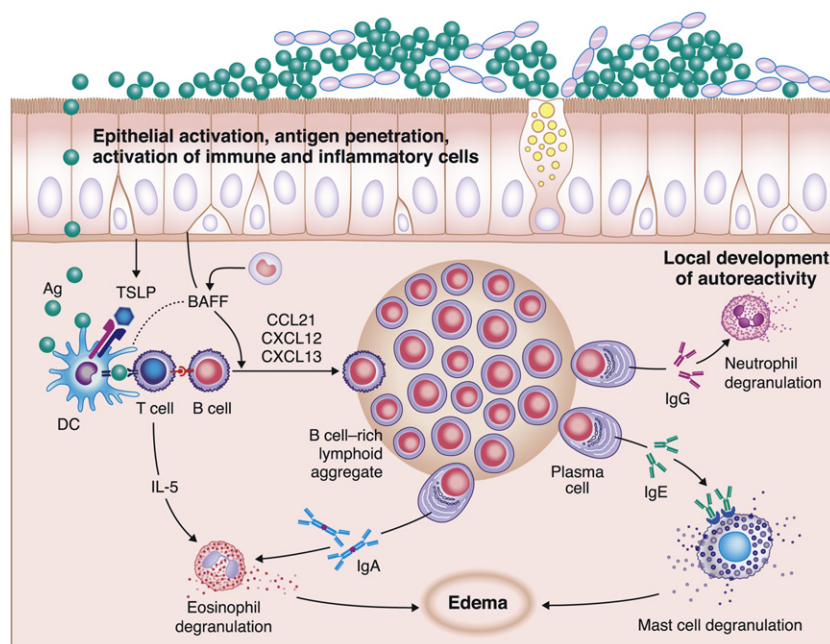


FIG 5. Overview of the role of B cells in patients with CRS. Studies of the pathogenesis of the polypoid form of CRS have implicated B cells and several isotypes of immunoglobulins in the pathogenesis of this important and common disease. Increased B-cell and plasma cell numbers are established in patients with CRS and are accompanied by increased levels of the chemokines that attract them (including CXCL12 and CXCL13), as well as thymic stromal lymphopoietin (TSLP) and BAFF. Formation of organized follicular structures occurs in some cases of CRS. Although the specificity of the IgG, IgA, and IgE antibodies made in nasal polyp tissue is not completely known, recent evidence shows the presence of autoantibodies of the IgA and IgG isotypes. Cellular targets of immunoglobulins that are enriched in nasal polyps include eosinophils (which respond briskly to IgA activation), mast cells (which respond to IgE receptor cross-linking), and neutrophils (which respond to IgG ICs). Release of mediators by these cell types is likely to be important in the pathogenesis of the disease, including formation of nasal polyps.

fungi, containing ample nutrients, an ideal temperature for growth, and a moist substrate. Were it not for the coordinated action of innate and adaptive immune responses, our respiratory system would be defenseless and rapidly overrun by these organisms. B-lineage cells in the airway exhibit a remarkable diversity in function, serving roles ranging from efficient immunoglobulin-producing plasma cells to regulatory B cells. Conversely, they can be agents of disease when a susceptible host is exposed to excessive antigen or when inappropriate sensitization occurs to self or to innocuous material. In such cases B cells and immunoglobulins can exert the same awe-inspiring toxicity toward the host, with devastating results.

The research and principles discussed in this review highlight the need for an improved understanding of the mechanisms of recruitment, activation, differentiation, and regulation of B-lineage cells in health and disease in the airways. Numerous important questions are still left unanswered about these important cells and their involvement in airways diseases. Are there specific molecular pathways through which B-lineage cells are recruited to the sinuses, the upper airways, or the lower airways, as opposed to the gastrointestinal tract or urogenital tract? Do naive B cells regularly traffic through the upper or lower airways as part of their lifecycle? Are plasmablasts or plasma cells activated within the airways and then subsequently recruited to remote locations, such as the bone marrow, to be long-lived antibody producers?

Although there are many unanswered questions, increased awareness of pathologic B-cell responses has increased the

recognition of a need to develop strategies to prevent, diagnose, treat, and manage B cell-mediated chronic inflammatory airway diseases. Consequently, numerous drugs designed to target elements of the B-cell lineage are presently available or under development (Table III).

As in most areas of medicine, the role of genetic or epigenetic variation in B-cell lineage pathways in disease susceptibility in the airways is unknown and likely to be important. In this framework subjects at high risk for airway disease, such as pigeon fanciers at risk for HP, might be found to have certain susceptibility genetic polymorphisms or epigenetic marks. In our own studies African American subjects with the minor A allele of the rs17564816 BAFF single nucleotide polymorphism were implicated to be at increased risk for asthma exacerbations.²⁵³ It seems likely that pharmaceutical agents targeted at suppressing B-cell activation will be found to be influenced by genetic and epigenetic variability. The success of rituximab in treating refractory RA demonstrates the potential for B cell-specific therapies in treating inflammatory conditions affecting the airways, but its rare potential for progressive multifocal leukoencephalopathy from the JC virus (fatal in 90% of affected subjects) is cautionary, particularly for airway diseases or disease stages in which attributable mortality is low.²⁵⁴

It is self-evident that any therapeutic strategies designed to prevent pathological B-cell responses should do so without undermining their essential roles in immunity. In this context, and given the critical role of B cells in tolerance and immunologic memory, long-term management of chronic inflammatory airway diseases by mechanisms inducing immune tolerance to specific

extrinsic antigens is a worthy goal, along with direct pharmacologic suppression of autoimmune responses.

We hope that this review has illustrated the importance of the B-lineage cell system in both host defense and disease within the respiratory system and that it will serve as a catalyst for the further study of B cells and plasma cells in the human respiratory system.

We extend our gratitude to Jacqi Schaffer for her input and guidance in preparation of the figures in this review.

What we do know?

- Airway B cells are capable of activation and differentiation independent of SLOs.
- The presence of B cells in airway inflammatory infiltrates correlates with disease severity in patients with many chronic airways diseases.
- B cells in the airways produce antibodies against extrinsic antigens, as well as autoantigens.
- The presence of autoantibodies in patients with several airways diseases correlates with poorer prognosis or more severe disease.

What is still unknown?

- The existence of airway-specific B cell-trafficking mechanisms
- The mechanisms driving CSR of airway IgD⁺IgM⁺ B cells
- The mechanisms of appearance of IgD in the airways
- The role that migration and local activation of naive B cells plays in airways disease
- The mechanisms that drive IgG subclass CSR in human subjects

REFERENCES

- Salvi S, Holgate ST. Could the airway epithelium play an important role in mucosal immunoglobulin A production? *Clin Exp Allergy* 1999;29:1597-605.
- Drolet JP, Frangie H, Guay J, Hajoui O, Hamid Q, Mazer BD. B lymphocytes in inflammatory airway diseases. *Clin Exp Allergy* 2010;40:841-9.
- Wilkes DS, Twigg HL 3rd. B-lymphocytes in the lung: a topic to be revisited. *Sarcoidosis Vasc Diffuse Lung Dis* 2001;18:34-49.
- Brandtzaeg P. Potential of nasopharynx-associated lymphoid tissue for vaccine responses in the airways. *Am J Respir Crit Care Med* 2011;183:1595-604.
- Kurosaki T, Shinohara H, Baba Y. B cell signaling and fate decision. *Annu Rev Immunol* 2010;28:21-55.
- Pillai S, Cariappa A. The follicular versus marginal zone B lymphocyte cell fate decision. *Nat Rev Immunol* 2009;9:767-77.
- Weill JC, Weller S, Reynaud CA. Human marginal zone B cells. *Annu Rev Immunol* 2009;27:267-85.
- Montecino-Rodriguez E, Leathers H, Dorshkind K. Identification of a B-1 B cell-specified progenitor. *Nat Immunol* 2006;7:293-301.
- Baumgarth N. The double life of a B-1 cell: self-reactivity selects for protective effector functions. *Nat Rev Immunol* 2011;11:34-46.
- Ansel KM, Harris RB, Cyster JG. CXCL13 is required for B1 cell homing, natural antibody production, and body cavity immunity. *Immunity* 2002;16:67-76.
- Berland R, Wortis HH. Origins and functions of B-1 cells with notes on the role of CD5. *Annu Rev Immunol* 2002;20:253-300.
- Dorner T, Kinnman N, Tak PP. Targeting B cells in immune-mediated inflammatory disease: a comprehensive review of mechanisms of action and identification of biomarkers. *Pharmacol Ther* 2010;125:464-75.
- Choi YS, Baumgarth N. Dual role for B-1a cells in immunity to influenza virus infection. *J Exp Med* 2008;205:3053-64.
- Griffin DO, Rothstein TL. Human b1 cell frequency: isolation and analysis of human b1 cells. *Front Immunol* 2012;3:122.
- Griffin DO, Holodick NE, Rothstein TL. Human B1 cells in umbilical cord and adult peripheral blood express the novel phenotype CD20+ CD27+ CD43+ CD70. *J Exp Med* 2011;208:67-80.
- Griffin DO, Rothstein TL. A small CD11b(+) human B1 cell subpopulation stimulates T cells and is expanded in lupus. *J Exp Med* 2011;208:2591-8.
- Griffin DO, Rothstein TL. Human "orchestrator" CD11b(+) B1 cells spontaneously secrete interleukin-10 and regulate T-cell activity. *Mol Med* 2012;18:1003-8.
- Inman CF, Murray TZ, Bailey M, Cose S. Most B cells in non-lymphoid tissues are naive. *Immunol Cell Biol* 2012;90:235-42.
- Carragher DM, Rangel-Moreno J, Randall TD. Ectopic lymphoid tissues and local immunity. *Semin Immunol* 2008;20:26-42.
- Reif K, Ekland EH, Ohl L, Nakano H, Lipp M, Forster R, et al. Balanced responsiveness to chemoattractants from adjacent zones determines B-cell position. *Nature* 2002;416:94-9.
- Cyster JG. B cell follicles and antigen encounters of the third kind. *Nat Immunol* 2010;11:989-96.
- Okada T, Miller MJ, Parker I, Krummel MF, Neighbors M, Hartley SB, et al. Antigen-engaged B cells undergo chemotaxis toward the T zone and form motile conjugates with helper T cells. *PLoS Biol* 2005;3:e150.
- Goodnow CC, Vinuesa CG, Randall KL, Mackay F, Brink R. Control systems and decision making for antibody production. *Nat Immunol* 2010;11:681-8.
- Bergtold A, Desai DD, Gavhane A, Clynes R. Cell surface recycling of internalized antigen permits dendritic cell priming of B cells. *Immunity* 2005;23:503-14.
- Mackay F, Schneider P. Cracking the BAFF code. *Nat Rev Immunol* 2009;9:491-502.
- Chan TD, Gatto D, Wood K, Camidge T, Basten A, Brink R. Antigen affinity controls rapid T-dependent antibody production by driving the expansion rather than the differentiation or extrafollicular migration of early plasmablasts. *J Immunol* 2009;183:3139-49.
- Inamine A, Takahashi Y, Baba N, Miyake K, Tokuhisa T, Takemori T, et al. Two waves of memory B-cell generation in the primary immune response. *Int Immunol* 2005;17:581-9.
- Paus D, Phan TG, Chan TD, Gardam S, Basten A, Brink R. Antigen recognition strength regulates the choice between extrafollicular plasma cell and germinal center B cell differentiation. *J Exp Med* 2006;203:1081-91.
- Chan TD, Gardam S, Gatto D, Turner VM, Silke J, Brink R. In vivo control of B-cell survival and antigen-specific B-cell responses. *Immunol Rev* 2010;237:90-103.
- Gatto D, Wood K, Brink R. EB12 operates independently of but in cooperation with CXCR5 and CCR7 to direct B cell migration and organization in follicles and the germinal center. *J Immunol* 2011;187:4621-8.
- Pereira JP, Kelly LM, Cyster JG. Finding the right niche: B-cell migration in the early phases of T-dependent antibody responses. *Int Immunol* 2010;22:413-9.
- Hannedouche S, Zhang J, Yi T, Shen W, Nguyen D, Pereira JP, et al. Oxysterols direct immune cell migration via EB12. *Nature* 2011;475:524-7.
- Liu C, Yang XV, Wu J, Kuei C, Mani NS, Zhang L, et al. Oxysterols direct B-cell migration through EB12. *Nature* 2011;475:519-23.
- Shlomchik MJ, Weisel F. Germinal center selection and the development of memory B and plasma cells. *Immunol Rev* 2012;247:52-63.
- McHeyzer-Williams LJ, McHeyzer-Williams MG. Antigen-specific memory B cell development. *Annu Rev Immunol* 2005;23:487-513.
- Gatto D, Brink R. The germinal center reaction. *J Allergy Clin Immunol* 2010;126:898-909.
- Vinuesa CG, Linterman MA, Goodnow CC, Randall KL. T cells and follicular dendritic cells in germinal center B-cell formation and selection. *Immunol Rev* 2010;237:72-89.
- Beltman JB, Allen CD, Cyster JG, de Boer RJ. B cells within germinal centers migrate preferentially from dark to light zone. *Proc Natl Acad Sci U S A* 2011;108:8755-60.
- Barnett BE, Ciocca ML, Goenka R, Barnett LG, Wu J, Laufer TM, et al. Asymmetric B cell division in the germinal center reaction. *Science* 2012;335:342-4.
- Crotty S. Follicular helper CD4 T cells (TFH). *Annu Rev Immunol* 2011;29:621-63.
- Allen CD, Okada T, Cyster JG. Germinal-center organization and cellular dynamics. *Immunity* 2007;27:190-202.
- Tsuji M, Komatsu N, Kawamoto S, Suzuki K, Kanagawa O, Honjo T, et al. Preferential generation of follicular B helper T cells from Foxp3+ T cells in gut Peyer's patches. *Science* 2009;323:1488-92.

43. Moyron-Quiroz JE, Rangel-Moreno J, Kusser K, Hartson L, Sprague F, Goodrich S, et al. Role of inducible bronchus associated lymphoid tissue (iBALT) in respiratory immunity. *Nat Med* 2004;10:927-34.
44. Lund FE, Partida-Sanchez S, Lee BO, Kusser KL, Hartson L, Hogan RJ, et al. Lymphotoxin-alpha-deficient mice make delayed, but effective, T and B cell responses to influenza. *J Immunol* 2002;169:5236-43.
45. Lee BJ, Santee S, Von Gesjen S, Ware CF, Sarawar SR. Lymphotoxin-alpha-deficient mice can clear a productive infection with murine gammaherpesvirus 68 but fail to develop splenomegaly or lymphocytosis. *J Virol* 2000;74:2786-92.
46. Aloisi F, Pujol-Borrell R. Lymphoid neogenesis in chronic inflammatory diseases. *Nat Rev Immunol* 2006;6:205-17.
47. McDonald KG, McDonough JS, Newberry RD. Adaptive immune responses are dispensable for isolated lymphoid follicle formation: antigen-naïve, lymphotoxin-sufficient B lymphocytes drive the formation of mature isolated lymphoid follicles. *J Immunol* 2005;174:5720-8.
48. Cerutti A, Chen K, Chorny A. Immunoglobulin responses at the mucosal interface. *Annu Rev Immunol* 2011;29:273-93.
49. Cameron L, Hamid Q, Wright E, Nakamura Y, Christodoulouopoulos P, Muro S, et al. Local synthesis of epsilon germline gene transcripts, IL-4, and IL-13 in allergic nasal mucosa after ex vivo allergen exposure. *J Allergy Clin Immunol* 2000;106:46-52.
50. Gould HJ, Sutton BJ. IgE in allergy and asthma today. *Nat Rev Immunol* 2008;8:205-17.
51. Tezuka H, Abe Y, Iwata M, Takeuchi H, Ishikawa H, Matsushita M, et al. Regulation of IgA production by naturally occurring TNF/iNOS-producing dendritic cells. *Nature* 2007;448:929-33.
52. Brandtzaeg P. Mucosal immunity: induction, dissemination, and effector functions. *Scand J Immunol* 2009;70:505-15.
53. Cortes B. Role of secretory immunoglobulin A and secretory component in the protection of mucosal surfaces. *Future Microbiol* 2010;5:817-29.
54. Kaetzel CS, Robinson JK, Chintalacharuvu KR, Vaerman JP, Lamm ME. The polymeric immunoglobulin receptor (secretory component) mediates transport of immune complexes across epithelial cells: a local defense function for IgA. *Proc Natl Acad Sci U S A* 1991;88:8796-800.
55. Tomasi TB Jr, Tan EM, Solomon A, Prendergast RA. Characteristics of an immune system common to certain external secretions. *J Exp Med* 1965;121:101-24.
56. Johansen FE, Baekkevold ES, Carlsen HS, Farstad IN, Soler D, Brandtzaeg P. Regional induction of adhesion molecules and chemokine receptors explains disparate homing of human B cells to systemic and mucosal effector sites: dispersion from tonsils. *Blood* 2005;106:593-600.
57. Sun K, Johansen FE, Eckmann L, Metzger DW. An important role for polymeric Ig receptor-mediated transport of IgA in protection against *Streptococcus pneumoniae* nasopharyngeal carriage. *J Immunol* 2004;173:4576-81.
58. Tjarnlund A, Rodriguez A, Cardona PJ, Guirado E, Ivanyi J, Singh M, et al. Polymeric IgR knockout mice are more susceptible to mycobacterial infections in the respiratory tract than wild-type mice. *Int Immunol* 2006;18:807-16.
59. Crago SS, Prince SJ, Kulhavy R, Mestecky J. Molecular-cellular interactions in the secretory IgA system. *Adv Exp Med Biol* 1978;107:209-17.
60. Czerkinsky C, Prince SJ, Michalek SM, Jackson S, Russell MW, Moldoveanu Z, et al. IgA antibody-producing cells in peripheral blood after antigen ingestion: evidence for a common mucosal immune system in humans. *Proc Natl Acad Sci U S A* 1987;84:2449-53.
61. Brandtzaeg P, Kiyono H, Pabst R, Russell MW. Terminology: nomenclature of mucosa-associated lymphoid tissue. *Mucosal Immunol* 2008;1:31-7.
62. Kiyono H, Fukuyama S. NALT- versus Peyer's-patch-mediated mucosal immunity. *Nat Rev Immunol* 2004;4:699-710.
63. Quiding-Jarbrink M, Granstrom G, Nordstrom I, Holmgren J, Czerkinsky C. Induction of compartmentalized B-cell responses in human tonsils. *Infect Immun* 1995;63:853-7.
64. Ogra PL. Effect of tonsillectomy and adenoidectomy on nasopharyngeal antibody response to poliovirus. *N Engl J Med* 1971;284:59-64.
65. Chen K, Xu W, Wilson M, He B, Miller NW, Bengten E, et al. Immunoglobulin D enhances immune surveillance by activating antimicrobial, proinflammatory and B cell-stimulating programs in basophils. *Nat Immunol* 2009;10:889-98.
66. Westermann J, Pabst R. How organ-specific is the migration of 'naïve' and 'memory' T cells? *Immunol Today* 1996;17:278-82.
67. Chvatchko Y, Kosco-Vilbois MH, Herren S, Lefort J, Bonnefoy JY. Germinal center formation and local immunoglobulin E (IgE) production in the lung after an airway antigenic challenge. *J Exp Med* 1996;184:2353-60.
68. Moyron-Quiroz JE, Rangel-Moreno J, Hartson L, Kusser K, Tighe MP, Klonowski KD, et al. Persistence and responsiveness of immunologic memory in the absence of secondary lymphoid organs. *Immunity* 2006;25:643-54.
69. GeurtsvanKessel CH, Willart MA, Bergen IM, van Rijt LS, Muskens F, Elewaut D, et al. Dendritic cells are crucial for maintenance of tertiary lymphoid structures in the lung of influenza virus-infected mice. *J Exp Med* 2009;206:2339-49.
70. Rangel-Moreno J, Moyron-Quiroz JE, Hartson L, Kusser K, Randall TD. Pulmonary expression of CXC chemokine ligand 13, CC chemokine ligand 19, and CC chemokine ligand 21 is essential for local immunity to influenza. *Proc Natl Acad Sci U S A* 2007;104:10577-82.
71. Cyster JG. Homing of antibody secreting cells. *Immunol Rev* 2003;194:48-60.
72. Goya S, Matsuoka H, Mori M, Morishita H, Kida H, Kobashi Y, et al. Sustained interleukin-6 signalling leads to the development of lymphoid organ-like structures in the lung. *J Pathol* 2003;200:82-7.
73. Vuilleminot BR, Rodriguez JF, Hoyle GW. Lymphoid tissue and emphysema in the lungs of transgenic mice inducibly expressing tumor necrosis factor-alpha. *Am J Respir Cell Mol Biol* 2004;30:438-48.
74. Lee JJ, McGarry MP, Farmer SC, Denzler KL, Larson KA, Carrigan PE, et al. Interleukin-5 expression in the lung epithelium of transgenic mice leads to pulmonary changes pathognomonic of asthma. *J Exp Med* 1997;185:2143-56.
75. Smith KG, Clatworthy MR. FcgammaRIIB in autoimmunity and infection: evolutionary and therapeutic implications. *Nat Rev Immunol* 2010;10:328-43.
76. Fairfax KA, Kallies A, Nutt SL, Tarlinton DM. Plasma cell development: from B-cell subsets to long-term survival niches. *Semin Immunol* 2008;20:49-58.
77. Oracki SA, Walker JA, Hibbs ML, Corcoran LM, Tarlinton DM. Plasma cell development and survival. *Immunol Rev* 2010;237:140-59.
78. Hirokawa S, Sato H, Kato I, Kudo A. EBF-regulating Pax5 transcription is enhanced by STAT5 in the early stage of B cells. *Eur J Immunol* 2003;33:1824-9.
79. Kwon H, Thierry-Mieg D, Thierry-Mieg J, Kim HP, Oh J, Tunyaplin C, et al. Analysis of interleukin-21-induced Prdm1 gene regulation reveals functional cooperation of STAT3 and IRF4 transcription factors. *Immunity* 2009;31:941-52.
80. Kato A, Schleimer RP. Beyond inflammation: airway epithelial cells are at the interface of innate and adaptive immunity. *Curr Opin Immunol* 2007;19:711-20.
81. Lambrecht BN, Hammad H. Biology of lung dendritic cells at the origin of asthma. *Immunity* 2009;31:412-24.
82. Mackay F, Figgett WA, Saulep D, Lepage M, Hibbs ML. B-cell stage and context-dependent requirements for survival signals from BAFF and the B-cell receptor. *Immunol Rev* 2010;237:205-25.
83. Kraus M, Alimzhanov MB, Rajewsky N, Rajewsky K. Survival of resting mature B lymphocytes depends on BCR signaling via the Igalpha/beta heterodimer. *Cell* 2004;117:787-800.
84. Cancro MP. Signalling crosstalk in B cells: managing worth and need. *Nat Rev Immunol* 2009;9:657-61.
85. Lazarus AH, Kawauchi K, Rapoport MJ, Delovitch TL. Antigen-induced B lymphocyte activation involves the p21ras and ras.GAP signaling pathway. *J Exp Med* 1993;178:1765-9.
86. King LB, Freedman BD. B-lymphocyte calcium influx. *Immunol Rev* 2009;231:265-77.
87. Rickert RC. Regulation of B lymphocyte activation by complement C3 and the B cell coreceptor complex. *Curr Opin Immunol* 2005;17:237-43.
88. Harwood NE, Batista FD. Early events in B cell activation. *Annu Rev Immunol* 2010;28:185-210.
89. Kurosaki T. Regulation of B-cell signal transduction by adaptor proteins. *Nat Rev Immunol* 2002;2:354-63.
90. Yusuf I, Zhu X, Kharas MG, Chen J, Fruman DA. Optimal B-cell proliferation requires phosphoinositide 3-kinase-dependent inactivation of FOXO transcription factors. *Blood* 2004;104:784-7.
91. Conley ME, Dobbs AK, Farmer DM, Kilic S, Paris K, Grigoriadou S, et al. Primary B cell immunodeficiencies: comparisons and contrasts. *Annu Rev Immunol* 2009;27:199-227.
92. van Zelm MC, Smet J, Adams B, Mascart F, Schandene L, Janssen F, et al. CD81 gene defect in humans disrupts CD19 complex formation and leads to antibody deficiency. *J Clin Invest* 2010;120:1265-74.
93. Eibel H, Salzer U, Warnatz K. Common variable immunodeficiency at the end of a prospering decade: towards novel gene defects and beyond. *Curr Opin Allergy Clin Immunol* 2010;10:526-33.
94. Alinari L, Christian B, Baiocchi RA. Novel targeted therapies for mantle cell lymphoma. *Oncotarget* 2012;3:203-11.
95. Buch MH, Emery P. New therapies in the management of rheumatoid arthritis. *Curr Opin Rheumatol* 2011;23:245-51.
96. Flaherty KT, Infante JR, Daud A, Gonzalez R, Keefe RF, Sosman J, et al. Combined BRAF and MEK inhibition in melanoma with BRAF V600 mutations. *N Engl J Med* 2012;367:1694-703.
97. Kantarjian H, le Coutre P, Cortes J, Pinilla-Ibarz J, Nagler A, Hochhaus A, et al. Phase 1 study of INNO-406, a dual Abl/Lyn kinase inhibitor, in Philadelphia chromosome-positive leukemias after imatinib resistance or intolerance. *Cancer* 2010;116:2665-72.

98. Lee L, Niu H, Goelzer P, Rueger R, Deutsch J, Busse-Reid R, et al. The safety, tolerability, pharmacokinetics, and pharmacodynamics of single oral doses of RO5068760, an MEK inhibitor, in healthy volunteers: assessment of target suppression. *J Clin Pharmacol* 2010;50:1397-405.
99. Leijen S, Middleton MR, Tresca P, Kraeber-Bodere F, Dieras V, Scheulen ME, et al. Phase I dose-escalation study of the safety, pharmacokinetics, and pharmacodynamics of the MEK inhibitor RO4987655 (CH4987655) in patients with advanced solid tumors. *Clin Cancer Res* 2012;18:4794-805.
100. Liu L, Halladay JS, Shin Y, Wong S, Coraggio M, La H, et al. Significant species difference in amide hydrolysis of GDC-0834, a novel potent and selective Bruton's tyrosine kinase inhibitor. *Drug Metab Dispos* 2011;39:1840-9.
101. Pal SK, Reckamp K, Yu H, Figlin RA. Akt inhibitors in clinical development for the treatment of cancer. *Expert Opin Investig Drugs* 2010;19:1355-66.
102. Patel SP, Lazar AJ, Papadopoulos NE, Liu P, Infante JR, Glass MR, et al. Clinical responses to selumetinib (AZD6244; ARRY-142886)-based combination therapy stratified by gene mutations in patients with metastatic melanoma. *Cancer* 2013; 119:799-805.
103. Singh D, Smyth L, Borrell Z, Sweeney L, Tal-Singer R. A randomized, placebo-controlled study of the effects of the p38 MAPK inhibitor SB-681323 on blood biomarkers of inflammation in COPD patients. *J Clin Pharmacol* 2010;50:94-100.
104. Vansteenkiste J, Ramlau R, von Pawel J, San Antonio B, Eschbach C, Szczesna A, et al. A phase II randomized study of cisplatin-pemetrexed plus either enzalutaurin or placebo in chemonaive patients with advanced non-small cell lung cancer. *Oncology* 2012;82:25-9.
105. Chen D, Frezza M, Schmitt S, Kanwar J, Dou QP. Bortezomib as the first proteasome inhibitor anticancer drug: current status and future perspectives. *Curr Cancer Drug Targets* 2011;11:239-53.
106. Navarra SV, Guzman RM, Gallacher AE, Hall S, Levy RA, Jimenez RE, et al. Efficacy and safety of belimumab in patients with active systemic lupus erythematosus: a randomised, placebo-controlled, phase 3 trial. *Lancet* 2011;377:721-31.
107. Stohl W. Biologic differences between various inhibitors of the BLYS/BAFF pathway: should we expect differences between belimumab and other inhibitors in development? *Curr Rheumatol Rep* 2012;14:303-9.
108. Kopp MV. Omalizumab: anti-IgE therapy in allergy. *Curr Allergy Asthma Rep* 2011;11:101-6.
109. Webb S. Attacks on asthma. *Nat Biotechnol* 2011;29:860-3.
110. Levine SJ, Wenzel SE. Narrative review: the role of Th2 immune pathway modulation in the treatment of severe asthma and its phenotypes. *Ann Intern Med* 2010;152:232-7.
111. Corren J, Lemanske RF, Hanania NA, Korenblat PE, Parsey MV, Arron JR, et al. Lebrikizumab treatment in adults with asthma. *N Engl J Med* 2011;365:1088-98.
112. Barnes PJ. The cytokine network in asthma and chronic obstructive pulmonary disease. *J Clin Invest* 2008;118:3546-56.
113. Fizazi K, De Bono JS, Flechon A, Heidenreich A, Voog E, Davis NB, et al. Randomised phase II study of siltuximab (CANTO 328), an anti-IL-6 monoclonal antibody, in combination with mitoxantrone/prednisone versus mitoxantrone/prednisone alone in metastatic castration-resistant prostate cancer. *Eur J Cancer* 2012;48:85-93.
114. Maurer MF, Garrigues U, Jaspers SR, Meengs B, Rixon MW, Stevens BL, et al. Generation and characterization of human anti-human IL-21 neutralizing monoclonal antibodies. *MAbs* 2012;4:69-83.
115. Dörner T, Radbruch A, Burmester GR. B-cell-directed therapies for autoimmune disease. *Nat Rev Rheumatol* 2009;5:433-41.
116. Elgueta R, Benson MJ, de Vries VC, Wasiuk A, Guo Y, Noelle RJ. Molecular mechanism and function of CD40/CD40L engagement in the immune system. *Immunol Rev* 2009;229:152-72.
117. Grammer AC, Lipsky PE. CD154-CD40 interactions mediate differentiation to plasma cells in healthy individuals and persons with systemic lupus erythematosus. *Arthritis Rheum* 2002;46:1417-29.
118. Muramatsu M, Kinoshita K, Fagarasan S, Yamada S, Shinkai Y, Honjo T. Class switch recombination and hypermutation require activation-induced cytidine deaminase (AID), a potential RNA editing enzyme. *Cell* 2000;102:553-63.
119. Vale AM, Schroeder HW Jr. Clinical consequences of defects in B-cell development. *J Allergy Clin Immunol* 2010;125:778-87.
120. Chatzigeorgiou A, Lyberi M, Chatzilymperis G, Nezos A, Kamper E. CD40/CD40L signaling and its implication in health and disease. *Biofactors* 2009;35: 474-83.
121. Chalermkulrat W, McKinnon KP, Brickey WJ, Neuringer IP, Park RC, Sterka DG, et al. Combined donor specific transfusion and anti-CD154 therapy achieves airway allograft tolerance. *Thorax* 2006;61:61-7.
122. Khan WN. B cell receptor and BAFF receptor signaling regulation of B cell homeostasis. *J Immunol* 2009;183:3561-7.
123. Warnatz K, Salzer U, Rizzi M, Fischer B, Gutenberger S, Böhm J, et al. B-cell activating factor receptor deficiency is associated with an adult-onset antibody deficiency syndrome in humans. *Proc Natl Acad Sci U S A* 2009; 106:13945-50.
124. Mackay F, Schneider P. TACI, an enigmatic BAFF/APRIL receptor, with new unappreciated biochemical and biological properties. *Cytokine Growth Factor Rev* 2008;19:263-76.
125. He B, Santamaria R, Xu W, Cols M, Chen K, Puga I, et al. The transmembrane activator TACI triggers immunoglobulin class switching by activating B cells through the adaptor MyD88. *Nat Immunol* 2010;11:836-45.
126. Yong PF, Thaventhiran JE, Grimbacher B. "A rose is a rose is a rose," but CVID is Not CVID common variable immune deficiency (CVID), what do we know in 2011? *Adv Immunol* 2011;111:47-107.
127. Rickert RC, Jellusova J, Miletic AV. Signaling by the tumor necrosis factor receptor superfamily in B-cell biology and disease. *Immunol Rev* 2011;244: 115-33.
128. Kato A, Peters A, Suh L, Carter R, Harris KE, Chandra R, et al. Evidence of a role for B cell-activating factor of the TNF family in the pathogenesis of chronic rhinosinusitis with nasal polyps. *J Allergy Clin Immunol* 2008;121: 1385-92, e1-2.
129. Kato A, Xiao H, Chustz RT, Liu MC, Schleimer RP. Local release of B cell-activating factor of the TNF family after segmental allergen challenge of allergic subjects. *J Allergy Clin Immunol* 2009;123:369-75.
130. Polverino F, Baraldo S, Bazzan E, Agostini S, Turato G, Lunardi F, et al. A novel insight into adaptive immunity in chronic obstructive pulmonary disease: B cell activating factor belonging to the tumor necrosis factor family. *Am J Respir Crit Care Med* 2010;182:1011-9.
131. Bilsborough J, Chadwick E, Mudri S, Ye X, Henderson WR Jr, Waggie K, et al. TACI-Ig prevents the development of airway hyperresponsiveness in a murine model of asthma. *Clin Exp Allergy* 2008;38:1959-68.
132. Kawai T, Akira S. The role of pattern-recognition receptors in innate immunity: update on Toll-like receptors. *Nat Immunol* 2010;11:373-84.
133. Chiron D, Bekeredjian-Ding I, Pellat-Deceunynck C, Bataille R, Jégou G. Toll-like receptors: lessons to learn from normal and malignant human B cells. *Blood* 2008;112:2205-13.
134. Dörner M, Brandt S, Tinguely M, Zucol F, Bourquin JP, Zauner L, et al. Plasma cell toll-like receptor (TLR) expression differs from that of B cells, and plasma cell TLR triggering enhances immunoglobulin production. *Immunology* 2009; 128:573-9.
135. Kelly-Scumpia KM, Scumpia PO, Weinstein JS, Delano MJ, Cuenca AG, Nacionales DC, et al. B cells enhance early innate immune responses during bacterial sepsis. *J Exp Med* 2011;208:1673-82.
136. Baccala R, Hoebe K, Kono DH, Beutler B, Theofilopoulos AN. TLR-dependent and TLR-independent pathways of type I interferon induction in systemic autoimmunity. *Nat Med* 2007;13:543-51.
137. Leadbetter EA, Rifkin IR, Hohlbaum AM, Beaudette BC, Shlomchik MJ, Marshak-Rothstein A. Chromatin-IgG complexes activate B cells by dual engagement of IgM and Toll-like receptors. *Nature* 2002;416:603-7.
138. Lau CM, Broughton C, Tabor AS, Akira S, Flavell RA, Mamula MJ, et al. RNA-associated autoantigens activate B cells by combined B cell antigen receptor/Toll-like receptor 7 engagement. *J Exp Med* 2005;202:1171-7.
139. Marshak-Rothstein A, Rifkin IR. Immunologically active autoantigens: the role of toll-like receptors in the development of chronic inflammatory disease. *Annu Rev Immunol* 2007;25:419-41.
140. Swigris JJ, Fischer A, Gillis J, Meehan RT, Brown KK. Pulmonary and thrombotic manifestations of systemic lupus erythematosus. *Chest* 2008;133:271-80.
141. Gao H, Neff T, Ward PA. Regulation of lung inflammation in the model of IgG immune-complex injury. *Annu Rev Pathol* 2006;1:215-42.
142. Stavnezer J, Guikema JE, Schrader CE. Mechanism and regulation of class switch recombination. *Annu Rev Immunol* 2008;26:261-92.
143. Fawaz LM, Sharif-Askari E, Hajoui O, Soussi-Gounni A, Hamid Q, Mazer BD. Expression of IL-9 receptor alpha chain on human germinal center B cells modulates IgE secretion. *J Allergy Clin Immunol* 2007;120:1208-15.
144. Avery DT, Bryant VL, Ma CS, de Waal Malefyt R, Tangye SG. IL-21-induced isotype switching to IgG and IgA by human naive B cells is differentially regulated by IL-4. *J Immunol* 2008;181:1767-79.
145. Avery DT, Ma CS, Bryant VL, Santner-Nanan B, Nanan R, Wong M, et al. STAT3 is required for IL-21-induced secretion of IgE from human naive B cells. *Blood* 2008;112:1784-93.
146. Kishimoto T. Interleukin-6: from basic science to medicine—40 years in immunology. *Annu Rev Immunol* 2005;23:1-21.
147. Peters AT, Kato A, Zhang N, Conley DB, Suh L, Tancowny B, et al. Evidence for altered activity of the IL-6 pathway in chronic rhinosinusitis with nasal polyps. *J Allergy Clin Immunol* 2010;125:397-403, e10.
148. Malin S, McManus S, Busslinger M. STAT5 in B cell development and leukemia. *Curr Opin Immunol* 2010;22:168-76.

149. Konforte D, Simard N, Paige CJ. IL-21: an executor of B cell fate. *J Immunol* 2009;182:1781-7.
150. Cupedo T, Crellin NK, Papazian N, Rombouts EJ, Weijer K, Grogan JL, et al. Human fetal lymphoid tissue-inducer cells are interleukin 17-producing precursors to RORC+ CD127+ natural killer-like cells. *Nat Immunol* 2009;10:66-74.
151. Mitsdoerffer M, Lee Y, Jager A, Kim HJ, Korn T, Kolls JK, et al. Proinflammatory T helper type 17 cells are effective B-cell helpers. *Proc Natl Acad Sci U S A* 2010;107:14292-7.
152. Maarof G, Bouchet-Delbos L, Gary-Gouy H, Durand-Gasselin I, Krzysiek R, Daloul A. Interleukin-24 inhibits the plasma cell differentiation program in human germinal center B cells. *Blood* 2010;115:1718-26.
153. Liu Z, Zou Y, Davidson A. Plasma cells in systemic lupus erythematosus: the long and short of it all. *Eur J Immunol* 2011;41:588-91.
154. Amoroso A, Hafsi S, Militello L, Russo AE, Soua Z, Mazzarino MC, et al. Understanding rituximab function and resistance: implications for tailored therapy. *Front Biosci* 2011;16:770-82.
155. Donvik KK, Omdal R. Churg-Strauss syndrome successfully treated with rituximab. *Rheumatol Int* 2011;31:89-91.
156. Debertin AS, Tschernig T, Tonjes H, Kleemann WJ, Troger HD, Pabst R. Nasal-associated lymphoid tissue (NALT): frequency and localization in young children. *Clin Exp Immunol* 2003;134:503-7.
157. Bienenstock J, McDermott MR. Bronchus- and nasal-associated lymphoid tissues. *Immunol Rev* 2005;206:22-31.
158. Park HS, Francis KP, Yu J, Cleary PP. Membranous cells in nasal-associated lymphoid tissue: a portal of entry for the respiratory mucosal pathogen group A streptococcus. *J Immunol* 2003;171:2532-7.
159. Ogasawara N, Kojima T, Go M, Takano K, Kamekura R, Ohkuni T, et al. Epithelial barrier and antigen uptake in lymphoepithelium of human adenoids. *Acta Otolaryngol* 2011;131:116-23.
160. Verbrugghe P, Kujala P, Waelput W, Peters PJ, Cuvelier CA. Clusterin in human gut-associated lymphoid tissue, tonsils, and adenoids: localization to M cells and follicular dendritic cells. *Histochem Cell Biol* 2008;129:311-20.
161. Kim DY, Sato A, Fukuyama S, Sagara H, Nagatake T, Kong IG, et al. The airway antigen sampling system: respiratory M cells as an alternative gateway for inhaled antigens. *J Immunol* 2011;186:4253-62.
162. Koshi R, Mustafa Y, Perry ME. Vimentin, cytokeratin 8 and cytokeratin 18 are not specific markers for M-cells in human palatine tonsils. *J Anat* 2001;199:663-74.
163. Hiller AS, Tschernig T, Kleemann WJ, Pabst R. Bronchus-associated lymphoid tissue (BALT) and larynx-associated lymphoid tissue (LALT) are found at different frequencies in children, adolescents and adults. *Scand J Immunol* 1998;47:159-62.
164. Gould SJ, Isaacson PG. Bronchus-associated lymphoid tissue (BALT) in human fetal and infant lung. *J Pathol* 1993;169:229-34.
165. Heier I, Malmstrom K, Sajantila A, Lohi J, Makela M, Jahnsen FL. Characterisation of bronchus-associated lymphoid tissue and antigen-presenting cells in central airway mucosa of children. *Thorax* 2010;66:151-6.
166. Richmond I, Pritchard GE, Ashcroft T, Avery A, Corris PA, Walters EH. Bronchus associated lymphoid tissue (BALT) in human lung: its distribution in smokers and non-smokers. *Thorax* 1993;48:1130-4.
167. Tschernig T, Pabst R. Bronchus-associated lymphoid tissue (BALT) is not present in the normal adult lung but in different diseases. *Pathobiology* 2000;68:1-8.
168. Tschernig T, Kleemann WJ, Pabst R. Bronchus-associated lymphoid tissue (BALT) in the lungs of children who had died from sudden infant death syndrome and other causes. *Thorax* 1995;50:658-60.
169. Delventhal S, Brandis A, Ostertag H, Pabst R. Low incidence of bronchus-associated lymphoid tissue (BALT) in chronically inflamed human lungs. *Virchows Arch B Cell Pathol Incl Mol Pathol* 1992;62:271-4.
170. Brandtzaeg P, Farstad IN, Johansen FE, Morton HC, Norderhaug IN, Yamanaka T. The B-cell system of human mucosae and exocrine glands. *Immunol Rev* 1999;171:45-87.
171. Zheng NY, Wilson K, Wang X, Boston A, Kolar G, Jackson SM, et al. Human immunoglobulin selection associated with class switch and possible tolerogenic origins for C delta class-switched B cells. *J Clin Invest* 2004;113:1188-201.
172. Cunningham-Rundles C, Ponda PP. Molecular defects in T- and B-cell primary immunodeficiency diseases. *Nat Rev Immunol* 2005;5:880-92.
173. Cunningham-Rundles C. Lung disease, antibodies and other unresolved issues in immune globulin therapy for antibody deficiency. *Clin Exp Immunol* 2009;157(suppl 1):12-6.
174. Buckley RH. Pulmonary complications of primary immunodeficiencies. *Paediatr Respir Rev* 2004;5(suppl A):S225-33.
175. Jesus AA, Duarte AJ, Oliveira JB. Autoimmunity in hyper-IgM syndrome. *J Clin Immunol* 2008;28(suppl 1):S62-6.
176. Holland SM, DeLeo FR, Elloumi HZ, Hsu AP, Uzel G, Brodsky N, et al. STAT3 mutations in the hyper-IgE syndrome. *N Engl J Med* 2007;357:1608-19.
177. Meyer-Bahlburg A, Renner ED, Rylaarsdam S, Reichenbach J, Schimke LF, Marks A, et al. Heterozygous signal transducer and activator of transcription 3 mutations in hyper-IgE syndrome result in altered B-cell maturation. *J Allergy Clin Immunol* 2012;129:559-62, e1-2.
178. Siegel AM, Heimall J, Freeman AF, Hsu AP, Brittain E, Brenchley JM, et al. A critical role for STAT3 transcription factor signaling in the development and maintenance of human T cell memory. *Immunity* 2011;35:806-18.
179. Speckmann C, Enders A, Woellner C, Thiel D, Rensing-Ehl A, Schlesier M, et al. Reduced memory B cells in patients with hyper IgE syndrome. *Clin Immunol* 2008;129:448-54.
180. Cunningham-Rundles C. Autoimmune manifestations in common variable immunodeficiency. *J Clin Immunol* 2008;28(suppl 1):S42-5.
181. Luger EO, Fokuhl V, Wegmann M, Abram M, Tillack K, Achatz G, et al. Induction of long-lived allergen-specific plasma cells by mucosal allergen challenge. *J Allergy Clin Immunol* 2009;124:819-26.e4.
182. Takhar P, Corrigan CJ, Smurthwaite L, O'Connor BJ, Durham SR, Lee TH, et al. Class switch recombination to IgE in the bronchial mucosa of atopic and nonatopic patients with asthma. *J Allergy Clin Immunol* 2007;119:213-8.
183. Rangel-Moreno J, Hartson L, Navarro C, Gaxiola M, Selman M, Randall TD. Inducible bronchus-associated lymphoid tissue (iBALT) in patients with pulmonary complications of rheumatoid arthritis. *J Clin Invest* 2006;116:3183-94.
184. Suda T, Chida K, Hayakawa H, Imokawa S, Iwata M, Nakamura H, et al. Development of bronchus-associated lymphoid tissue in chronic hypersensitivity pneumonitis. *Chest* 1999;115:357-63.
185. Aguilar Leon DE, Novelo Retana V, Martinez-Cordero E. Anti-avian antibodies and rheumatoid factor in pigeon hypersensitivity pneumonitis. *Clin Exp Allergy* 2003;33:226-32.
186. Araiza MT, Aguilar Leon DE, Retana VN, Martinez-Cordero E. IgM, IgG, and IgA rheumatoid factors in pigeon hypersensitivity pneumonitis. *J Clin Lab Anal* 2007;21:315-21.
187. Hogg JC, Chu F, Utokaparch S, Woods R, Elliott WM, Buzatu L, et al. The nature of small-airway obstruction in chronic obstructive pulmonary disease. *N Engl J Med* 2004;350:2645-53.
188. Gosman MM, Willemse BW, Jansen DF, Lapperre TS, van Schadewijk A, Hiemstra PS, et al. Increased number of B-cells in bronchial biopsies in COPD. *Eur Respir J* 2006;27:60-4.
189. Lee SH, Goswami S, Grudo A, Song LZ, Bandi V, Goodnight-White S, et al. Antiasthma autoimmunity in tobacco smoking-induced emphysema. *Nat Med* 2007;13:567-9.
190. Feghali-Bostwick CA, Gadgil AS, Otterbein LE, Pilewski JM, Stoner MW, Csizmadia E, et al. Autoantibodies in patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2008;177:156-63.
191. Berra A, Sterin-Borda L, Bacman S, Borda E. Role of salivary IgA in the pathogenesis of Sjogren syndrome. *Clin Immunol* 2002;104:49-57.
192. Borchers AT, Chang C, Keen CL, Gershwin ME. Idiopathic pulmonary fibrosis—an epidemiological and pathological review. *Clin Rev Allergy Immunol* 2011;40:117-34.
193. Wallace WA, Howie SE, Krajewski AS, Lamb D. The immunological architecture of B-lymphocyte aggregates in cryptogenic fibrosing alveolitis. *J Pathol* 1996;178:323-9.
194. Seetharam A, Tiriveedhi V, Mohanakumar T. Alloimmunity and autoimmunity in chronic rejection. *Curr Opin Organ Transplant* 2010;15:531-6.
195. KleinJan A, Vinke JG, Severijnen LW, Fokkens WJ. Local production and detection of (specific) IgE in nasal B-cells and plasma cells of allergic rhinitis patients. *Eur Respir J* 2000;15:491-7.
196. Van Zele T, Gevaert P, Holtappels G, van Cauwenberge P, Bachert C. Local immunoglobulin production in nasal polyposis is modulated by superantigens. *Clin Exp Allergy* 2007;37:1840-7.
197. Sabirov A, Hamilton RG, Jacobs JB, Hillman DE, Lebowitz RA, Watts JD. Role of local immunoglobulin E specific for *Alternaria alternata* in the pathogenesis of nasal polyposis. *Laryngoscope* 2008;118:4-9.
198. Tan BK, Li QZ, Suh L, Kato A, Conley DB, Chandra RK, et al. Evidence for intranasal antinuclear autoantibodies in patients with chronic rhinosinusitis with nasal polyps. *J Allergy Clin Immunol* 2011;128:1198-206.e1.
199. Liu M, Subramanian V, Christie C, Castro M, Mohanakumar T. Immune responses to self-antigens in asthma patients: clinical and immunopathological implications. *Hum Immunol* 2012;73:511-6.
200. Simpson BM, Custovic A, Simpson A, Hallam CL, Walsh D, Marolia H, et al. NAC Manchester Asthma and Allergy Study (NACMAAS): risk factors for asthma and allergic disorders in adults. *Clin Exp Allergy* 2001;31:391-9.

201. Nunez B, Sauleda J, Anto JM, Julia MR, Orozco M, Monso E, et al. Anti-tissue antibodies are related to lung function in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2011;183:1025-31.
202. Iwata T, Philipovskiy A, Fisher AJ, Presson RG Jr, Chiyo M, Lee J, et al. Anti-type V collagen humoral immunity in lung transplant primary graft dysfunction. *J Immunol* 2008;181:5738-47.
203. Sato M, Hirayama S, Hwang DM, Lara-Guerra H, Wagnetz D, Waddell TK, et al. The role of intrapulmonary de novo lymphoid tissue in obliterative bronchiolitis after lung transplantation. *J Immunol* 2009;182:7307-16.
204. Kurosu K, Takiguchi Y, Okada O, Yumoto N, Sakao S, Tada Y, et al. Identification of annexin 1 as a novel autoantigen in acute exacerbation of idiopathic pulmonary fibrosis. *J Immunol* 2008;181:756-67.
205. Salomonsson S, Wahren-Herlenius M. Local production of Ro/SSA and La/SSB autoantibodies in the target organ coincides with high levels of circulating antibodies in sera of patients with Sjogren's syndrome. *Scand J Rheumatol* 2003;32:79-82.
206. Smurthwaite L, Durham SR. Local IgE synthesis in allergic rhinitis and asthma. *Curr Allergy Asthma Rep* 2002;2:231-8.
207. Blyth DI, Pedrick MS, Savage TJ, Hessel EM, Fattah D. Lung inflammation and epithelial changes in a murine model of atopic asthma. *Am J Respir Cell Mol Biol* 1996;14:425-38.
208. Randall TD. Bronchus-associated lymphoid tissue (BALT) structure and function. *Adv Immunol* 2010;107:187-241.
209. McSharry C. B lymphocytes in allergic alveolitis. *Clin Exp Allergy* 2003;33:159-62.
210. Drent M, van Velzen-Blad H, Diamant M, Wagenaar SS, Donckerwolck-Bogaert M, van den Bosch JM. Differential diagnostic value of plasma cells in broncho-alveolar lavage fluid. *Chest* 1993;103:1720-4.
211. Martinez-Cordero E, Negrete-Garcia MC, Mendoza A. Rheumatoid factor activity in serum and bronchoalveolar lavage fluid from patients with acute hypersensitivity pneumonitis. *J Investig Allergol Clin Immunol* 1992;2:254-7.
212. Churg A, Zay K, Shay S, Xie C, Shapiro SD, Hendricks R, et al. Acute cigarette smoke-induced connective tissue breakdown requires both neutrophils and macrophage metalloelastase in mice. *Am J Respir Cell Mol Biol* 2002;27:368-74.
213. Saetta M, Turato G, Facchini FM, Corbino L, Lucchini RE, Casoni G, et al. Inflammatory cells in the bronchial glands of smokers with chronic bronchitis. *Am J Respir Crit Care Med* 1997;156:1633-9.
214. Bosken CH, Hards J, Gatter K, Hogg JC. Characterization of the inflammatory reaction in the peripheral airways of cigarette smokers using immunocytochemistry. *Am Rev Respir Dis* 1992;145:911-7.
215. Kelsen SG, Aksoy MO, Georgy M, Hershtman R, Ji R, Li X, et al. Lymphoid follicle cells in chronic obstructive pulmonary disease overexpress the chemokine receptor CXCR3. *Am J Respir Crit Care Med* 2009;179:799-805.
216. van der Strate BW, Postma DS, Brandsma CA, Melgert BN, Luinge MA, Geerlings M, et al. Cigarette smoke-induced emphysema: a role for the B cell? *Am J Respir Crit Care Med* 2006;173:751-8.
217. Zhu J, Qiu Y, Valobra M, Qiu S, Majumdar S, Matin D, et al. Plasma cells and IL-4 in chronic bronchitis and chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2007;175:1125-33.
218. Polosukhin VV, Cates JM, Lawson WE, Zaynagetdinov R, Milstone AP, Massion PP, et al. Bronchial secretory immunoglobulin A deficiency correlates with airway inflammation and progression of chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2011;184:317-27.
219. Crestani B. The respiratory system in connective tissue disorders. *Allergy* 2005;60:715-34.
220. Lafyatis R, O'Hara C, Feghali-Bostwick CA, Matteson E. B cell infiltration in systemic sclerosis-associated interstitial lung disease. *Arthritis Rheum* 2007;56:3167-8.
221. Salomonsson S, Jonsson MV, Skarstein K, Brokstad KA, Hjelmstrom P, Wahren-Herlenius M, et al. Cellular basis of ectopic germinal center formation and autoantibody production in the target organ of patients with Sjogren's syndrome. *Arthritis Rheum* 2003;48:3187-201.
222. Pulkkinen V, Salmenkivi K, Kinnula VL, Sutinen E, Halme M, Hodgson U, et al. A novel screening method detects herpesviral DNA in the idiopathic pulmonary fibrosis lung. *Ann Med* 2012;44:178-86.
223. Campbell DA, Poulter LW, Janossy G, du Bois RM. Immunohistological analysis of lung tissue from patients with cryptogenic fibrosing alveolitis suggesting local expression of immune hypersensitivity. *Thorax* 1985;40:405-11.
224. Marchal-Somme J, Uzunhan Y, Marchand-Adam S, Valeyre D, Soumelis V, Crestani B, et al. Cutting edge: nonproliferating mature immune cells form a novel type of organized lymphoid structure in idiopathic pulmonary fibrosis. *J Immunol* 2006;176:5735-9.
225. Wallace WA, Howie SE. Upregulation of tenascin and TGFbeta production in a type II alveolar epithelial cell line by antibody against a pulmonary auto-antigen. *J Pathol* 2001;195:251-6.
226. Sato A, Chida K, Iwata M, Hayakawa H. Study of bronchus-associated lymphoid tissue in patients with diffuse panbronchiolitis. *Am Rev Respir Dis* 1992;146:473-8.
227. Huggins KG, Brostoff J. Local production of specific IgE antibodies in allergic rhinitis patients with negative skin tests. *Lancet* 1975;2:148-50.
228. Nakajima S, Gillespie DN, Gleich GJ. Differences between IgA and IgE as secretory proteins. *Clin Exp Immunol* 1975;21:306-17.
229. Merrett TG, Houri M, Mayer AL, Merrett J. Measurement of specific IgE antibodies in nasal secretion—evidence for local production. *Clin Allergy* 1976;6:69-73.
230. Deuschl H, Johansson SG. Specific IgE antibodies in nasal secretion from patients with allergic rhinitis and with negative or weakly positive RAST on the serum. *Clin Allergy* 1977;7:195-202.
231. Platts-Mills TA. Local production of IgG, IgA and IgE antibodies in grass pollen hay fever. *J Immunol* 1979;122:2218-25.
232. Small P, Barrett D, Frenkiel S, Rochon L, Cohen C, Black M. Local specific IgE production in nasal polyps associated with negative skin tests and serum RAST. *Ann Allergy* 1985;55:736-9.
233. Jones E, Frenkiel S, Small P, Rochon L. Immunopathological characteristics of nasal polyps. *J Otolaryngol* 1987;16:19-22.
234. Hoffmann-Sommergruber K, Ferreira ED, Ebner C, Barisani T, Korninger L, Kraft D, et al. Detection of allergen-specific IgE in tears of grass pollen-allergic patients with allergic rhinoconjunctivitis. *Clin Exp Allergy* 1996;26:79-87.
235. Shatkin JS, Delsupehe KG, Thisted RA, Corey JP. Mucosal allergy in the absence of systemic allergy in nasal polyposis and rhinitis: a meta-analysis. *Otolaryngol Head Neck Surg* 1994;111:553-6.
236. Yoshida T, Usui A, Kusumi T, Inafuku S, Sugiyama T, Koide N, et al. A quantitative analysis of cedar pollen-specific immunoglobulins in nasal lavage supported the local production of specific IgE, not of specific IgG. *Microbiol Immunol* 2005;49:529-34.
237. Bellanti JA, Sanga RL, Klutinis B, Brandt B, Artenstein MS. Antibody responses in serum and nasal secretions of children immunized with inactivated and attenuated measles-virus vaccines. *N Engl J Med* 1969;280:628-33.
238. Cameron LA, Durham SR, Jacobson MR, Masuyama K, Juliusson S, Gould HJ, et al. Expression of IL-4, Cepsilon RNA, and Iepsilon RNA in the nasal mucosa of patients with seasonal rhinitis: effect of topical corticosteroids. *J Allergy Clin Immunol* 1998;101:330-6.
239. Chu VT, Frohlich A, Steinhäuser G, Scheel T, Roch T, Fillatreau S, et al. Eosinophils are required for the maintenance of plasma cells in the bone marrow. *Nat Immunol* 2011;12:151-9.
240. Tan BK, Schleimer RP, Kern RC. Perspectives on the etiology of chronic rhinosinusitis. *Curr Opin Otolaryngol Head Neck Surg* 2010;18:21-6.
241. Tieu DD, Kern RC, Schleimer RP. Alterations in epithelial barrier function and host defense responses in chronic rhinosinusitis. *J Allergy Clin Immunol* 2009;124:37-42.
242. Tan BK, Zirkle W, Chandra RK, Lin D, Conley DB, Peters AT, et al. Atopic profile of patients failing medical therapy for chronic rhinosinusitis. *Int Forum Allergy Rhinol* 2011;1:88-94.
243. Settipane GA, Chafee FH. Nasal polyps in asthma and rhinitis. A review of 6,037 patients. *J Allergy Clin Immunol* 1977;59:17-21.
244. Patadia M, Dixon J, Conley D, Chandra R, Peters A, Suh LA, et al. Evaluation of the presence of B-cell attractant chemokines in chronic rhinosinusitis. *Am J Rhinol Allergy* 2010;24:11-6.
245. Kato A, Truong-Tran AQ, Scott AL, Matsumoto K, Schleimer RP. Airway epithelial cells produce B cell-activating factor of TNF family by an IFN-beta-dependent mechanism. *J Immunol* 2006;177:7164-72.
246. Litinskiy B, Nardelli B, Hilbert DM, He B, Schaffer A, Casali P, et al. DCs induce CD40-independent immunoglobulin class switching through BLyS and APRIL. *Nat Immunol* 2002;3:822-9.
247. Ghaffar O, Lavigne F, Kamil A, Renzi P, Hamid Q. Interleukin-6 expression in chronic sinusitis: colocalization of gene transcripts to eosinophils, macrophages, T lymphocytes, and mast cells. *Otolaryngol Head Neck Surg* 1998;118:504-11.
248. Hulse KE, Norton J, Suh L, Zhong Q, Mahdavinia M, Simon P, et al. Chronic rhinosinusitis with nasal polyps is characterized by B-cell inflammation and EBV-induced protein 2 expression. *J Allergy Clin Immunol* 2013;131:1075-83.
249. Sanchez-Segura A, Brieve JA, Rodriguez C. Regulation of immunoglobulin secretion by plasma cells infiltrating nasal polyps. *Laryngoscope* 2000;110:1183-8.

250. Van Zele T, Gevaert P, Watelet JB, Claeys G, Holtappels G, Claeys C, et al. Staphylococcus aureus colonization and IgE antibody formation to enterotoxins is increased in nasal polyposis. *J Allergy Clin Immunol* 2004;114:981-3.
251. Castigli E, Wilson SA, Scott S, Dedeoglu F, Xu S, Lam KP, et al. TACI and BAFF-R mediate isotype switching in B cells. *J Exp Med* 2005;201:35-9.
252. Mackay F, Woodcock SA, Lawton P, Ambrose C, Baetscher M, Schneider P, et al. Mice transgenic for BAFF develop lymphocytic disorders along with autoimmune manifestations. *J Exp Med* 1999;190:1697-710.
253. Kumar R, Williams LK, Kato A, Peterson EL, Favoreto S Jr, Hulse K, et al. Genetic variation in B cell-activating factor of the TNF family (BAFF) and asthma exacerbations among African American subjects. *J Allergy Clin Immunol* 2012;130:996-9.e6.
254. Carson KR, Evens AM, Richey EA, Habermann TM, Focosi D, Seymour JF, et al. Progressive multifocal leukoencephalopathy after rituximab therapy in HIV-negative patients: a report of 57 cases from the Research on Adverse Drug Events and Reports project. *Blood* 2009;113:4834-40.

Reviewer Board

The JACI Reviewer Board is selected by the Editor-in-Chief to acknowledge a strong record of service to the Journal. The Editors thank the current members of our Reviewer Board:

Daniel C. Adelman, MD	Azzeddine Dakhama, PhD	Fu-Tong Liu, MD, PhD
Ioana Agache, MD	Stephen C. Dreskin, MD, PhD	Eric M. Macy, MD
Zuhair K. Ballas, MD, PhD	Markus J. Ege, MD, MPH	Bruce D. Mazer, MD
Donald H. Beezhold, PhD	Thomas Eiwegger, MD	Akio Mori, MD, PhD
Bruce G. Bender, PhD	Matthew Greenhawt,	Ariel Munitz, PhD
Malcolm N. Blumenthal, MD	MD, MBA, MSc	Oscar Palomares, PhD
Eric B. Brandt, PhD	Gabriele Grunig, DMV, PhD	Lars K. Poulsen, PhD
Albrecht Bufe, MD, PhD	David P. Huston, MD	Robert E. Reisman, MD
Javier Chinen, MD, PhD	Daniel Jackson, MD	Stephen I. Rosenfeld, MD
Donald W. Cockcroft, MD, FRCP(C)	John M. Kelso, MD	Ulla Seppälä, PhD
Scott P. Commins, MD, PhD	Alex KleinJan, PhD	Jonathan M. Spergel, MD, PhD
Ronina A. Covar, MD	Daphne Koinis-Mitchell, PhD	P. Brock Williams, PhD
		Bruce L. Zuraw, MD