Insights into the heterogeneity of human B cells: diverse functions, roles in autoimmunity, and use as therapeutic targets

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Abstract

B cells are critical players in the orchestration of properly regulated immune responses, providing protection against infectious agents without inflicting auto-inflammatory damage. A balanced B cell compartment is also essential to create protective immunity in response to vaccines. This difficult compromise is achieved through the finely regulated participation of multiple B cell populations with different antibody-dependent and independent functions. Both types of functions allow B cells to powerfully modulate other components of the innate and adaptive immune system. For the most part however, the necessary division of labor among different B cell populations is poorly understood. B cell dysfunction has been implicated in multiple autoimmune conditions. The physiological importance and complexity of B cell functions has been brought to the fore in recent years by the success of rituximab-based B cell depletion therapy (BCDT) in multiple autoimmune diseases including Rheumatoid Arthritis (RA) and Multiple Sclerosis (MS) which are conventionally viewed as T-cell mediated conditions. Given the widespread utilization of BCDT in malignant and autoimmune diseases and the key role of B cells in both protective immunity and pathogenic autoimmunity, a better understanding of B cell functions is of the essence and a focus of the research in our division. We are investigating these issues through a variety of approaches, including the study of the phenotype and function of human B cell populations in health, their perturbation in autoimmune disease states, the effects of targeted biologic therapies, and the study of relevant murine models.

Keywords

B cells; autoimmunity; systemic lupus; tolerance

Introduction

B cells play critical protective roles in maintaining health (defense against infection, effectiveness of vaccines, protection against atherosclerosis, cancer surveillance), but also play central pathogenic roles in multiple autoimmune diseases (affecting up to 5% of the entire population), malignant diseases, and transplant rejection. In addition to antibody production, both protective and pathogenic roles of B cells are mediated by poorly understood antibody-independent mechanisms. We have postulated that the physiological balance between protective and pathogenic functions is ensured by a division of labor among multiple B cell populations (Figure 1).

Moreover, the clinical and immunological outcome of B cell depletion therapy (BCDT) will depend on the relative balance of protective and pathogenic B cell subsets established upon B cell repopulation. BCDT with rituximab is widely used for the treatment of non-Hodgkin follicular lymphoma (NHL). It is estimated that close to one million lymphoma patients have been treated with rituximab since the drug was FDA-approved in 1997 (1). More recently, this modality has also been approved for the treatment of rheumatoid arthritis refractory to TNF blockade. In addition, rituximab is increasingly used off-label and is being formally tested for the treatment of multiple autoimmune diseases. Given that repeated BCDT leading to sustained depletion of B cells is expected to be used in millions of patients in the near future, there is a pressing need to better understand antibody-independent B cell functions, their role in health and disease, and the impact of BCDT on protective and pathogenic functions. In this review, we will discuss our current understanding of human B cell populations from a phenotypic and functional perspective, as well as the dysregulation that occurs in autoimmunity.

1. Heterogeneity of human B cells

1.1 Definition using surface markers

Human B cells have been classified on the basis 4 major surface markers (CD19, IgD, CD38 and CD27) measured in 3 or 4-color combinations (2,3) (Table 1). The Bm1-Bm5 (B mature) classification identifies multiple tonsil B cell subsets: virgin naïve cells (Bm1: IgD+CD38-); activated naïve cells (Bm2: IgD+CD38lo); pre-GC cells (Bm2': IgD+CD38++); GC cells (Bm3-centroblasts and Bm4-centrocytes: IgD-CD38++); and memory cells (Bm5: IgD-CD38+/-). Bm5 memory cells have been further divided into early Bm5 (CD38lo) and Bm5 (CD38-) (4). The implied chronological relationship between the latter subset has never been formally established, although CD38 is likely to identify activated memory cells (3,5,6). While this classification represents an almost mandatory point of reference, it suffers from significant limitations. Thus, Bm1 and Bm2 cells also contain unswitched memory cells and the Bm2' subsets also contains transitional cells (3,7,8).

The IgD/CD27 classification builds on the notion of CD27 as a universal marker of human memory B cells to distinguish between memory cells (CD27+) and naïve B cells (CD27-/IgD+). In turn, CD27+ memory cells can be divided into IgD+ (unswitched memory; usually together with IgM) and IgD- (switched memory; predominantly IgG+ or IgA+) (3). Mature IgD+/CD27- naïve cells (Bm1/2) represent the human B2 follicular compartment, and switched CD27+ cells are thought to represent their post-GC memory progeny. In addition, the human spleen contains a population of marginal zone (MZ) B cells with a phenotype similar to its murine counterpart (3). The vast majority of MZ B cells express CD27 and CD1c and some degree of somatic hypermutation. Moreover, in contrast to the mouse, a subset of human spleen MZ cells also express IgD. Whether these cells indeed represent real memory cells and whether unswitched CD27+ cells (about 15% of all PBL B cells) represent a recirculating MZ population remains contested in the field (9–11). The existence and actual identity of a human B1 cell equivalent continues to be debated due in part to the inability of CD5 expression to identify a separate population.

1.2. Human memory B cells

As discussed by ourselves and others, our understanding of memory B cells is hampered by pre-conceived definitions (3,12). Despite these caveats, memory responses can be heterogeneous in mice and include conventional B2 IgG memory to T cell-dependent (TD) antigens and B1b-mediated IgM memory to T cell-independent type 2 (TI-2) antigens (13–15). B2 cells with a phenotype different from both follicular and marginal zone (MZ) B cells can also provide IgG memory to TI-2 antigens (16). Several populations of B cells,

including a highly mutated CD21/CD35^{low} subset, provide memory responses to a single antigen (17). A similar population of human CD21[−] cells is also likely to represent activated effector memory cells (5,6,18). In humans, the use of CD27 as a universal marker of memory has hampered progress. Yet, over the last few years we and others have demonstrated the existence of important populations of CD27[−] memory cells (5,19–22). Our results with multi-color flow cytometry also demonstrate additional heterogeneity within both CD27⁺ and CD27[−] memory cells (3,5). A summary of our current understanding of human memory B cell diversity is included in Table 1.

1.3. Antibody independent functions and cytokine producing ability of B cells

It has been known for many years that B cells can produce cytokines with immunosuppressive, polarizing, inflammatory and tissue-organizing properties, yet the potential biologic relevance of cytokine-producing B cells was largely unappreciated. However, recent findings have renewed interest in this area and have raised the intriguing possibility that cytokine-producing B cells actively modulate both humoral and cellular immune responses (23,24). Overwhelming evidence for the participation of specialized effector T cell responses in multiple autoimmune diseases has been provided by mouse models (25). Ample evidence also exists for specialized subsets of effector and regulatory human T cells although their regulation, antigen-specificity and skewing in different autoimmune diseases remain to be fully elucidated (25–28). Much less is known about B cell division of labor. However, the elegant description *in vitro* and *in vivo* by one of our investigators (Dr. Lund) of mouse effector B cell subsets (Be1 and Be2) with polarized cytokine secretion has been a major first step (29,30). The notion that different B cell subsets may induce separate T cell subsets has recently been expanded by the demonstration that B1 cells may promote Th1 and Th17 differentiation (31). Of note, both protective and pathogenic B cell functions may be mediated by antibody-independent mechanisms (24). Dr. Lund's work with pneumocystis and H polygyrus infection powerfully illustrates the importance of protective antibody-independent B cell functions (24,32).

From an autoimmunity standpoint, B cells may either stimulate or inhibit pathogenic responses. Numerous examples of pathogenic roles have been provided by mouse models using either B cell deficient mice or B cell depletion (33,34). On the other hand, growing evidence is accumulating for regulatory B cells capable of preventing or suppressing autoimmunity in different mouse models (23,35). This protective role can be mediated by inducing T cell anergy during antigen presentation or inducing Treg expansion or activity, (31,35,36). B cells may also directly suppress Th1 and Th17-mediated diseases (37). These activities are mediated, at least in part, by the production of IL-10 or TGF β and may control a variety of auto-inflammatory diseases including: inflammatory arthritis, inflammatory bowel disease, autoimmune diabetes, experimental autoimmune encephalitis and contact hypersensitivity (31,38–47).

However, the actual nature and mechanisms of action of Breg cells remain unclear. Mouse Breg activity has been variously assigned to cells with a transitional (in particular, T2-Marginal Zone Precursors – T2/MZP), Marginal Zone (MZ) or B1/transitional/MZ intermediate phenotype and lupus resistance has been associated with expansion of MZ cells (45,48–50). As we have discussed elsewhere, the existence of human Bregs remains to be fully demonstrated (51,52). However, our observation that expansion of transitional B cells correlates with long-term remission in SLE patients treated with BCDT (B cell depletion therapy) is consistent with a regulatory nature. Finally, it has been proposed that IL-10 production may be impaired in naïve B cells of patients with active MS and that a reversal of this deficit correlates with good response to BCDT (53). While not establishing the actual function of a given B cell subset, the ability to produce certain cytokines provides a first clue as to function and a starting point to define functional changes in human disease. Human B

cells can secrete multiple cytokines depending on the B cell subset tested and the stimulatory conditions used (23,51–60). Moreover, cytokine-producing human B cells can induce Th1 and Th2 cell (54–56). It has also been proposed that CD27+ Be1 cells may be accumulate in the salivary glands in primary Sjögren's syndrome (pSS) and that CD27+ cell producing TNF may contribute to Multiple Sclerosis (MS) (52,53).

Cytokine-producing B cells can also regulate lymphoid tissue formation and maintenance. Ectopic lymphoid tissue is often observed in the target organs of autoimmune patients and mice (61). While it is still debated whether these tissues directly lead to disease or form in response to disease, it is clear that these inducible lymphoid tissues can participate in local immune responses and sustain autoreactive B and T cells. It is also known that B cells are involved in the formation and/or maintenance of tertiary lymphoid tissues, as these tissues do not efficiently develop in B cell deficient mice (62). Furthermore, elegant experiments from the Weyand laboratory revealed that depleting B cells in synovial biopsies from RA patients caused diminishment of the ectopic follicles and, even more importantly, reduced the autoreactive T cell response (63). Interestingly, recent studies revealed the presence of cytokine-producing B cells (IFN γ , IL-6, TNF α , LT α) directly within the ectopic lymphoid tissues (62,64). The question of how B cells regulate the development or maintenance of ectopic lymphoid tissues is still unanswered. However, B cell-derived LT α plays important roles in the maintenance of lymphoid architecture in lymph nodes (LN) and spleen. For example, B cell-derived LT α is required for the development of follicular dendritic cell networks, subsets of splenic stromal cells, the production of homeostatic chemokines, the subsequent recruitment of DCs, T cells and additional B cells, and the formation of germinal centers (65). Likewise, recent data suggests that B cells, particularly VEGF-A-producing B cells, play important roles in regulating local lymphangiogenesis in antigen-stimulated LNs (66). Based on these data, investigators in our group (Dr. Randall) have proposed that cytokine-producing B cells contribute to the induction and maintenance of the local inflammatory response in autoimmune target organs and in this way exacerbate pathology (67).

We have found that B cell depletion may alter lymphoid architecture by eliminating LT α bearing and TNF secreting B cells (68–70), resulting in a prolonged delay in memory B cell reconstitution (7). Interestingly, we have further shown that treatment of RA with etanercept (TNF receptor-Ig p75 decoy that binds both TNF and LT α) may mimic these findings via blockade of TNF and LT signaling pathways and inhibition of FDCs (71). Our findings suggest an unexpected convergence of mechanism between TNF blockade and B cell depletion therapy as both seemingly divergent approaches can reduce memory B cells.

2. The opposing roles of B cells in autoimmunity

2.1. Subversion of B cell function in human SLE

The functional versatility of B cells enables them to play either protective or pathogenic roles in autoimmunity. As we have recently reviewed and alluded to above (1,51), B cells may be deleterious through the production of pathogenic autoantibodies, activation of autoreactive T cells, production of pro-inflammatory cytokines and organization of ectopic lymphoid tissue. On the other hand, B cells may prevent or suppress established autoimmunity through anti-inflammatory cytokines such as IL-10 and TGF β and the expansion of Tregs and/or inhibition of effector T cells (1,23,33,42,51). At a population level, separate B cell subsets may exert different functions as indicated by the differential ability of mouse Be1, Be2 and B1 cells to induce effector T cell subsets that might contribute to disease (Th1, Th2 and Th1/Th17, respectively) while B2 cells may preferentially induce Treg differentiation (31). In other cases however, B cells may contribute to disease by suppressing Tregs (72). Understanding the imbalance between these

opposing B cell functions in disease is a critical focus of ongoing research in the division (Fig. 1).

2.2. Abnormal B cell homeostasis in SLE

Multiple B cell alterations have been reported in human SLE. A summary of alterations described in SLE and their correlation with disease activity is provided in Table 2. The most prominent abnormalities seen in active disease include naïve lymphopenia, relative increase in CD27+ switched memory and expansion of CD27+++ PB. Expansion of T1 cells has also been reported. Our studies (highlighted in blue), have identified the expansion of other subsets including switched CD27– memory cells and transitional T2/T3 cells. Of significant interest, these abnormalities have been described either in isolation or in different combinations due, to a large extent, to the use of pauci-color FACS analysis. Still, it seems obvious that B cell alterations in SLE peripheral blood are more prominent and fluid than in other autoimmune diseases. We surmise that this reflects the systemic nature of the autoimmune process with frequent cycles of activation, differentiation and traffic between secondary lymphoid organs and target tissues.

3. Impact of B cell targeted therapies on B cell abnormalities in SLE

3.1. B cell depletion therapy (BCDT)

FDA-approved for RA refractory to anti-TNF, BCDT has also proven effective in randomized placebo controlled trials of Relapsing-Remitting MS (73,74). Phase III are also underway for other autoimmune diseases including Type 1 Diabetes (75,76). Significant benefit has also been reported (often for severe refractory cases) in open label studies and case series for many other autoimmune diseases (1,77–79). As for SLE, our initial phase I/II study and other similar open-label trials have provided strong support for this therapy which has become a useful tool for refractory SLE with either renal, CNS or hematological involvement (80–85). Different mechanisms of action have been invoked to explain the benefit of rituximab in SLE and other diseases including the elimination of autoantibodies, the restoration of proper Th1/Th2 balance, the expansion of Treg cells, the disruption of ectopic lymphoid tissue, and the elimination of effector B cells from target organs (1,86–92). Our own work has suggested that at least early benefit cannot be simply explained by autoantibody reduction (19,93). A major focus of our research efforts is to better understand the different functions played by B cells and the impact of BCDT on protective and pathogenic B cell functions.

Along these lines, our group has provided many of the original observations regarding the timing and quality of B cell depletion and repopulation after treatment of SLE with rituximab (7,19,93,94). We have proposed that a repopulation with transitional cells is associated with sustained clinical remission while a quick resurgence of memory cells portends a poor outcome. These observations have been confirmed by other groups and also appear to apply to RA patients (albeit a long clinical remission is less common at least in TNF-refractory patients) (95).

3.2. B cell depletion reverses B cell abnormalities in SLE

Systematic follow-up of BCDT in SLE for longer than 7 years has provided significant insight into the immunological consequences of B cell depletion. Of central importance, we have identified two different groups with dramatically diverging clinical and immunological responses (Figure 2) (1,19,93). Group A (30%), has maintained clinical remission to date without additional therapy with progressive disappearance of lupus-specific autoantibodies (anti-DNA and 9G4) antibodies) and in some cases of all ANA. Group B (70%) experiences transient improvement and disease flares ensue with the re-emergence of B cells.

Immunologically, both groups are also dramatically different. Thus, while significant B cell depletion was initially achieved in both groups, the CD27⁺ memory compartment in group A remained significantly depleted for up to 3–5 years after treatment ($p=0.0000046$ compared to normals) and there was no re-accumulation of switched CD27⁺ cells expanded at baseline (7,19) (Table 3). Moreover, B cell tolerance (measured by proper GC censoring of 9G4 cells) was restored in these patients (96). This is a remarkable finding given that our studies of hundreds of follicles in 12 SLE tonsils and 3 SLE spleens demonstrate that defective tolerance resulting in productive 9G4⁺ GCs is a universal feature of SLE. In contrast to good responders, group B was characterized by rapid re-accumulation of memory B-cells and abnormal GC tolerance (Figure 2). We concluded that group A undergoes slow de novo immune reconstitution without reemergence of autoimmunity (97,98). In contrast, reconstitution in poor responders may be explained by expansion of residual autoreactive memory B cells in a lymphopenic environment. Consistent with this model, we have recently shown that poor responders are also more likely to have an increased fraction of CD19^{high} B cells characterized by an activated, pre-plasma cell phenotype, enrichment for anti-Sm autoantibody precursors and propensity to migration towards inflamed tissues as indicated by expression of CXCR3 and chemotaxis to CXCL9 (99).

3.3. Understanding the response to BCDT. The importance of comprehensive B cell phenotyping

Good responders display a dominance of IgD⁺ CD27⁺ cells that is sustained for several years in the absence of significant numbers of memory cells. While these cells are widely assumed to be naïve, the use of multi-color analysis indicates instead a transitional population initially comprised of T1 and T2 subsets. After approximately 18 months, a new population becomes apparent with phenotypic and functional properties intermediate between T2 and mature naïve B cells (1,7,94). This population, which we have labeled T3 by analogy with similar mouse transitional cells, can only be differentiated from fully mature B cells by inability to extrude R123. Interestingly, our data indicate that the majority of T3 are CD10⁺ and therefore, the absence of this marker cannot be taken as an absolute definition of naïve cells in human PBL (100). Of note, T3 cells represent the major PBL fraction in good responders for up to 3 years. Of significant interest, expansion of transitional cells also correlates with prevention of diabetes after B cell depletion in NOD mice (46). One critical question that remains to be addressed is whether the benefit of BCDT is directly mediated by the expanded transitional cells or instead reflects the absence of effector B cells. Of course, these scenarios are not mutually exclusive since transitional cells could themselves be responsible for suppressing effector response.

Poorer outcome with preferential memory repopulation has been reproduced in SLE, RA and MS (74,95). These studies raise critical questions regarding the factors that influence depletion, the kinetics and quality of repopulation and the mechanisms responsible for long-term remission in a subset of patients. In SLE, we showed that poor B cell depletion is determined by race, low-avidity FcγRIIIa 158V/F gene polymorphism and very high levels of HACA (93). Importantly, we also showed that autoantibodies against RNA-binding proteins (RBP: Ro, La, Sm/RNP) strongly predict poor response (7). We propose that RBP antibodies, good inducers of IFNα production, contribute to activation and expansion of memory B cells (101,102). Moreover, the persistence of RBP antibodies would help explain the dominant expansion of memory B cells during repopulation. We are currently exploring these hypotheses in mouse models utilizing the murine equivalent of rituximab (Dr. Anolik). As supported by our findings and others (92), we surmise that RBP persistence is due to preferential production by long-lived bone marrow PC that are not targeted by rituximab.

4. Conclusion and Perspectives

In summary, the heterogeneity in human B cell populations is becoming increasingly better elucidated. However, an important limitation of current knowledge regarding the role of B cells in autoimmunity is that very little has been known in human disease beyond the production of autoantibodies. Yet, accumulating data in diverse autoimmune diseases, including SLE and RA, indicates that B cells likely contribute to disease through multiple mechanisms that include both antibody-dependent and antibody-independent functions. The latter include antigen-presentation, T-cell activation and polarization, and dendritic cell modulation and we propose are critically mediated by the ability of B cells to produce cytokines. Indeed, we suggest that human B cells display phenotypic diversity that reflects division of labor for effector and regulatory functions and a striking imbalance in SLE, one that may be restored with targeted biologic therapies. Further understanding the imbalance between opposing B cell functions in disease remains an important focus of study.

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Figure 1. Model of B cell effector and regulatory functions in health and disease

Immunologic homeostasis is dependent on the balance between protective (regulatory, anti-inflammatory) B cells and effector (pro-inflammatory) B cells and their corresponding cytokines. We postulate that physiologically, transitional cells predominantly produce IL10 which in a normal environment exerts anti-inflammatory actions, opposing IL12-mediated DC induction of Th1 cells, contributing to the induction of Treg cells, and suppressing autoreactive Th1 cells. This situation would be altered in SLE due to abnormal numbers or function of transitional cells and the cytokines they produce, as well as the type I interferon dominated milieu which primes proinflammatory functions of IL10. In addition, dysregulated effector memory B cells in SLE would produce an excess of pro-inflammatory cytokines (IFN γ , IL-12p40, TNF), shifting the scales toward a proinflammatory phenotype. Finally, we postulate that prolonged absence of B cells induced by BCDT has the potential to either restore physiological balance between protective and pathogenic B cell functions or even to create at least temporarily an environment dominated by transitional cells with anti-inflammatory and tolerogenic functions and Treg inducing activity

Figure 2. Timing and intensity of depletion and repopulation in two groups of SLE responses to BCDT

(A/B) Both groups depleted to < 5 CD19⁺ cells/ μ l. Group A repopulates with high absolute B cell numbers. Good responders are characterized by dominant transitional (T) and absent memory (M) cells for up to 3 years after BCDT in a pattern resembling neonatal ontogeny. In contrast, memory cells dominate in poor responders. (C) Comparison of total and autoreactive 9G4 tonsil B cells in normal healthy control, RA and SLE patients. Active SLE (but not RA) is characterized by expansions of GC and M cells (in particular activated CD38^{low} early Bm5). These abnormalities are stably reversed by BCDT as illustrated in a good responder studied 4 years after BCDT. Autoreactive 9G4 cells are effectively excluded from GC in healthy control and RA but not in active SLE. Strikingly, this breach of tolerance is restored with good Rituximab response.

Table 1

Classification of human PBL B cell subsets.
Table 1. Surface phenotype of human peripheral B calls

Markers	CD27	IgD	IgM	IgG	CD38	B220	CD21	CD1c	CD24	CD10	R123
Transitional	T1	-	+++	-	+++	ND	+/-	ND	+++	+	+
	T2	-	++	-	++		+		++	(+),-	+
	T3	-	++	-	+		+		+	(-),+	++
Naive		-	++	+/low	+/low	+	+	-	+	-	-
Memory	CD27+ memory	UNS. IgD+	+	-	(+),-	(-),+	(+),-	(+),-	+	-	+
		UNS. IgD-	+	-	+,-	-	NA	-	+	-	+
		SW. IgD-	+	+	+,-	(-),+	(+),-	(-),+	(+),-	-	+
	CD27- memory	UNS. IgD-	+	-	+,-	-	NA	-	+	-	+
		SW. IgD-	-	+	+,-	(+),-	+,-	-	+,-	-	+
	Bm5 memory	Early Bm5	(-),+	(+),-	+	(-),+	(+),-	(-),+	(+),-	-	+
Plasmablasts (PB)	Late Bm5	(+),-	(-),+	(+),-	-	(-),+	(+),-	(-),+	(+),-	-	+
		+++	sIg (IgG/IgA) Low/-	Low/-	+++	Low/-	-	-	-	(-),+	+

This table incorporates our own data with multi-color flow cytometry. B cell subsets found in human PBL are classified according to the expression of different surface markers and the extrusion of dyes like Rhodamine-123. + and - symbols denote marker expression or absence. +(-) or (-) denotes bi-modal expression with the dominant subset shown first in parentheses. + to +++ indicates relative intensity of expression; +/low and low/- indicate spread of expression of a marker with a Bell curve distribution.

Table 2

B cell abnormalities in human SLE

B cell Subset	Change	Tissue	Activ.	Design	REF
Naïve ✓	↓	PBL, TN	+	CRS	(19, 103)
Naïve CD25+	↓	PBL	+	CRS	(60)
SW. CD27+ ✓	↑	PBL	+	CRS	(19, 103)
CD27+++ PB ✓	↑	PBL	+	CRS	(103, 104)
SW. CD27- FCRL4-	↑	PBL	+	CRS	(5)
SW. B220+ (CD21-CD95+)	↑	PBL	+	CRS	(5)
SW. 9G4+ CD27+	↑	PBL	+	CRS	(97, 98)
9G4+ PB	↑	PBL, TN	+	CRS	(97, 98, 105)
Early Bm5	↑	PBL, TN	=	CRS	(5, 98)
CD19High pre-PB ✓	↑	PBL	+	CRS	(99)
Transitional T1 ✓	↑	PBL	=	CRS	(7, 106)
Transitional T2-T3	↑	PBL	=	CRS	(7, 107)
FcγIIb in memory cells	↓	PBL	+	CRS	(108)
Pre-GC (Bm2')	↑	PBL	+	CRS	(104)
SW. CD27+, ICOSL+	↓	PBL, kidney	=	CRS	(109)

Blue font indicates abnormalities originally described by our group. ✓ sign denotes findings reproduced and/or extended in our lab. +: positive correlation with disease activity (typically by SLAM/SLEDAI); = correlation not studied or established; M: memory; MZ: marginal zone; PB: plasmablasts; PBL: peripheral blood; TN: tonsil CRS: Cross-sectional. We have reviewed this information elsewhere (1, 3, 51).

Table 3

Post-Rituximab B cell changes in human SLE

B cell Subset	Change		Tissue	Design	REF (1,3,7,19,51,93,94,110)
	Good response	Poor response			
Naïve	↑	↓	PBL, TN	LGT	
SW, CD27+	↓↓↓	↑↑↑	PBL, TN	LGT	
CD27+++ PB	↓↓↓	↑	PBL	LGT	
SW, CD27−	↓↓↓	↑	PBL, TN	LGT	
9G4+ SW, CD27+ and PB	↓↓↓	↑	PBL	LGT	
Transitional T1/T2	↑↑↑	↓	PBL	LGT	
Transitional T3	↑↑↑	↓	PBL	LGT	
MZ-like circulating M	=	+/-	PBL	LGT	
IL-10 producers (subset?)	ND*			(53)	

↑ indicates abnormalities originally described by our group. = correlation not studied or established; M: memory; MZ: marginal zone; PB: plasmablasts ND* changes in IL-10-producing B cells in SLE have not yet been published. However, restored production of IL-10 by B cells has been reported in Multiple Sclerosis patients with good response to Rituximab. LGT: longitudinal; PBL: peripheral blood; TN: tonsil