Editorial

New paradigm of B-cell biology regarding the elucidation of a new mechanism of tissue fibrosis in IgG_4-related disease

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Fibrosis, which occurs in many organs including the lung, liver, kidney, and pancreas, is defined as an excessive deposition of extracellular matrix, which leads to the destruction of organ structure and impairment of organ function. Multiple immune and molecular pathways are involved in the pathophysiology of fibrosis. Many researchers have focused on the involvement of various types of immune cells, including CD4 T cells, dendritic cells, macrophages, and mast cells, in tissue fibrosis.[1] Unfortunately, despite extensive studies regarding the contribution of these cells to tissue fibrosis, experimental data have not been successfully translated to clinical application.

IgG_4-related disease (IgG_4-RD) is a systemic condition of unknown cause characterized by highly fibrotic lesions, with dense lymphoplasmacytic infiltrates containing a preponderance of IgG_4-expressing plasma cells. $CD4^+$ T cells and B cells constitute the major inflammatory cell populations in IgG_4-RD lesions. Among the $CD4^+$ T-cell subsets, T follicular helper (Tfh) cells, which provide help to B cells during T-cell–dependent immune responses and contribute to the processes of isotype switching, are considered to contribute to IgG_4-RD pathogenesis. Akiyama et al[2] reported a potential role of circulating Tfh2 cells in IgG_4-RD. Recently, it was found that in IgG_4-RD, specific $CD4^+$ cytotoxic T lymphocytes clonally expand, infiltrate disease sites, and are reactivated locally, presumably by activated cognate B cells that capture the driving antigen through the B-cell receptor, internalize it, and present it to the $CD4^+$ cytotoxic T lymphocytes, causing them to drive the inflammatory and fibrotic processes that characterize the disease.[2]

Since their discovery approximately 50 years ago, B cells have been largely recognized as being active in antibody-dependent immune responses. Interestingly, recent studies indicate that B cells appear to have an antibody-independent role and appear to be one of the emerging important players regarding tissue fibrosis because several animal studies have shown that B-cell depletion ameliorated fibrosis in lung, liver, and kidney models.[1] It is well known that IgG_4-RD is characterized by a fibroinflammatory condition with tissue fibrosis. Typical presentation and imaging findings include mass-forming synchronous or metachronous lesions in almost any organ, but most commonly in the pancreas, bile duct, retroperitoneum, kidneys, lungs, salivary and lacrimal glands, orbit, and lymph nodes.[3]

Generally, one of the most important processes of the observed pathological features of IgG_4-RD is the induction of a polarized $CD4^+$ T-cell population, which activates innate immune cells (including macrophages, myofibroblasts, and fibroblasts) to drive fibrosis, which could involve the collaboration of activated B-lineage cells, possibly expanding plasmablasts that enter the damaged tissue along with activated $CD4^+$ T cells.[4] The initial rationale for rituximab treatment for immune-mediated diseases has been to blunt the effects of disease-associated autoantibodies. For example, B-lymphocyte depletion by rituximab is effective for pemphigus vulgaris in which the autoimmune condition is mediated by IgG_4 autoantibodies.[5] In IgG_4-related disease, total IgG_4 level has been used as a surrogate for autoantibody levels, and rapid decline in serum IgG_4 levels associated with prompt clinical improvement has been demonstrated in the clinical trial of Khosroshahi et al[6] as well as the study of Carruthers et al.[7]

In this regard, mechanistic studies on patients with IgG_4-RD treated with rituximab should lead to novel insights about the pathophysiology of the disease; however, there has been no direct evidence supporting the possible mechanism of B- cell involvement in the tissue fibrosis of IgG_4-RD.

In this issue of the *Journal of Allergy and Clinical Immunology*, Della-Torre et al[8] reported the direct contribution of B lymphocytes to tissue fibrosis in patients with IgG_4-RD. Of note, they provided the first evidence that plasmablasts (which the authors focused on as a population with major involvement in IgG_4-RD pathophysiology) represent a B-cell subset with intrinsic fibrogenic potential in IgG_4-RD in contrast to both naive and memory B cells.

First, they cocultured primary human fibroblasts with $CD19^+$ B cells (naive, memory, and plasmablast) isolated from the peripheral blood of patients with IgG_4-RD and examined the profibrotic gene expression and collagen production of the cultured fibroblasts by transcriptomic analyses. The data revealed that B cells from patients with IgG_4-related autoimmune pancreatitis induced the activation of profibrotic pathways in the cocultured fibroblasts, with a significant upregulation of genes involved in epithelial-mesenchymal transition as well as increased collagen production compared with that in control fibroblasts. Second, they examined soluble profibrotic molecules secreted from B lymphocytes that contribute to the development of fibrosis by a Luminex-based approach. Interestingly, coculture of B cells from healthy controls with fibroblasts was also associated with increased collagen secretion; however, the amount of this secretion was less than that of the coculture with IgG_4-RD B cells. In

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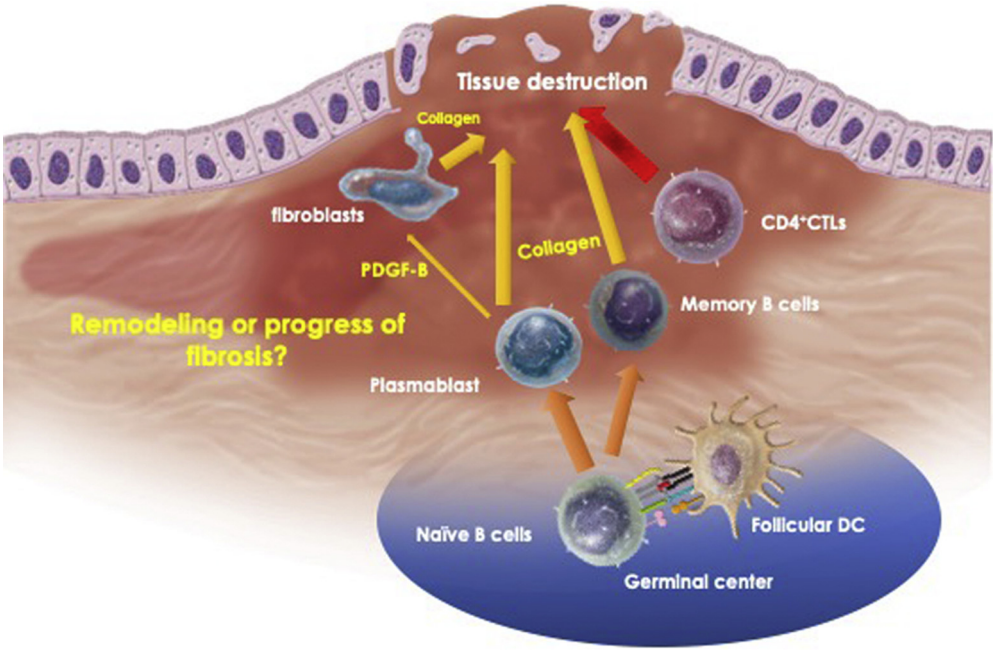

FIG 1. The contribution of B cells to tissue remodeling and fibrosis. The naive B cells differentiate into memory B cells and plasmablasts after antigen presentation by follicular dendritic cells. In IgG_4-RD, plasmablasts or memory B cells may play an important role in tissue fibrosis both by indirect and by direct collagen production. However, in normal conditions, these cells might contribute to tissue remodeling.

addition, B cells from both patients with IgG_4-RD and healthy controls could induce platelet-derived growth factor (PDGF)-B, CCL4, CCL5, and CCL11. Among these molecules, PDGF-B most biologically contributed to B-lymphocyte–dependent collagen production from human fibroblasts, which was confirmed by PDGF-B blockade examination. Moreover, in an *in vivo* study, the authors showed that not only B cells but also alpha-smooth muscle actin (SMA)+ myofibroblasts produced PDGF-B, CCL4, CCL5, and CCL11. Overall, both cells could drive inflammation in the affected lesions by orchestrating these molecules, which finally leads to tissue fibrosis.

The question is why the collagen production induced by B cells from patients with IgG_4-RD was significantly higher than that from a healthy donor despite similar levels of PDGF-B in the supernatants. A previous study demonstrated a significant correlation between the expansion of plasmablasts in patients with IgG_4-RD and serological markers as well as decreasing plasmablasts during the remission of IgG_4-RD. The authors previously showed that a population of circulating plasmablasts is disproportionally expanded in patients with active IgG_4-RD.

In this regard, the authors examined whether circulating plasmablasts have profibrotic properties. The experimental data indicated that fibroblasts cocultured with plasmablasts produced significantly more collagen than did the control fibroblasts or fibroblasts cocultured with naive or memory B cells. However, we must acknowledge that these data do not necessarily reflect the exact contribution of B cells to pancreatic fibrosis because of the use of skin fibroblasts, not pancreatic fibroblasts. In addition, although the authors strongly suggest that plasmablasts directly contribute to fibrosis in the pathogenesis of IgG_4-RD, it remains unclear whether the profibroblastic property of plasmablasts is specific for patients with IgG_4-RD. In this regard, it would be interesting to examine whether plasmablasts from patients with allergies or asthma have profibroblastic properties.

The next question is why plasmablasts have a stronger profibrotic property than naive and memory B cells. To elucidate the mechanism, the authors focus on lysyl oxidase-like 2 (LOXL2) expression in B cells. LOXL2 is an enzyme that promotes the network establishment of collagen fibers in the extracellular matrix. The inhibition of LOXL2 can decrease cell numbers, proliferation, colony formation, and cell growth, induce cell cycle arrest, and increase apoptosis. Recent reports have shown that LOXL2 is associated with various types of fibrotic diseases.[9]

In the study by Della-Torre et al,[8] immunohistochemistry data demonstrated abundant LOXL2 expression in B cells infiltrating IgG_4-RD tissue. Moreover, the intriguing finding is that plasmablasts from patients with IgG_4-RD expressed type I collagen genes. We still have the following question: why do plasmablasts mimic the phenotype of myofibroblasts despite their inherently short lives, irrespective of the nature of the antigen? To answer this question, additional studies with next-generation sequencing, single-cell sequencing, and DNA methylation sequencing would enable us to better identify the factors contributing to the profibrotic phenotype of plasmablasts.

Despite several limitations in this current study, it provides mechanisms for direct involvement of B cells, in particular plasmablasts, in the tissue fibrosis of patients with IgG_4-RD. What about autoantibody-mediated effects? Could some of the B-cell–mediated effects in IgG_4-related disease be mediated by autoantibodies? Recently, Shiokawa et al[10] reported laminin 511 as one of the target antigens in autoimmune pancreatitis and the induction of antibodies and pancreatic injury by the immunization of mice with human laminin 511-E8. It is possible that plasmablasts may play an important role in IgG_4-related

disease both by directed cell-mediated effects and by antigen-specific tissue damage (Fig 1).

In conclusion, the pathophysiology of IgG_4-RD is not simple. Therefore, further study is needed to understand how immune cells such as CD4 T cells (including Tfh cells) and B cells orchestrate with target autoantigens in many organs of IgG_4-RD. The current data can cause controversy regarding B-cell biology in elucidating the mechanism of autoimmune diseases, including IgG_4-RD. Thus, the development of B- cell biology continues.

REFERENCES

1. Meng XM, Nikolic-Paterson DJ, Lan HY. Inflammatory processes in renal fibrosis. Nature Rev Nephrol 2014;10:493-503.
2. Akiyama M, Suzuki K, Yamaoka K, Yasuoka H, Takeshita M, Kaneko Y, et al. Number of circulating follicular helper 2 T cells correlates with IgG4 and interleukin-4 levels and plasmablast numbers in IgG4-related disease. Arthritis Rheumatol 2015;67:2476-81.
3. Kamisawa T, Zen Y, Pillai S, Stone JH. IgG4-related disease. Lancet 2015;385:1460-71.
4. Mattoo H, Mahajan VS, Maehara T, Deshpande V, Della-Torre E, Wallace ZS, et al. Clonal expansion of CD4(+) cytotoxic T lymphocytes in patients with IgG4-related disease. J Allergy Clin Immunol 2016;138:825-38.
5. Mouquet H, Musette P, Gougeon ML, Jacquot S, Lemercier B, Lim A, et al. B-cell depletion immunotherapy in pemphigus: effects on cellular and humoral immune responses. J Invest Dermatol 2008;128:2859-69.
6. Khosroshahi A, Bloch DB, Deshpande V, Stone JH. Rituximab therapy lead to rapid decline of serum IgG4 levels and prompt clinical improvement in IgG4-related systemic disease. Arthritis Rheum 2019;62:1755-62.
7. Carruthers MN, Topazian MD, Khosroshahi A, Witzig TE, Wallace ZS, Hart PA, et al. Rituximab for IgG4-related disease: a prospective, open-label trial. Ann Rheum Dis 2015;74:1171-7.
8. Della-Torre E, Rigamonti E, Perugino C, Baghai-Sain S, Sun N, Kaneko N, et al. B lymphocytes directly contribute to tissue fibrosis in patients with IgG4-related disease. J Allergy Clin Immunol 2020;145:968-81.e14.
9. Puente A, Fortea JI, Cabezas J, Arias Loste MT, Iruzubieta P, Lierena S, et al. LOXL2-A new target in antifibrogenic therapy? Int J Mol Sci 2019;20.
10. Shiokawa M, Kodama Y, Sekiguchi K, Kuwada T, Tomono T, Kuriyama K, et al. Laminin 511 is a target antigen in autoimmune pancreatitis. Sci Transl Med 2018;10.