Celiac disease: from food hypersensitivity to autoimmunity

When exogenous antigen drives harmful T cell-B cell interactions

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Celiac disease (CeD) is a common gastrointestinal disorder that can be diagnosed at all ages. The disease is related to intake of cereal gluten proteins, and the diagnostic scheme initially relied on elimination/provocation diets, similar to other food intolerances. Today, the guidelines for pediatric disease allows diagnosis to be made in a large fraction of patients solely based on the presence of high levels of autoantibodies to the autoantigen transglutaminase 2 (TG2). Autoimmunity is thus clearly implicated in CeD. Although autoimmune disorders are a collection of heterogeneous conditions, for which a single unifying mechanism is unlikely, some autoimmune diseases might share key pathogenic aspects with CeD. In this respect, the most provocative notion would be that immune reactions to exogenous antigens can drive autoimmune diseases other than CeD (1). Correspondingly, environmental factors such as viral or bacterial infections have commonly been associated with development of autoimmunity. Yet, CeD is the only autoimmune disease for which disease-driving T-cell epitopes originating from an exogenous antigen have been identified. Our recent observations suggest that interactions between gluten-specific CD4⁺ T cells and TG2-specific B cells are important for driving CeD pathogenesis. While T cell-B cell interactions have been implicated in other autoimmune diseases, the well-characterized target epitopes for both T cells and B cells in CeD offer unique possibilities to study pathogenic mechanisms involving disease-relevant antigen-specific immune cells.

A key role of CD4⁺ T cells in CeD is highlighted by an unusually strong HLA association with almost all patients expressing one of the HLA-DQ allotypes HLA-DQ2.5, HLA-DQ2.2 or HLA-DQ8. These HLA-DQ molecules harbor positively charged pockets in their peptide binding grooves making them particularly well suited to accommodate negatively charged gluten peptides. Negative charges are introduced via the catalytic activity of TG2 through a process known as deamidation. Notably, gluten-reactive CD4⁺ effector T cells in CeD patients are specific to epitopes that have been deamidated by TG2. Although the CD4⁺ T cells are not directly responsible for destruction of the gut epithelium, it is believed that these cells orchestrate the killing of enterocytes through secretion of pro-inflammatory cytokines, which act on the epithelium and cause activation of intraepithelial cytotoxic lymphocytes.

The dual role of TG2 in CeD as being both the target of autoantibodies and responsible for creating HLA-binding T-cell epitopes cannot be coincidental. The connection between activation of gluten-reactive T cells and production of autoantibodies against TG2 can be explained in the context of T cell-B cell collaboration. Further, accumulating evidence suggest that B cells can play an important role as antigen-presenting cells (APCs) for pathogenic T cells in CeD (and likely also in other autoimmune conditions). TG2-specific B cells can take up TG2-gluten enzyme-substrate complexes via B-cell receptor (BCR)-mediated endocytosis followed by presentation of deamidated gluten peptides on surface HLA molecules. This process directly links gluten uptake by TG2-specific B cells to deamidation and presentation of antigen to T cells, resulting in mutual activation of B and T cells, generation of TG2-specific autoantibodies and cytokine release (see figure).

Similar to the deamidation of gluten in CeD, post-translational modifications have been implicated in other autoimmune diseases. Yet, it has to be established whether T cells specific to modified (self)-peptides are controlling tissue destruction in patients or if post-translational modifications are merely a side-effect of autoimmune reactions. Post-translational modifications can potentially create neoepitopes that the immune system perceives as foreign, thereby facilitating escape of autoreactive cells from tolerance. The underlying mechanisms of neoepitope formation and their potential role in autoimmunity are poorly understood. Environmental factors such as smoking and viral infections are candidate triggers that may induce inflammatory tissue alterations, accompanied by dysregulation of post-translational modifications that could lead to autoimmunity. Other potential explanations for the connection between viral infections and different autoimmune disorders include molecular mimicry
between viral and self-antigens, leading to activation of cross-reactive T cells, and killing of host cells in infected tissues caused by virus-specific immune responses.

A prominent example of the connection between viral infections and autoimmunity is the association of Epstein-Barr virus (EBV) with development of multiple sclerosis (MS) (2). EBV persists in a latent state in memory B cells, which could serve as a permanent reservoir of viral antigens with the ability to stimulate immune cells. The disease is traditionally considered T cell-mediated. Nevertheless, B-cell depletion therapy with anti-CD20 antibodies has a beneficial effect and limits relapses in MS, suggesting that B cells play an important role. If persistent viral antigens are important drivers of autoimmunity, a possible explanation for the clinical observations is that EBV-infected B cells are depleted by anti-CD20 therapy, thereby effectively removing the driving antigen (3). In this case, B-cell depletion in MS would resemble the exclusion of gluten from the diet in CeD. In both cases, exogenous antigens could be targets of inappropriate immune responses causing damage to affected tissues.

Although intriguing, the idea that viral antigens can drive autoimmune disease has not been proven experimentally. Thus, the exact role of EBV in MS remains unclear, and it is not established if EBV-specific T cells are pathogenic or if EBV-infected B cells in the CNS give rise to inflammation. Importantly, B cells may have several other functions than being reservoirs of viral antigens, including antibody production, antigen presentation and secretion of cytokines. As anti-CD20 therapy does not deplete antibody-secreting cells, the clinical benefits of the treatment strongly suggest that B cells have antibody-independent functions in MS. It was recently shown that circulating B cells in MS patients are responsible for stimulating autoreactive, potentially pathogenic T cells that home to the brain (4). In addition, knockout of MHC class II expression specifically on B cells resulted in amelioration of symptoms in experimental autoimmune encephalomyelitis, the primary mouse model of MS (5). Similar findings were also obtained in a mouse model of systemic lupus erythematosus (SLE) (6), suggesting that antigen presentation by B cells to CD4+ T cells plays a key role in development of destructive immune reactions, at least in some autoimmune diseases.

We have recently obtained several lines of evidence pointing to interactions between antigen-specific B and T cells as essential for development of CeD. By using a combination of mass cytometry and RNA sequencing, it was shown that gluten-specific CD4+ T cells in blood and gut biopsies of CeD patients have a distinct phenotype with features resembling T follicular helper (Tfh) cells (7). Thus, the cells expressed high levels of IL-21 and CXCL13, which are important for activating and attracting B cells, but they lacked expression of the chemokine receptor CXCR5, which is required for homing to B-cell follicles. A similar expression profile was also observed in other autoimmune diseases and was previously reported for T cells found in synovial tissue of patients with rheumatoid arthritis (8). As CD4+ T cells rely on antigen-dependent interactions with B cells for Tfh-cell differentiation, the observed phenotype of pathogenic gluten-specific T cells indicates an important role of B cells as APCs in CeD. The lack of CXCR5 expression, however, suggests that T cell-B cell interactions do not take place in conventional germinal centers (GCs) but rather at extrafollicular sites. Curiously, activated B cells in SLE patients were also found to lack CXCR5, consistent with a non-GC-dependent origin (9). Extrafollicular activation of B cells in CeD is supported by the observation that TG2-specific antibodies rapidly disappear when patients start a gluten-free diet, indicating that GC-dependent long-lived plasma cells are not generated. Further, these antibodies contain relatively few mutations, consistent with B cells being activated extrafollicularly rather than in GCs (10). Not all types of autoantibodies, however, show this behavior. For example, rheumatoid arthritis autoantibodies against citrullinated proteins are heavily mutated, indicating a GC-dependent origin (11).

The potential role of B-lineage cells as APCs in CeD was substantiated through development of peptide-MHC-specific antibodies, which were used to identify cells presenting an immunodominant gluten
epitope on HLA-DQ2.5 in gut biopsies of CeD patients (12). Surprisingly, the most abundant cell type presenting antigen in the tissue was found to be plasma cells, which, in contrast to common belief, expressed surface MHC class II and co-stimulatory molecules for interaction with T cells. While the ability of plasma cells to stimulate T cells has yet to be demonstrated, support for plasma cells acting as APCs was obtained by RNA sequencing data demonstrating strong expression of invariant chain (CD74) and some expression of HLA-DQA1 and HLA-DQB1 (13). Importantly, IgA and IgM positive plasma cells express cell surface immunoglobulins (10), which serve as functional BCRs (14), allowing receptor-mediated uptake of cognate antigen. We observed that plasma cells presenting gluten antigen were enriched for TG2-specific cells which would be in line with the observation that TG2-specific B cells can present deamidated gluten peptide to T cells in a B-cell-epitope-dependent manner (15). Curiously, formation of antibodies targeting the preferred TG2 epitope correlated with onset of clinical disease, suggesting that efficient T cell-B cell collaboration is important for CeD development.

Identification of the primary target antigens in CeD has allowed the characterization of disease-relevant immune cells and antibodies isolated from patient samples. Recent studies focusing on different parts of the adaptive immune system suggest that interactions between antigen-specific T and B cells play a critical role in CeD pathogenesis. Importantly, clonally expanded B cells that efficiently take up antigen via receptor-mediated endocytosis are likely to be the main APCs for T cells both in CeD and in other autoimmune diseases. In many cases, however, the lack of well-defined antigens on both the T-cell and B-cell side complicates studies of the interactions between disease-relevant immune cells. It is important to note that autoimmune disorders are a heterogeneous group of diseases with widely different manifestations and pathogeneses. Nevertheless, we believe that the mechanisms that are beginning to unravel in CeD can be relevant for other autoimmune conditions. Likewise, we consider it possible that gluten is just one example of exogenous antigens that can drive autoimmunity in genetically susceptible individuals.

REFERENCES

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Path leading to (celiac) autoimmunity. Several of the steps involved in CeD pathogenesis have also been implicated in other autoimmune diseases. In CeD, however, we have more knowledge about the target antigens and the basis for the HLA association than in other cases. It is plausible that the mechanisms outlined for CeD are mirrored in some, but not all, other types of autoimmunity.