Pathogenic T-Cell Responses in Patients With Autoimmune Thrombocytopenic Purpura

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Summary: Autoimmune thrombocytopenic purpura (AITP) is a bleeding disorder in which autoantibodies are directed against an individual's own platelets, leading to enhanced clearance through Fc receptor (R)-mediated phagocytosis by macrophages residing in the reticuloendothelial system, particularly in the spleen. This review surveys the recent current literature and updates our understanding of the cell-mediated immunology of AITP. It will focus on the relationship between T-cell reactivities and cytokine profiles in patients with AITP. Understanding these cellular immune aspects of AITP is vital for developing antigen-specific immunotherapies to treat the disease.

Key Words: autoimmune thrombocytopenic purpura, platelets, HLA, T cells, autoimmunity, immunoregulation, cytokines

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Autoimmune thrombocytopenic purpura (AITP) is a bleeding disorder in which autoantibodies are directed against an individual's own platelets, leading to enhanced destruction through Fc receptor (R)-mediated phagocytosis by macrophages in the reticuloendothelial system.[1] Although the immunopathogenesis of the disease is antibody-mediated, the disease-producing autoantibodies are under the control of Th cells and the cytokines that they produce. These abnormal T-cell responses direct autoreactive B cells to differentiate and secrete IgG antiplatelet autoantibodies. Understanding the immune mechanisms controlling these cell-mediated mechanisms is vital for developing antigen-specific immunotherapies to treat the disease. Many studies on the cellular immunity of chronic AITP have been performed.[2–4] This paper will focus on the abnormal T-cell activation in chronic AITP, particularly related to T-cell specificity and cytokines and how these abnormalities may be related to autoantibody production.

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T-CELL ABNORMALITIES IN AITP

In 1991, we reported the novel finding that chronic AITP was associated with a $CD4^+$ T-helper cell defect in which peripheral blood T cells could secrete interleukin-2 upon stimulation with autologous platelets.[5] It suggested that chronic AITP may be the result of an abnormal Th cell defect that could direct autoreactive B cells to differentiate and secrete IgG autoantibodies. These interleukin-2 results were confirmed,[6] and subsequently it was reported that the abnormal T-cell responses observed in chronic AITP might be the result of a breakdown in T-cell tolerance against platelet autoantigens related to interleukin-2 expression.[7] In addition, it was demonstrated that there was an oligoclonal accumulation of $CD4^+$ Th cells in the peripheral blood of patients with AITP that frequently used Vβ3, 6, 10, and 13.1 to 14 genes for their T-cell receptors.[8] These findings suggested that antigen-driven T-cell clones accumulate in patients with chronic AITP and are related to the disease pathogenesis. However, the antigen specificities of the T cells, or their ability to stimulate autoreactive immunoglobulin production, are just beginning to be elucidated.

In 1998, Kuwana et al[9] demonstrated that T cells from patients with chronic AITP could proliferate in vitro to disulfide-reduced GPIIb/IIIa or the molecule's tryptic peptides. This suggested that autoreactive $CD4^+$ Th cells in chronic AITP need to recognize a modified GPIIb/IIIa molecule, implying that antigen-processing mechanisms within recipient APC may be required to present GPIIb/IIIa autoantigens in the context of self-HLA-DR molecules. More importantly, however, they showed that the modified GPIIb/IIIa proteins could also induce autoantibody formation in vitro.[9] Related to this, a recent study by Roark et al[10] demonstrated that platelet-specific autoantibodies derived from splenic B cells by Fab phage display had undergone somatic mutation, a genetic process strictly dependent on T-cell help. Taken together, these results are probably the first to link T-cell activation and B-cell autoantibody production in patients with AITP.

With respect to the platelet epitopes recognized by T cells in chronic AITP, Kuwana et al[11] also mapped the general

specificity of GPIIb/IIIa-reactive Th cells using six recombinant fragments encoding different portions of the GPIIbα and GPIIIa chains. They demonstrated that the T cells frequently recognized the amino terminal portion of the two GP chains (GPIIbα 18-259 and GPIIIa22-262) and that these molecules also stimulated the production of antiplatelet antibodies.[11] Since no Th cell reactivity against other portions of the two GP molecules was observed, they concluded that the amino terminal portion of GP IIb/IIIa was primarily responsible for the stimulation of autoreactive Th cells and subsequent autoantibody production.[11] In contrast, work from our laboratory examining the fine specificity of Th cell clones from the spleens of patients with chronic AITP revealed that at least one minimal peptide corresponding to amino acid residues 496–510 of the GPIIIa molecule was sufficient to stimulate T-cell interleukin-2 secretion.[12] These different results may reflect the heterogeneity of T-cell responsiveness in chronic AITP but more importantly suggest that T-cell epitopes can be found throughout the GPIIb/IIIa molecule. These results are a breakthrough because they will possibly allow for the design of antigen-specific therapies targeted at autoreactive T cells (e.g., modified and suppressive MHC binding peptides for inhibition of Th cells).

CYTOKINE DEFECTS IN AITP

Several cytokine abnormalities have been found to be associated with chronic AITP, and it appears that the disease is associated with a T_H0 cytokine pattern that is skewed toward a T_H1 activation pattern.[13–20] Other cytokines not classically associated with the T_H1/T_H2 paradigm have also been shown to be abnormal in chronic AITP and may play a role in the development of autoantibodies.[21–24] For example, Andersson et al[25] found that patients with chronic AITP in remission had significantly elevated levels of transforming growth factor (TGF)-β, and this was recently confirmed by Mouzaki et al.[20] TGF-β is a potent immunosuppressive cytokine and is thought to mark a T-cell subset now termed Th3 cells.[26] The results suggest that TGF-β may be part of a bystander immunosuppression in patients with AITP in remission.[25] Interestingly, Andersson et al[25] further suggested that elevating the in vivo levels of TGF-β by oral tolerance induction might be a viable immunotherapeutic approach. Taken together, these results suggest that modulating certain cytokine levels in patients with chronic AITP may be an effective therapy for raising platelet counts.

In summary, several T-cell activation and cytokine abnormalities have been identified in patients with chronic AITP. It appears that autoreactive T cells recognize several epitopes on the platelet surface and these events lead to abnormal activation and the generation of help for platelet-reactive B cells. Additionally, cytokine abnormalities associated with the T-cell activation may play an important role in regulating autoantibody production. However, although GPIIIa peptides have been identified that are recognized by autoreactive T cells, it is still not clear whether these peptides are the initiators of the autoimmune response. Further research is required to elucidate the general initiation mechanism of autoimmunity in chronic AITP and to develop more antigen-specific therapies to combat this disorder.

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