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Immunomodulation of immunothrombocytopenia

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ABSTRACT

Current treatment strategies in patients with immunothrombocytopenia (ITP) include immunosuppression and the stimulation of platelet production. Research over the last decade has emphasized the important role of Fc-receptors as key molecules responsible for autoantibody-mediated platelet depletion, allowing for a more specific targeting of this pathway instead of a generalized suppression of the immune system. This short review will discuss approaches aiming at interfering with key steps in this pathway, such as preventing the interaction of the platelet autoantibody immune complex with phagocytic cells, enhancing the clearance of autoantibodies, and interfering with or modulating the signaling pathways responsible for innate immune effector cell activation.

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The loss of humoral tolerance is one of the first steps preceding the development of many autoimmune diseases. In immunothrombocytopenia (ITP) the development of platelet specific antibodies results in the removal of opsonized platelets via innate immune effector cells [1]. Direct evidence that serum components are causative for the disease were provided by early serum transfer studies from ITP patients into healthy human volunteers and by preclinical mouse model systems documenting a rapid drop in platelet counts upon autoantibody infusion [2,3]. Studies in mice have emphasized that once the platelet autoantibody immune complex has been formed in vivo the binding to activating Fc γ -receptors (Fc γ R) expressed on innate immune effector cells is the key event responsible for platelet removal via phagocytosis [4,5]. Of note, several factors, such as the autoantibody IgG subclass or autoantibody glycosylation may directly impact the severity of the disease. Thus, platelet-specific antibodies of the mouse IgG2c/IgG2b or the human IgG1/IgG3 subclasses, which interact well with activating Fc γ Rs, have a much greater capacity to trigger platelet depletion than the same antibody with a mouse IgG3 or a human IgG2/IgG4 constant region [4,6]. Moreover, the affinity of the individual IgG subclass to bind to the inhibitory Fc γ RIIb, which is coexpressed with activating Fc γ Rs on mononuclear phagocytes, is another important parameter determining autoantibody activity (Fig. 1). Further along these lines, a low level of fucosylation in platelet specific alloantibodies targeting Hpa1, which will result in a strong increase in affinity for the human Fc γ RIIIa and Fc γ RIIIb, was shown to correlate with a higher disease activity [7]. Taken together, all of these studies emphasize that phagocytosis of autoantibody-platelet immune complexes via cellular Fc γ Rs is a key event in the pathology of ITP.

1. Therapeutic interventions targeting activating Fc γ Rs

Consistent with this central role of FcγRs in the disease process, many experimental strategies to treat ITP focus on blocking either the interaction of immune complexes with activating FcγRs or preventing the induction of phagocytosis (Fig. 1). The general validity of such an approach was demonstrated in humans thirty years ago by showing that a monoclonal antibody blocking the interaction of immune complexes with FcγRIIIa and FcγRIIIb was able to restore platelet counts in ITP patients [8]. Further confirming the important role of FcγRIIIa in this process, experimental ITP could be blocked with FcγRIII-specific antibodies in mice transgenic for human FcγRIIIa or with a cocktail of FcγR-specific antibodies in humanized mice [6,9]. Apart from using blocking antibodies, the induction of small immune complexes by injection of anti-D-specific antibodies in rhesus factor D-positive ITP patients or the use of multimeric IgG Fc-portions may achieve the same effect and compete for immune complex binding to FcγRs [10-12]. Instead of blocking immune complex binding to activating FcγRs another successful strategy to interfere with phagocyte activation is to block the signaling pathways induced after FcγR crosslinking (Fig. 1). Indeed, the inhibition of the spleen tyrosin kinase (SYK) was able to ameliorate ITP in mice and a human ITP patient cohort [13]. Apart from inhibiting activating FcγR-dependent signaling pathways, a depletion or inactivation of the monocyte and macrophage subsets responsible for platelet removal was another very efficient strategy to block autoantibody-dependent platelet depletion at least in preclinical model systems [5,6].

2. Therapeutic interventions targeting autoantibody half-life

The serum half-life of IgG antibodies is critically dependent on the neonatal Fc-receptor FcRn and mice deficient in this receptor are protected from autoantibody mediated induction of autoimmune diseases [14]. One promising strategy to achieve this therapeutically is the use of so

called Abdegs (antibodies that enhance IgG degradation), which are FcRn-specific monoclonal antibodies preventing serum IgG binding to FcRn. The potential of such an approach was demonstrated in murine models of inflammatory arthritis and experimental autoimmune encephalitis and may also be an interesting treatment option for acute or chronic ITP [15,16].

3. Immunomodulation via intravenous immunoglobulins

ITP was the first autoimmune disease in which the potent immunomodulatory activity of intravenous IgG preparations (IVIg) was demonstrated [17]. Later studies showed that the infusion of the isolated IVIg Fc-fragments was sufficient to raise platelet counts in ITP patients, as well as in a variety of mouse model systems, starting in depth investigations into the mechanism of this Fc fragment-dependent mechanism of IVIg activity in animal model systems [3]. Based on the large amounts of IgG infused at doses of 1–3 g/kg, the two most obvious mechanisms of IVIg-mediated suppression of ITP would be the interference with immune complex binding to activating FcγRs and/or the competition for access to the neonatal receptor. While there is compelling evidence that IVIg infusion indeed can reduce the level of autoantibodies [18], two more recent studies in classical and humanized mouse models demonstrated that IVIg ameliorates ITP independent of a mechanism involving FcRn at least during the acute phase of the disease [6,19]. With respect to a general block of activating FcγRs by IVIg, it was demonstrated that IVIg indeed binds to all types of immune cells upon infusion, but irrespective of the presence of activating or the inhibitory FcγRIIb [6,20]. Moreover, the depletion of B cells by cytotoxic antibodies (such as rituximab), which is also dependent on activating FcγRs, was not blocked by IVIg, arguing against a mechanism involving the general blockade of activating FcγRs [5,6]. In contrast, IVIg-mediated suppression of murine ITP was shown to be dependent on IgG glycovariants rich in terminal sialic acid residues, which was

confirmed more recently in a humanized mouse model system [6,21,22]. One current concept of how sialylated IgG glycovariants could mediate their activity suggests that they acquire the capacity to bind type II FcRs, such as the C-type lectin family members SignR1 (specific ICAM3 grabbing non-integrin-related 1), DC-SIGN (dendritic cell-specific intercellular adhesion molecule-3-grabbing non-integrin), or DCIR (dendritic cell inhibitory receptor), which may trigger an increase of inhibitory FcγRIIb expression on innate immune effector cells responsible for platelet phagocytosis [23]. Although it remains to be determined whether similar mechanisms account for IVIg activity in human ITP patients, the enhanced activity of highly sialylated IVIg preparations across a range of preclinical animal model systems, including two models of ITP, suggests that enhancing IVIg activity or generating recombinant IVIg products to treat ITP patients may become feasible in the near future [24].

4. Conclusion

Taken together, our more in depth understanding along which pathways autoantibodies mediate their activity may allow us to develop novel strategies to treat acute or chronic ITP. Compared to a generalized immunosuppression or invasive strategies such as splenectomy an advantage of these more targeted approaches may be that they have less unwanted side effects. Most forward, therapeutic concepts aiming at a modulation instead of a suppression of the immune system may help us to further optimize available therapies, such as the use of IVIg.

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Fig 1. Strategies to interfere with autoantibody-dependent platelet depletion. Shown is the pathway along which autoantibody–platelet immune complexes are removed from the circulation and how this process can be interfered with. Therapeutic options may include the inactivation or removal of phagocytes responsible for platelet phagocytosis, the modulation of Fc γ R expression as observed upon IVIg infusion, the blockade of activating Fc γ R signaling pathways, the block of immune complex binding to activating Fc γ Rs, and the enhancement of autoantibody clearance via blocking the neonatal FcRn. See text for further details.

Figure 1