Immune modulation with IVIg is commonly used for autoimmune and inflammatory diseases, such as Immune thrombocytopenic purpura, Kawasaki Syndrome, Dermatomyositis, Gullaine-Barre Syndrome and a large number of other neurological conditions. In all of these, treatment with IVIg uses doses that are 4-5 times higher than standard immune regulatory doses. This suggests that the regulatory component of IVIg is a minor fraction of the constituent IgG molecules.

IgG molecules are glycopolypeptides. They have various sites on both the Fc and the Fab region that can have a various sugar moieties as an integral part of the structure (Figure 1). These glycosylation sites alter the configuration of IgG and increase or decrease Fc receptor binding affinity, antigen trapping and other functions. One particular pattern of glycosylation, linkage of sialic acid residues to the Asn297 of the FC region, has been associated with regulatory responses. Seminal work by the Ravetch lab has identified this structure as crucial to the inhibition of murine models of serum induced arthritis and ITP. In their work, sialylated Fc sequences interact with SIGN-R1 or DC-SIGN on macrophages or dendritic cells, which induces downstream regulatory pathways involving IL-33 (Anthony et al., 2011; Anthony et al., 2008). Moreover, there are several clinical observations suggesting that the presence of sialylated IgG is associated with remissions or decreases in clinical symptoms. IgG sialylation may be a natural regulatory pathway for immune tolerance (Oefner et al., 2012; Orczyk-Pawilowicz et al., 2012; Otani et al., 2012).

Figure 1.

**IVIg in the Murine Model of Allergic Airways Disease**: Our laboratory has studied the immune regulatory role of IVIg in the murine model of allergic airways disease driven by ovalbumin (OVA) or Ragweed. Mice are sensitized intranasally to the antigen and then challenged on day 28-33. One day prior to challenge, IVIg 2 gm/kg or Human Serum Albumin control is infused i.p. Mice are sacrificed and airway hyperresponsiveness (AHR), pulmonary histology and regulatory T-cell numbers and function are assessed. The basic experimental protocol is outlined in Figure 2.
**IVIg induces regulatory T-cells:** Treatment of OVA-sensitized or ragweed sensitized mice with IVIg inhibited AHR, diminished pulmonary inflammation, reduced Th2 cytokines IL-4 and IL-13 and impaired splenocyte proliferation to the allergens. This was accompanied by a doubling of regulatory T-cells (Treg) in the pulmonary parenchyma. The Treg were antigen specific, suppressed antigen-driven lymphocyte proliferation and were enriched 10-fold in the peribronchial infiltrate compared to effector T-cells (Kaufman et al., 2011; Massoud et al., 2012).

**How does IVIg induce Regulatory T-cells?** We have identified specific event that lead to the induction of Treg by IVIg. IVIg interacts with pulmonary CD11c+ dendritic cells, leading to a modified, tolerogenic phenotype, with lower CD80 and CD86 than normally activated DC. Adoptive transfer of pulmonary CD11c+ DC from mice treated with IVIG completely abrogates AHR and increases Treg to the same extent as IVIg. A) PBS; B) OVA, C) IVIG treated DC, D) OVA+IVIG treated DC, E) Treg enumeration in lung digests. Both DCs exposed to IVIg alone or IVIg + OVA induced Treg and inhibited AHR.

**Sialylated IgG reproduces the action of IVIg:** We purified sialic acid fractions from IVIG, which is a 3-5% constituent of normal IVIg. We also deglycosylated the remaining IVIg fraction to ensure that it had no sialic acid residues. Sialylated IgG (SA-IVIG), at a 10 fold lower dose than IVIg, inhibited all AHR (A) and pulmonary inflammation and induced Treg (B) to the same extent as IVIg.
IVIg. Deglycosylated IgG (nonSA-IVIg). It also inhibited production of both IL-4 and IL-17 from pulmonary mononuclear cells (C-F).

Thus, the immune regulatory function of IVIg in this Th2 mediated inflammatory model indicates that IgG functions as a bridge between the innate and adaptive immune systems. Sialylated residues on IgG bind to lectin receptors and induce a tolerogenic phenotype in the DC (Figure 5). In the presence of antigen, T-effector cells home to the DC-T-cell interface, leading to induction of Treg. We are currently exploring the mechanism by which DC will induce Treg; this may include up-regulation of inhibitory phosphatases, alteration in expression of co-stimulatory molecules, production of tolerogenic cytokines such as IL-10. IgG may also be internalized and processed by inhibitory C-type lectin receptors on DC followed by presentation of peptide epitopes to the TCR. Further understanding of this IgG mediated bridge and its regulatory activities will lead to improved molecular design for novel therapies for autoimmune and inflammatory diseases.
References


