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IgG-effector functions: "The Good, The Bad and The Ugly"

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ABSTRACT

IgG-antibodies are potent and versatile mediators of host protection. They elicit their biological effects through specific interaction of the Fc-part with complement, specific cellular receptors, or both. Several factors should be taken into consideration when analyzing the nature and intensity of the immunological response elicited via IgG-effector functions, especially for the family of IgG-Fc receptors (FcγRs) exclusively expressed on immune cells. These include the various classes of leukocyte FcγR, expressed variably on different immune cells, each with distinct affinity for every IgG subclass, as well as genetic FcγR-polymorphisms affecting expression and affinity for IgG. Furthermore, various aspects of the IgG itself are also crucial for the outcome of the biological response. These include endogenously encoded IgG-polymorphisms, such as IgG3 polymorphisms, and post-transcriptional IgG-modifications, in particular IgG-Fc-glycosylation, affecting IgG effector functions through modified binding affinity to FcγR. These latter aspects concerning the variability in IgG3 on its half-life and placental transport and the clinical consequences of altered IgG-quality through glycosylation, will be the focus of this review.

1. Introduction

Immunoglobulins are crucial mediators of immunological protection against invading pathogens. They exert their biological effects through several effector systems. For IgG-antibodies the most important effector functions are mediated through complement and/or the Fcγ-receptors (FcγRs), a family of cell surface receptors on leukocytes that specifically bind to the Fc portion of IgG-antibodies, bridging the adaptive and innate immune systems [1]. In humans these receptors are termed FcγRIa (CD64a), FcγRIb (CD64b), FcγRIIa (CD32a), FcγRIIb (CD32b), FcγRIIc (CD32c), FcγRIIIa (CD16a), and FcγRIIIb (CD16b). With the exception of FcγRIIb, all the FcγR mediate activating functions (e.g. phagocytosis, antibody-dependent cell-mediated cytotoxicity (ADCC), release of inflammatory mediators and superoxide radicals) after FcγR-crosslinking by IgG-opsonized targets, while FcγRIIb mainly exhibits an inhibitory function, inhibiting the function of the activating FcγR. Exceptions include induction of anti-tumor responses of agonistic CD40 antibodies through FcγRIIb [2,3]. These receptors also have a varying distribution on cells, with FcγRI mostly restricted to macrophages, monocytes and activated granulocytes, FcγRIIa having a widest range of expression on myeloid cells, FcγRIIIb only present on granulocytes (neutrophils, basophils and possibly on eosinophils) [4] and FcγRIIIa on NK cells, macrophages and a subpopulation of monocytes, particularly in the spleen. Although it is clear that the inhibitory FcγRIIb is expressed on B cells and macrophages, the expression on other cells is less certain and may vary between individuals [1].

Two other receptors for IgG are also expressed ubiquitously in almost all cells; the neonatal Fc-receptor (FcRn), a homologue of the major histocompatibility complex (MHC) class I molecules, and the tripartite motif-containing protein 21 (TRIM21), an E3 ubiquitin-protein ligase. Both receptors are expressed inside cells, particularly of myeloid origin. FcRn is found within vacuoles and tubules transporting FcRn along with its cargo, binding both endocytosed IgG and albumin in a pH-dependent manner, recycling both ligands and thereby extending their half-life [5]. FcRn-mediated transport across cellular (e.g. epithelial, endothelial, syncytiotrophoblast) barriers is responsible for the IgG-transmission across mucosal barriers and from mother to child. In addition, FcRn participates in the process of IgG-mediated phagocytosis in myeloid cells in a pH-dependent manner independent of the IgG recycling [6]. Downstream of this pathway FcRn can also deliver immune complex-bound antigens into the antigen presenting pathway, boosting secondary responses [7,8].

TRIM21 on the other hand, is expressed in the cytosol, binding IgG with an even higher affinity than the other human IgG-receptors. It recognizes opsonized non-enveloped viruses,

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intracellular bacteria and targets them for ubiquitination and proteasome degradation [9,10]. Subsequently, TRIM21 activation has also been found to stimulate immune signaling via transcription factor pathways (NF-κB, AP-1, IRF3, IRF5, and IRF7), resulting in downstream secretion of pro-inflammatory cytokines, modulation of natural killer stress ligands and inducing an antiviral state [11]. Antibody-engagement through the surface-exposed FcγRs can also trigger intracellular signaling cascades (induced proximally and sequentially through Src- and Syk-kinases) [12]. This can result in immediate degranulation of the cells, with concomitant release of inflammatory mediators and initiation of cellular responses, such as phagocytosis (by phagocytes such as monocytes, macrophages, neutrophils) or ADCC (for example by NK-cells or all myeloid cells). Under normal conditions these processes are utilized beneficially by the host to eliminate invading pathogens. However, these processes can also cause adverse reactions for the host in numerous auto-or alloimmune diseases [13,14], for instance by antibody-mediated platelet destruction (immune thrombocytopenia; ITP, or fetal or neonatal alloimmune thrombocytopenia; FNAIT) [15] or antibody-mediated red blood cell destruction in hemolytic disease of the fetus or the newborn (HDFN) [16].

Besides different cellular distribution of FcγRs, their affinity and specificity for the different IgG subclasses (IgG1, IgG2, IgG3, IgG4) also varies considerably, reflecting the distinct biological effects of each subclass in triggering different cell types (reviewed by Hogarth et al. [1]). For instance, IgG2 only binds FcγRIIa, with slightly lower affinity for FcγRIIIa, while IgG1 and IgG3 bind all of the FcγRs. This enables IgG1 and IgG3 to co-cross link all of the FcγR (for instance FcγRIIa and FcγRIIIb), while IgG2 targets only the FcγRIIa on neutrophils and possibly FcγRIIIa on NK cells, macrophages and some macrophages [17]. The affinity for the receptor types differ greatly between FcγRI and the rest, but is also depending on the IgG subclass, as IgG1, IgG3 and IgG4 bind FcγRI in the nanomolar range, while IgG1 and IgG3 (and IgG4, but with slightly less affinity) bind FcγRII in the micromolar range [1,17].

In last two decades we have become increasingly aware of the importance of how these binding affinities are affected by genetic polymorphisms within the FcγR family. A good example is FcγRIIa, which can have either arginine (R) or histidine (H) at position 131. The former binds human IgG2 with lower affinity than the latter, while the opposite is true for human IgG4 and mouse IgG1 [1,17].

Another important polymorphism in the FcγR-family can be found within FcγRIIIa, which contains either a valine (V) or phenylalanine (F) at position 158, with the V-variant showing stronger binding affinity for all the IgG subclasses [1,17–19]. Consequently, the V-variant is associated with a higher incidence of ITP [20,21], as well as with faster clearance of Rhesus D (RhD) expressing erythrocytes in the presence of anti-RhD IgG [22]. Conversely, the lower affinity FcγR variants are associated with increased susceptibility to infectious diseases [23].

Within FcγRIIb, the inhibitory FcγR, additional polymorphism exist within the trans-membrane region, with either a isoleucine (I) or threonine (T) at position 232 that determines their efficacy to co-aggregate with the activating FcγR into lipid rafts (T232 being less efficient) and thereby their capacity to down regulate the activating responses [24,25].

An added layer of variability exist within the FcγR locus on the long arm of chromosome one, which entails copy number polymorphisms of the FcγRIII genes as well as the FcγRIIc gene, which is a pseudogene in approximately 80% of healthy individuals. Those individuals expressing a functional copy of IIc have been found to be at increased risk for acquiring ITP (present in 34.4% of ITP patients), indicating that this polymorphism may also be important negative predictor for other autoimmune diseases [20]. These polymorphisms and their importance for infectious diseases and auto- and allo- immunity have been extensively investigated and reviewed before [23,26–31]. However, polymorphisms within the IgG, both endogenously encoded as well as through posttranscriptional modification, can also greatly affect immune responses, as will be described further below.

2. Functional polymorphism within IgG3

Of the four IgG subclasses, human IgG3 stands out compared to the other IgG subclasses – also to other species – with up to four exons encoding for the hinge, extending its reach to up to 62 amino acids (compared with 15 for IgG1, 12 for IgG2 and IgG4) (Fig. 1). IgG3 has higher affinity for complement component C1q and FcγRs, providing it with its relatively stronger effector functions [1,17,32,33]. However, it has a relatively short half-life (7 days vs. 21 days for the other subclasses), is generally known to have poor placental transport and to have impaired transport to mucosal surfaces [34], suggesting IgG3 not to be compatible for FcRn-mediated transport [35–37]. However, we recently identified that IgG3 has a short half-life because of a single amino acid change at position 435; a position bearing a histidine in IgG in all known mammalian species. This histidine is normally neutral at physiological pH of 7.4, but becomes protonated in acidic lysosomes (pH < 6.5) after uptake by a pino- or endocytotic event, acquiring a positive charge. This protonation is required for binding to FcRn, triggering transport of IgG away from the developing lysosome back to the cell surface, where it is released again at neutral pH. Unlike the other IgG subclasses, IgG3 generally bears an arginine at position 435 and is thus always positively charged at all physiological pH. Despite this difference, IgG3 can be transported normally by FcRn – but only when IgG3 is present alone. This has been observed both *in vitro* using transwell systems [33] and *in vivo* as IgG3 has a normal half-life in SCID mice [38] as opposed to wild type mice [39]. Using *in vitro* transwell assays, IgG3 was not transported effectively together with other IgG subclasses but was degraded instead. However, the transport was rescued after replacing arginine at position 435 with a histidine [33]. Conversely, IgG1 bearing R435 was rescued and transported normally by FcRn *in vitro*, but only when present alone, as after mixing with IgG3 H435 – or other IgG WT subclasses – both transport and recovery of both IgG1 was dramatically decreased. *In vivo* experiments in mice confirmed these findings, indicating the decreased transport and enhanced loss of IgG3 to be solely due to the arginine at position 435 [33].

In contrast to the other IgG subclasses, IgG3 is highly polymorphic in humans, consisting of at least 17 distinct variants, many of which are known to give rise to allo-specific antibodies (allotypes) [33,40]. Three distinct genetic IgG3 variants, previously identified by two different allo-epitopes (G3m15 and G3m16), do actually naturally contain histidine at position 435. They are uncommon in Europe (~1%) but much more prevalent in Asia (10–50%) [40,41]. We recently found that these H435-IgG3 allotypes do have an extended half-life in humans that is comparable to the other subclasses (Fig. 1A) [33]. In agreement with this, and with FcRn being the only receptor involved in transplacental transport, G3m16 IgG3-antibodies found in pregnant women were readily transported across the placenta by FcRn [41] as well as H435-containing but not R435-containing IgG3 in *ex vivo* perfused human placenta (Fig. 1A) [42]. As IgG3-antibodies are frequently found in pregnancy related cellular allo-immune pathologies, and generally mediate stronger effector functions than IgG1, fetuses and newborns of G3m16 (and the less common G4m15) positive mothers might therefore be more susceptible for adverse HDFN or FNAIT, in which the red blood cells or platelets, respectively, of the fetus or newborn are destroyed [43,44].

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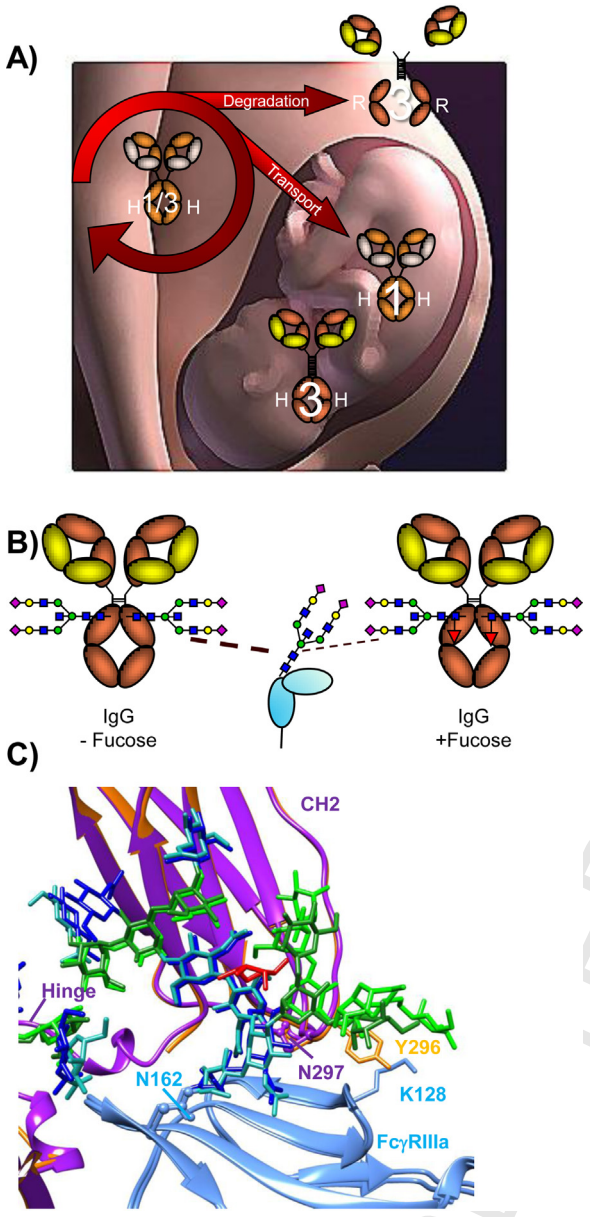

Q4 Fig. 1. IgG pharmacology and modulation of effector functions. (A) Due to FcRn-mediated recycling, all human IgG subclasses, except some allotypes of IgG3, have an extended half-life of three weeks whereas similar size proteins and IgG3 containing arginine (R) at position 435, have a half-life of only one week and are degraded. This is because the amino acid at position 435 resides at a key position for FcRn-binding, causing IgG3 to lose binding to FcRn in competition with the other subclasses – all bearing a histidine (H) at position 435. The same holds true for FcRn-mediated transport of IgG across the placenta, which is ineffective for the classical form of IgG3 with R435. However, H435-containing allotypes of IgG3 do exist at high frequency in some populations (allotypes G3m15 and G3m16, also termed G3m(s,t)), and these have normal half-life of three weeks and placental transport. As IgG3 has the highest affinity for FcγR and C1q, this may be beneficial for the young, providing it with enhanced immunity for the first weeks after birth. However, they are also of potential higher risk for allo-immune mediated diseases that occur frequently in pregnancy such as anemia and thrombocytopenia. 1:IgG1, 3:IgG3. (B) The approximate locations of the glycan attached to N297 in the IgG-Fc, and N162 for FcγRIIIa and IIIb affecting binding between IgG and FcγR. Although normal variations in the Fc-glycan have been shown to slightly affect binding to all FcγR, the IgG-Fc glycan is absolutely required for binding. Only the presense or absence of the core-Fc-fucose (red triangle, right) strongly affects binding to FcγR, but only to the FcγRIII family, bearing N162 that interacts directly with the IgG-Fc glycan. These allotypic variations and posttranslational modifications, and how they add an altered layer of complexity and conrol over the biology of human IgG, is discussed in more details in the main text. The thicker dashed line indicates a stronger binding affinity of nonfucosylated IgG with the human FcγRIII family members, compared to the thinner dashed line respresenting the weaker binding affinity of fucosylated IgG. (C) Three-dimensional alignment of FcγRIIIa-Fc crystal structures, crystallized with- (accession number 3SGJ, Fc-heavy chains purple, darker glycan colors) and without- (accession number 3SGK, Fc-heavy chains orange, lighter glycan colors) core Fc-fucose, based on structures published by Ferrara et al. [47]. FcγRIIIa is depicted in light blue for both structures. The glycans are colored according to standard schemes with N-acetylglucosamine in blue, mannoses in green, and fucose in red. Without fucose, the FcγRIIIa glycan, in particular the mannoses of FcγRIII (right), structures shifts with increased hydrogen bond formation, but also clamping of the Y296 in the Fc onto K128 on FcγRIIIa [47], which may however also be influenced by the extended number of mannose residues found in that structure. (For interpretation of the references to color in figure legend, the reader is referred to the web version of the article.)

3. Regulation of IgG effector functions on the B-cell level through Fc-glycosylation

IgG antibodies are glycoproteins containing a highly-conserved branched sugar moiety attached to the asparagine (ASN)297 part of the antibody-Fc domain. This glycan is essential for the maintenance of a functional structure and is required for binding of IgG with FcγR [45–47]. Normally, this glycan consists of a core structures of N-acetylglucosamine and mannoses but with variable levels of galactose, sialic acid, bisecting N-acetylglucosamine (GlcNAc) and core fucose. Variation in this composition influences antibody affinity to FcγR and thus antibody effector activity. Increased sialic acid content of IgG-Fc has received a great amount of attention over the past years due their protective effect in IVIG, as the presence of sialic acid in IVIG has been described to enable IgG-binding to the mouse lectin SIGN-R1 (also known as DC-SIGN – dendritic cell-specific ICAM3-grabbing non-integrin – in humans), upregulating a cytokine cascade initiated by IL-33 and eventually leading to enhanced myeloid expression of FcγRIIb via IL-4 [48–50]. The enrichment of sialic acid has been demonstrated to have an approximate 7-fold reduced binding affinity of murine IgG to FcγRIII in mice [48]. However, these findings in mice have recently been challenged by a study of Rheumatoid Arthritis, a disease well known for frequent remission during pregnancy. Although this remission seemed to go hand in hand with both increased galactosylation and sialylation, a further independent analysis of di-galactosylated (G2) IgG indicated that the association with sialylation to be an epiphenomenon due to its requirement for previous galactosylation. In fact, while G2- IgG was associated with remission, di-sialylated G2-IgG had an opposing effect apparent by the minor but positive association with disease activity [51].

Of the variable amount of glycans present in the IgG-Fc, the removal of the IgG-core fucose has by far the greatest effect on binding to FcγRs. A lack of core fucose has been demonstrated to result in an up to 50-fold stronger binding affinity to FcγRIIIa [45,52] and FcγRIIIb [52] but not to the other FcγRs. This restriction is caused by a conserved N-glycan in human FcγRIIIa and FcγRIIIb at position 162 (N162), which interacts with the Fc but also the Fc-glycan. This configuration is greatly affected by the presence or absence of the core-fucose within the IgG-Fc (Fig. 1B) [47,53]. The enhanced binding to the GPI-linked FcγRIIIb has been shown to result in enhanced neutrophil phagocytosis of rituximab-opsonized $CD20^+$ lymphoma cells [52] and of platelets [54]. In addition, the enhanced binding to FcγRIIIa resulted in enhanced ADCC on mononuclear cells [45,55–59], but remarkably not through neutrophil FcγRIIIb [60].

Until recently, the importance of the core fucosylation of the IgG-Fc has only received attention because of the possibilities to produce more efficacious therapeutic antibodies, like rituximab (reviewed by Yamane-Ohnuki et al. [61]). This is because core fucosylation was not known to be variable in humans until recently [54,62,63]. Indeed, ~94% of all naturally occurring IgG-derived Fc-glycopeptides in humans have the core-fucose attached [51,64]. Theoretically, this means that ~99.7% of all IgG

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Table 1
Overview of IgG-glycosylation patterns and their effects in disease-settings, observed in literature to date.

IgG-glycan	Disease setting	Affected-IgG	Effect of IgG-glycan
Galactosylation	Rheumatoid arthritis (RA)	Total IgG	Increased [66–68]; increased in remission of RA in pregnancy [51,66,67,69], decreased during relapse of RA in pregnancy [69], inversely related with N-acetylglucosamine [70,71]
	Juvenile onset chronic arthritis	Total IgG	Inversely related with N-acetylglucosamine [70,71]
	Crohn's disease	Total IgG	Inversely related with N-acetylglucosamine [71]
	Primary osteoarthritis	Total IgG	Decreased [72]
	Osteoarthritis	Total IgG	Parallel increase with N-acetylglucosamine [71]
	Sjögren's Syndrome	Total IgG	Decreased [70]. Parallel increase with N-acetylglucosamine [71]
	Tuberculosis	Total IgG	Decreased [73]
	Ovarian cancer	Total IgG	Decreased [74]
	Fetal or neonatal alloimmune thrombocytopenia (FNAIT)	Anti-platelet IgG	Increased [54,62]
	Refractory thrombocytopenia (RT)	Anti-HLA IgG	Increased [54]
	HIV	Anti-HIV in elite controllers	Decreased [63]
	Gastric cancer	Total IgG	Decreased [75]
	Lambert–Eaton myasthenic syndrome	IgG1-anti presynaptic voltage gated Ca^{2+}-channels	Decreased [76]
Sialylation	RA	Total IgG	Increased in remission of RA in pregnancy [51,67]
	Gastric cancer	Total IgG	Decreased [75]
	FNAIT	Anti-platelet IgG	Increased [54,62]
	HIV	Anti-HIV in elite controllers	Decreased [63]
Fucose	FNAIT	Anti-platelet IgG	Decreased [54,62]
	HIV	Anti-HIV in elite controllers	Decreased [63]
	Gastric cancer (stage II–III)	Total IgG	Increased [75]
	Infertile leukospermic patients	Seminal IgG	Decreased compared to normal, fertile normozoospermic patients [77]
Bisecting N-acetylglucosamine	RT	Anti-HLA IgG	Decreased [54]
	Gastric cancer (stage II–III)	Total IgG	Decreased [75]
	Lambert–Eaton myasthenic syndrome (<50 years)	IgG1-anti presynaptic voltage gated Ca^{2+}-channels	Increased [76]

contain either one (combination of a heavy chain without and with fucose on either side or $2 \times 0.06 \times 0.94 \times 100\% = 11.3\%$) or two ($0.94 \times 0.94 \times 100\% = 88.4\%$) core-Fc fucoses, with only 0.3% of circulating IgG completely devoid of core-fucose. However, individual differences do exist. This was recently exemplified in a genome-wide association study, identifying the loci containing fucosyltransferase 8 (FUT8) and the transcription regulator Ikaros to influence these levels – loci that in turn were previously reported to be associated with other diseases such as systemic lupus erythematosus, Crohn's disease, and multiple sclerosis [65]. In addition, antigen-specific IgG-fucosylation can be skewed in certain immune responses. In a previous pilot-study we observed that the anti-platelet specific antibodies, which mediated alloimmune reactions against platelets in pregnancies, displayed a decreased fucosylation [62]. In a follow-up study, we evaluated the glycosylation patterns of a large cohort of FNAIT -serum samples containing anti-platelet specific antibodies (anti-human platelet antigen (HPA)-1a antibodies) in pregnancy and found a clear skewing toward decreased Fc-fucosylation in the majority of anti-platelet-, but not total-IgG [54]. Most patients were identified because of their adverse clinical symptoms, but those identified without clinical symptoms had normal fucosylation of their anti-platelet IgG, while those most severely affected had fucosylation down to 10%. This indicated the immune setting during the pregnancy, the nature of the antigen, or both, to play a role in steering the quality of the antibodies. In this case with an adverse effect for the patient, as the lack of core-fucose in the anti-platelet IgG resulted in increased platelet destruction due to enhanced binding to either FcγRIIIa and/or FcγRIIIb. Furthermore, a significant relationship was observed between the level of anti-platelet IgG-core fucosylation and neonatal platelet numbers in FNAIT, as well as bleeding tendency, with decreased core-fucosylation of the pathogenic anti-HPA1a-antibodies being associated with decreased number of platelets and increased clinical severity [54]. Conversely, a lack of Fc-fucose in IgG can theoretically be highly beneficial for responses against infectious diseases, as has also recently been demonstrated for HIV-specific antibodies in elite controllers – apparently a signature for improved HIV-neutralization and clearance [63]. The remarkable variation in IgG-Fc glycosylation patterns in various disease settings and their subsequent effects reported in the literature are summarized in Table 1.

In conclusion, more variables besides FcγR polymorphisms, IgG titer, and subclass responses affect IgG-effector functions and clinical outcomes. Strong effector functions and high binding affinities (FcγR profiles, FcγRIIc-ORF, a high number of FcγRIII and IIc gene copies, IgG3-G3m15/16, non-fucosylated IgG-variants) can be associated with a "good" outcome in terms of protection against infectious diseases or antibody-mediated therapies. However, these can also turn out "bad" or even "ugly" in allo- and auto-immune mediated diseases, for instance resulting in intracranial hemorrhages in FNAIT. All these factors should be taken into consideration when looking into the biological response, both from a diagnostic as well as a therapeutic perspective.

References

[1] Hogarth PM, Pietersz GA. Fc receptor-targeted therapies for the treatment of inflammation, cancer and beyond. Nat Rev Drug Discov 2012;11:311–31.
[2] Li F, Ravetch JV. Inhibitory Fcγ receptor engagement drives adjuvant and anti-tumor activities of agonistic CD40 antibodies. Science 2011;333:1030–4.
[3] White AL, Chan HT, Roghanian A, French RR, Mockridge CI, Tutt AL, et al. Interaction with FcγRIIB is critical for the agonistic activity of anti-CD40 monoclonal antibody. J Immunol 2011;187:1754–63.
[4] Meknache N, Jonsson F, Laurent J, Guinnepain MT, Daeron M. Human basophils express the glycosylphosphatidylinositol-anchored low-affinity IgG receptor FcγRIIIB (CD16B). J Immunol 2009;182:2542–50.
[5] Vaccaro C, Zhou J, Ober RJ, Ward ES. Engineering the Fc region of immunoglobulin G to modulate in vivo antibody levels. Nat Biotechnol 2005;23:1283–8.

Please cite this article in press as: Kapur R, et al. IgG-effector functions: "The Good, The Bad and The Ugly". Immunol Lett (2014), http://dx.doi.org/10.1016/j.imlet.2014.01.015

[6] Vidarsson G, Stemerding AM, Stapleton NM, Spliethoff SE, Janssen H, Rebers FE, et al. FcRn: an IgG receptor on phagocytes with a novel role in phagocytosis. Blood 2006;108:3573–9.
[7] Baker K, Qiao SW, Kuo TT, Aveson VG, Platzer B, Andersen JT, et al. Neonatal Fc receptor for IgG (FcRn) regulates cross-presentation of IgG immune complexes by CD8-CD11b+ dendritic cells. Proc Natl Acad Sci USA 2011;108:9927–32.
[8] Qiao SW, Kobayashi K, Johansen FE, Sollid LM, Andersen JT, Milford E, et al. Dependence of antibody-mediated presentation of antigen on FcRn. Proc Natl Acad Sci USA 2008;105:9337–42.
[9] Mallery DL, McEwan WA, Bidgood SR, Towers GJ, Johnson CM, James LC. Antibodies mediate intracellular immunity through tripartite motif-containing 21 (TRIM21). Proc Natl Acad Sci USA 2010;107:19985–90.
[10] Hauler F, Mallery DL, McEwan WA, Bidgood SR, James LC. AAA ATPase p97/VCP is essential for TRIM21-mediated virus neutralization. Proc Natl Acad Sci USA 2012;109:19733–8.
[11] McEwan WA, Tam JC, Watkinson RE, Bidgood SR, Mallery DL, James LC. Intracellular antibody-bound pathogens stimulate immune signaling via the Fc receptor TRIM21. Nat Immunol 2013;14:327–36.
[12] Bezbradica JS, Medzhitov R. Role of ITAM signaling module in signal integration. Curr Opin Immunol 2012;24:58–66.
[13] Schwab I, Nimmerjahn F. Intravenous immunoglobulin therapy: how does IgG modulate the immune system. Nat Rev Immunol 2013;13:176–89.
[14] Karsten CM, Kohl J. The immunoglobulin, IgG Fc receptor and complement triangle in autoimmune diseases. Immunobiology 2012;217:1067–79.
[15] Semple JW, Italiano Jr JE, Freedman J. Platelets and the immune continuum. Nat Rev Immunol 2011;11:264–74.
[16] Urbaniak SJ, Greiss MA. RhD haemolytic disease of the fetus and the newborn. Blood Rev 2000;14:44–61.
[17] Bruhns P, Iannascoli B, England P, Mancardi DA, Fernandez N, Jorieux S, et al. Specificity and affinity of human Fcγ receptors and their polymorphic variants for human IgG subclasses. Blood 2009;113:3716–25.
[18] de Haas M, Koene HR, Kleijer M, de VE, Simsek S, van Tol MJ, et al. A triallelic Fcγ receptor type IIIA polymorphism influences the binding of human IgG by NK cell FcγRIIIa. J Immunol 1996;156:2948–55.
[19] Koene HR, Kleijer M, Algra J, Roos D, von dem Borne AE, de HM. FcγRIIIa-158V/F polymorphism influences the binding of IgG by natural killer cell FcγRIIIa, independently of the FcγRIIIa-48L/R/H phenotype. Blood 1997;90:1109–14.
[20] Breunis WB, van ME, Bruin M, Geissler J, de BM, Peters M, et al. Copy number variation of the activating FCGR2C gene predisposes to idiopathic thrombocytopenic purpura. Blood 2008;111:1029–38.
[21] Bruin M, Bierings M, Uiterwaal C, Revesz T, Bode L, Wiesman ME, et al. Platelet count, previous infection and FCGR2B genotype predict development of chronic disease in newly diagnosed idiopathic thrombocytopenia in childhood: results of a prospective study. Br J Haematol 2004;127:561–7.
[22] Miescher S, Spycher MO, Amstutz H, de HM, Kleijer M, Kalus UJ, et al. A single recombinant anti-RhD IgG prevents RhD immunization: association of RhD-positive red blood cell clearance rate with polymorphisms in the FcγRIIA and FcγIIIA genes. Blood 2004;103:4028–35.
[23] van Sorge NM, van Der Pol WL, van de Winkel JG. FcγR polymorphisms: implications for function, disease susceptibility and immunotherapy. Tissue Antigens 2003;61:189–202.
[24] Vidarsson G, van de Winkel JG. Fc receptor and complement receptor-mediated phagocytosis in host defence. Curr Opin Infect Dis 1998;11:271–8.
[25] Aman MJ, Tosello-Trampont AC, Ravichandran K. FcγRIIB1/SHIP-mediated inhibitory signaling in B cells involves lipid rafts. J Biol Chem 2001;276:46371–8.
[26] Tsuchiya N, Kyogoku C, Miyashita R, Kuroki K. Diversity of human immune system multigene families and its implication in the genetic background of rheumatic diseases. Curr Med Chem 2007;14:431–9.
[27] Cantsilieris S, White SJ. Correlating multiallelic copy number polymorphisms with disease susceptibility. Hum Mutat 2013;34:1–13.
[28] Bournazos S, Woof JM, Hart SP, Dransfield I. Functional and clinical consequences of Fc receptor polymorphic and copy number variants. Clin Exp Immunol 2009;157:244–54.
[29] Fanciulli M, Vyse TJ, Aitman TJ. Copy number variation of Fcγ receptor genes and disease predisposition. Cytogenet Genome Res 2008;123:161–8.
[30] de Haas M. IgG-Fc receptors and the clinical relevance of their polymorphisms. Wien Klin Wochenschr 2001;113:825–31.
[31] Rascu A, Repp R, Westerdaal NA, Kalden JR, van de Winkel JG. Clinical relevance of Fcγ receptor polymorphisms. Ann NY Acad Sci 1997;815:282–95.
[32] Vidarsson G, van Der Pol WL, van Den Elsen JM, Vile H, Jansen M, Duijs J, et al. Activity of human IgG and IgA subclasses in immune defense against Neisseria meningitidis serogroup B. J Immunol 2001;166:6250–6.
[33] Stapleton NM, Andersen JT, Stemerding AM, Bjarnarson SP, Verheul RC, Gerritsen J, et al. Competition for FcRn-mediated transport gives rise to short half-life of human IgG3 and offers therapeutic potential. Nat Commun 2011;2:599.
[34] Raux M, Finkielsztejn L, Salmon-Ceron D, Bouchez H, Excler JL, Dulioust E, et al. IgG subclass distribution in serum and various mucosal fluids of HIV type 1-infected subjects. AIDS Res Hum Retroviruses 2000;16:583–94.
[35] Stussi G, Huggel K, Lutz HU, Schanz U, Rieben R, Seebach JD. Isotype-specific detection of ABO blood group antibodies using a novel flow cytometric method. Br J Haematol 2005;130:954–63.
[36] Einarsdottir HK, Selman MH, Kapur R, Scherjon S, Koeleman CA, Deelder AM, et al. Comparison of the Fc glycosylation of fetal and maternal immunoglobulin G. Glycoconj J 2013;30:147–57.
[37] Malek A, Sager R, Schneider H. Maternal-fetal transport of immunoglobulin G and its subclasses during the third trimester of human pregnancy. Am J Reprod Immunol 1994;32:8–14.
[38] Hassan MS, Abedi-Valugerdi M, Lefranc G, Hammarstrom L, Smith CI. Biological half-life of normal and truncated human IgG3 in scid mice. Eur J Immunol 1991;21:1319–22.
[39] Kim JK, Firan M, Radu CG, Kim CH, Ghetie V, Ward ES. Mapping the site on human IgG for binding of the MHC class I-related receptor, FcRn. Eur J Immunol 1999;29:2819–25.
[40] Lefranc MP, Lefranc G. Human Gm, Km, and Am allotypes and their molecular characterization: a remarkable demonstration of polymorphism. Methods Mol Biol 2012;882:635–80.
[41] Einarsdottir H, Ji Y, Visser R, Mo C, Luo G, Scherjon S, et al. H435-containing immunoglobulin G3 allotypes are transported efficiently across the human placenta: implications for alloantibody-mediated diseases of the newborn. Transfusion 2013 [Epub ahead of print].
[42] Mathiesen L, Nielsen LK, Andersen JT, Grevys A, Sandlie I, Michaelsen TE, et al. Maternofetal transplacental transport of recombinant IgG antibodies lacking effector functions. Blood 2013;122:1174–81.
[43] Brouwers HA, Overbeeke MA, Ouwehand WH, Keuning K, van E, van Leeuwen IEF, et al. Maternal antibodies against fetal blood group antigens A or B: lytic activity of IgG subclasses in monocyte-driven cytotoxicity and correlation with ABO haemolytic disease of the newborn. Br J Haematol 1988;70:465–9.
[44] Pollock JM, Bowman JM. Anti-Rh(D) IgG subclasses and severity of Rh hemolytic disease of the newborn. Vox Sang 1990;59:176–9.
[45] Shields RL, Lai J, Keck R, O'Connell LY, Hong K, Meng YG, et al. Lack of fucose on human IgG1 N-linked oligosaccharide improves binding to human FcγRIII and antibody-dependent cellular toxicity. J Biol Chem 2002;277:26733–40.
[46] Sondermann P, Huber R, Oosthuizen V, Jacob U. The 3.2-Å crystal structure of the human IgG1 Fc fragment-FcγRIII complex. Nature 2000;406:267–73.
[47] Ferrara C, Grau S, Jager C, Sondermann P, Brunker P, Waldhauer I, et al. Unique carbohydrate–carbohydrate interactions are required for high affinity binding between FcγRIII and antibodies lacking core fucose. Proc Natl Acad Sci USA 2011;108:12669–74.
[48] Kaneko Y, Nimmerjahn F, Ravetch JV. Anti-inflammatory activity of immunoglobulin G resulting from Fc sialylation. Science 2006;313:670–3.
[49] Anthony RM, Kobayashi T, Wermeling F, Ravetch JV. Intravenous gammaglobulin suppresses inflammation through a novel T(H)2 pathway. Nature 2011;475:110–3.
[50] Sondermann P, Pincetic A, Maamary J, Lammens K, Ravetch JV. General mechanism for modulating immunoglobulin effector function. Proc Natl Acad Sci USA 2013;110:9868–72.
[51] Bondt A, Selman MH, Deelder AM, Hazes JM, Willemsen SP, Wuhrer M, et al. Association between galactosylation of immunoglobulin G and improvement of rheumatoid arthritis during pregnancy is independent of sialylation. J Proteome Res 2013;12:4522–31.
[52] Shibata-Koyama M, Iida S, Misaka H, Mori K, Yano K, Shitara K, et al. Nonfucosylated rituximab potentiates human neutrophil phagocytosis through its high binding for FcγRIIIb and MHC class II expression on the phagocytotic neutrophils. Exp Hematol 2009;37:309–21.
[53] Mizushima T, Yagi H, Takemoto E, Shibata-Koyama M, Isoda Y, Iida S, et al. Structural basis for improved efficacy of therapeutic antibodies on defucosylation of their Fc glycans. Genes Cells 2011;16:1071–80.
[54] Kapur R, Kustiawan I, Vestrheim A, Koelman CA, Visser R, Einarsdottir HK, et al. A prominent lack of IgG1 Fc-fucosylation of platelet-alloantibodies in pregnancy. Blood 2013;123(4):471–80, Q2 http://dx.doi.org/10.1182/blood-2013-09-527978.
[55] Junttila TT, Parsons K, Olsson C, Lu Y, Xin Y, Theriault J, et al. Superior in vivo efficacy of afucosylated trastuzumab in the treatment of HER2-amplified breast cancer. Cancer Res 2010;70:4481–9.
[56] Masuda K, Kubota T, Kaneko E, Iida S, Wakitani M, Kobayashi-Natsume Y, et al. Enhanced binding affinity for FcγRIIIa of fucose-negative antibody is sufficient to induce maximal antibody-dependent cellular cytotoxicity. Mol Immunol 2007;44:3122–31.
[57] Suzuki E, Niwa R, Saji S, Muta M, Hirose M, Iida S, et al. A nonfucosylated anti-HER2 antibody augments antibody-dependent cellular cytotoxicity in breast cancer patients. Clin Cancer Res 2007;13:1875–82.
[58] Niwa R, Shoji-Hosaka E, Sakurada M, Shinkawa T, Uchida K, Nakamura K, et al. Defucosylated chimeric anti-CC chemokine receptor 4 IgG1 with enhanced antibody-dependent cellular cytotoxicity shows potent therapeutic activity to T-cell leukemia and lymphoma. Cancer Res 2004;64:2127–33.
[59] Niwa R, Natsume A, Uehara A, Wakitani M, Iida S, Uchida K, et al. IgG subclass-independent improvement of antibody-dependent cellular cytotoxicity by fucose removal from Asn297-linked oligosaccharides. J Immunol Methods 2005;306:151–60.
[60] Peipp M, Lammerts van Bueren JJ, Schneider-Merck T, Bleeker WW, Dechant M, Beyer T, et al. Antibody fucosylation differentially impacts cytotoxicity mediated by NK and PMN effector cells. Blood 2008;112:2390–9.
[61] Yamane-Ohnuki N, Satoh M. Production of therapeutic antibodies with controlled fucosylation. mAbs 2009;1:230–6.
[62] Wuhrer M, Porcelijn L, Kapur R, Koeleman CA, Deelder A, de HM, et al. Regulated glycosylation patterns of IgG during alloimmune responses against human platelet antigens. J Proteome Res 2009;8:450–6.

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[63] Ackerman ME, Crispin M, Yu X, Baruah K, Boesch AW, Harvey DJ, et al. Natural variation in Fc glycosylation of HIV-specific antibodies impacts antiviral activity. J Clin Invest 2013;123:2183–92.
[64] Bakovic MP, Selman MH, Hoffmann M, Rudan I, Campbell H, Deelder AM, et al. High-throughput IgG Fc N-glycosylation profiling by mass spectrometry of glycopeptides. J Proteome Res 2013;12:821–31.
[65] Lauc G, Huffman JE, Pucic M, Zgaga L, Adamczyk B, Muzinic A, et al. Loci associated with N-glycosylation of human immunoglobulin G show pleiotropy with autoimmune diseases and haematological cancers. PLoS Genet 2013;9:e1003225.
[66] Rook GA, Steele J, Brealey R, Whyte A, Isenberg D, Sumar N, et al. Changes in IgG glycoform levels are associated with remission of arthritis during pregnancy. J Autoimmun 1991;4:779–94.
[67] van de Geijn FE, Wuhrer M, Selman MH, Willemsen SP, de Man YA, Deelder AM, et al. Immunoglobulin G galactosylation and sialylation are associated with pregnancy-induced improvement of rheumatoid arthritis and the postpartum flare: results from a large prospective cohort study. Arthritis Res Ther 2009;11:R193.
[68] Selman MH, Derks RJ, Bondt A, Palmblad M, Schoenmaker B, Koeleman CA, et al. Fc specific IgG glycosylation profiling by robust nano-reverse phase HPLC–MS using a sheath-flow ESI sprayer interface. J Proteomics 2012;75:1318–29.
[69] Alavi A, Arden N, Spector TD, Axford JS. Immunoglobulin G glycosylation and clinical outcome in rheumatoid arthritis during pregnancy. J Rheumatol 2000;27:1379–85.
[70] Bond A, Alavi A, Axford JS, Youinou P, Hay FC. The relationship between exposed galactose and N-acetylglucosamine residues on IgG in rheumatoid arthritis (RA), juvenile chronic arthritis (JCA) and Sjogren's syndrome (SS). Clin Exp Immunol 1996;105:99–103.
[71] Bond A, Alavi A, Axford JS, Bourke BE, Bruckner FE, Kerr MA, et al. A detailed lectin analysis of IgG glycosylation, demonstrating disease specific changes in terminal galactose and N-acetylglucosamine. J Autoimmun 1997;10:77–85.
[72] Parekh RB, Dwek RA, Sutton BJ, Fernandes DL, Leung A, Stanworth D, et al. Association of rheumatoid arthritis and primary osteoarthritis with changes in the glycosylation pattern of total serum IgG. Nature 1985;316:452–7.
[73] Parekh R, Isenberg D, Rook G, Roitt I, Dwek R, Rademacher T. A comparative analysis of disease-associated changes in the galactosylation of serum IgG. J Autoimmun 1989;2:101–14.
[74] Saldova R, Royle L, Radcliffe CM, Abd Hamid UM, Evans R, Arnold JN, et al. Ovarian cancer is associated with changes in glycosylation in both acute-phase proteins and IgG. Glycobiology 2007;17:1344–56.
[75] Kodar K, Stadlmann J, Klaamas K, Sergeyev B, Kurtenkov O. Immunoglobulin G Fc N-glycan profiling in patients with gastric cancer by LC–ESI–MS: relation to tumor progression and survival. Glycoconj J 2012;29:57–66.
[76] Selman MH, Niks EH, Titulaer MJ, Verschuuren JJ, Wuhrer M, Deelder AM. IgG fc N-glycosylation changes in Lambert–Eaton myasthenic syndrome and myasthenia gravis. J Proteome Res 2011;10:143–52.
[77] Kratz EM, Ferens-Sieczkowska M, Faundez R, Katnik-Prastowska I. Changes in fucosylation of human seminal IgG and secretory component of IgA in leukocytospermic patients. Glycoconj J 2014;31(1):51–60. Q3

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