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Immunoglobulin G in IgE-mediated allergy and allergen-specific immunotherapy

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Summary

Allergen-specific IgG antibodies play a significant role in allergen-specific tolerance, either naturally induced or generated by specific immunotherapy. Nevertheless, the underlying mechanisms are still debated, and allergen-specific IgG determinations are not recommended as a diagnostic tool in IgE-mediated allergy. This review summarizes the latest findings on the immunological and diagnostic role of IgG antibodies in respiratory and food allergies, and during allergen-specific immunotherapy.

Allergen-specific IgG antibodies in respiratory allergy

Type-1 allergy is mainly based on the production and effects of IgE antibodies (1); however, other immunoglobulin classes such as IgG (2,3) with its subclasses and IgA (4,5) have also gained considerable attention in allergy research. Allergen-specific IgG antibodies appear increasingly within the course of and after immunotherapy (6,7) and gained the attribute of “blocking antibodies” (8,9) against antigens involved in IgE-mediated allergy. On the other hand, allergen-specific IgG may also play a role in the occurrence of anaphylactic events (10). This broadens the line-up for possible functions of IgG and its subclasses in type-1 allergy.

The evidence that IgE deficient mice could still develop anaphylaxis (11,12) introduced IgG in the group of allergy promoting factors. Recent studies on mouse models confirmed the role of the high-affinity human IgG receptors FcγRIIA (CD32) and FcγRI (CD64) in IgG-mediated allergic inflammation and ana-

phylaxis (10,13). As the binding affinity of IgG antibodies to the antigen is much lower than that of IgE (14), their blocking function seems to be based mainly on the sheer quantity of antibodies able to bind the allergen before it reaches the surface of the mast cells. This higher concentration of antibodies is among other factors promoted by the significantly longer half-life of IgG compared to IgE (15,16). Although various studies point out a contributing effect of allergen-specific IgG in the pathogenesis of allergic disease (17,18), the overall results remain controversial and vary according to the antigen and exposure levels (19). For example, the appearance and protective role of IgG antibodies in cat allergy may be related to the dose of exposure to the major allergen Fel d 1 (20). By contrast, although the correlation between cat ownership and higher IgG levels, especially of the subclass IgG4, could also be shown in a study on 412 Swedish children (21), no significant protective effect of these antibodies could be demonstrated. Finally, among 227

children aged 12 to 14 years (22), those exposed to higher antigen concentrations showed a higher risk of being sensitized to house dust mite or cat (OR 4.0, 99% CI 1.49-10.00). Within this group, only high concentrations of IgG antibodies to Fel d 1 correlated with a decreased prevalence of sensitization. Other studies among children reported a relation of lower IgG4 levels with positive skin prick test (SPT) (5), an increased risk of rhinoconjunctivitis (23), and a modifying effect of IgG (not IgG4) on the association between cat-specific IgE and childhood wheezing, with decreasing symptoms related to higher IgG levels (24). This goes along with the results of the German Multicentre Allergy Study (MAS), which reported a low risk of wheezing in children with high IgG levels to cat (25). However, these specific IgG levels were only protective in the absence of IgE and not in children with IgE-mediated sensitization. Serum levels of mouse related IgG or IgG4 were initially suggested as markers for clinical tolerance among 23 laboratory animal workers (26), but following tests among an increased number of probands (n = 110) could not confirm this evidence (27). Various studies on the above-mentioned antigens (28,29) and on *Malassezia* (30) or *Alternaria* (31), report on parallel trends in the appearance of IgE, IgG and IgG4 antibodies, suggesting a complementary role. In addition to these findings, Jenmalm et al. repeatedly discovered a strong correlation between elevated IgG4 serum levels and atopic sensitization to birch, egg and cat allergens in childhood (32,33).

Allergen-specific IgG antibodies in food allergy

Food-specific IgG antibodies can be found in most children at the age of three months, independently from their atopic status (34). In a trial on 89 food-allergic children with eczema, the levels of serum and salivary antibodies were examined as potential biomarkers predicting safe reintroduction of previously eliminated foods (35). Interestingly, high pre-diet serum IgG4 levels and IgG4/IgE ratios correlated to established allergen-specific tolerance. The importance of allergen-specific IgE/IgG4 ratios in tolerance induction has been repeatedly underlined (36-38) and recently confirmed in 107 egg-allergic children (39) undergoing an oral food challenge with baked egg. While children with a low IgE/IgG4 ratio to ovomucoid and/or ovalbumin were able to tolerate baked egg, higher levels of this ratio were related to a positive challenge and even anaphylactic reactions. Then, a low IgE/IgG4 to ovalbumin and ovomucoid has been suggested as a marker for tolerance to baked egg in egg-allergic children. Similarly, tolerance was associated in cow's milk-allergic children with a decrease in epitope binding by IgE in combination with an opposed increase in IgG4 binding to the corresponding epitopes (40,41). Among 95 infants with eczema, low serum IgG4 levels to β -lactoglobulin differentiated those with a clear from those with only suspected cow's milk allergy (4). Ac-

ordingly, various clinical trials showed that the efficacy of oral immunotherapy for different antigens, such as peanut (42,43), milk (44) and egg (45), is related to a significant increase of IgG and IgG4 concentrations. By contrast, some studies reported elevated IgG levels in IgE sensitized children to peanut, milk and egg (46,47), warning that the role of IgG in food allergy or tolerance has not been fully determined yet.

Allergen-specific IgG antibodies in drug allergy

Although food allergens are more frequently the cause of anaphylactic events, hypersensitivity to drugs can also lead to severe and potentially life threatening allergic reactions (48). Especially adverse reactions to penicillins are reported by patients and can be observed in daily clinical practice, which made their immunological base a matter of interest already in the 1990s. After reporting on diverse isotypes and specificities of IgG and IgE antibodies to penicillins at individual level (49), a Spanish research group evaluated the role of IgG antibodies in immediate allergic reactions to different determinants of benzylpenicillin, amoxicillin, and ampicillin (50). The study on 59 patients could not confirm its hypothesis of a protective role of allergen-specific IgG in the development of anaphylaxis. A later study on 249 patients with penicillin allergy (51) reported on higher IgG levels specific to various allergen components in allergic subjects, also in patients with negative skin tests but typical symptoms. These findings underline the role of allergen-specific IgG antibodies in the development of drug hypersensitivity, but further research on this topic and on reactions to other drugs is still needed.

The role of IgG antibodies in allergen-specific immunotherapy

To date, allergen-specific immunotherapy (SIT) is the only recognized disease modifying and clinical effective treatment for allergic rhinitis and allergic asthma, as well as IgE-mediated venom allergy. Unlike symptomatic pharmacotherapy for allergy, SIT can reduce both, symptoms and use of medication, prevent sensitization to new allergens, and induce prolonged allergen-specific tolerance after discontinuation of the treatment (52-54). However, the immunological mechanisms underlying SIT still remain incompletely understood. Successful SIT has been associated with several immunological changes, including reduction in mucosal recruitment of basophils and eosinophils, suppression of peripheral Th2 effector cells, immune deviation of cytokine responses from an allergic Th2 to a Th1 pattern, and induction of regulatory T-cells, which suppress the specific Th2 response to allergens through cell-to-cell contact and release of immunosuppressive cytokines (such as TGF- β and interleukin IL-10) (55,56). In addition, there is increasing evidence that clinically effective SIT is associated with an increase in allergen-specific IgG antibodies, particularly the IgG4 subclass. Several studies, involving either

sublingual immunotherapy (SLIT) with aeroallergens (57-60) or subcutaneous immunotherapy (SCIT) with aeroallergens (60-64) and hymenoptera venoms (65,66), have documented an induction of allergen-specific IgG and IgG4 in sera. Furthermore, the duration of clinical reactivity (67) or tolerance (68) has also been shown to be related to the level of specific IgG4. It should be stressed that an increase in IgG and IgG4 antibodies has been related not only to a “naturally” acquired food tolerance, but also to the development of tolerance induced by oral immunotherapy (OIT or SOTI) (69-73). Additionally, the specific IgG4/IgG1 ratio as well as the IgG4/IgE ratio have been proposed in some studies as predictive parameters of a beneficial response to SIT (74, 75). However, there is no consensus on using these antibodies as biomarkers to predict the clinical response to SIT (76). It is still a matter of debate whether the efficacy of SIT could depend on allergen-specific IgG induction. According to the same studies, the induction of allergen-specific IgG antibodies during SIT is mainly an “epiphenomenon”, reflecting the development of favorable conditions for tolerance such as the appearance of IL-10 producing regulatory T-cells, which also increase IgG4 production (77,78). Furthermore, a link between increased allergen-specific IgG4 titers and favorable response to SIT has not always been found, particularly with hymenoptera venoms immunotherapy (79).

A possible explanation for the lack of correlation in some studies is that successful SIT seems to induce changes not only in allergen-specific-IgG concentrations, but also in their biological activity, which require qualitative rather than quantitative assays for the detection (2). SIT-induced IgG4 antibodies have been shown to act as “blocking antibodies”, which prevent both immediate and late-phase responses by inhibiting IgE-mediated basophils and mast cells degranulation, and allergen presentation to T-cells (3,7-9,80,81). Noteworthy, these blocking activities do not solely depend on allergen-specific-IgG concentrations. Changes in the antigenic reactivity and specificity of SIT-induced IgG antibodies have been reported (82). Moreover, it has been shown that long-term clinical tolerance after discontinuation of SIT is associated with persistence of the IgG4-associated blocking activities (particularly after SIT with aeroallergens). In contrast, SIT-induced allergen-specific IgG4 levels tend to decrease after withdrawal of immunotherapy (83,84). Therefore, the measurement of the IgG inhibitory activities with functional assays, rather than IgG serum titers with quantitative assays, seems a more reliable biomarker to predict the clinical response to SIT (76,85).

In light of these evidences, an effective role of allergen-specific IgG antibodies in the induction and maintenance of the beneficial effects of SIT has been reconsidered. In a very recent experimental study in mouse models, the potential therapeutic and preventive effects of passive immunization with allergen-specific IgG antibodies on allergy have been tested, showing promising results (86).

Lack of diagnostic value of allergen-specific IgG in routine clinical practice

Especially in food allergy, an accurate diagnostic procedure is fundamental to avoid potentially life-threatening reactions (87,88); hence, the clinical history, a controlled food challenge or skin prick testing and serum IgE determinations should be used as a combination of diagnostic tools. When the diagnosis of IgE-mediated allergy cannot be established and serologically confirmed, it is not rare that patients seek for alternative test methods to meet their expectations for results. These procedures often include the determination of allergen-specific IgG antibodies and subclasses offered by commercial laboratories and pharmacies. These measures are not only expensive for the patient and a burden for any health system but do also lack sufficient scientific background. In 2008, a task force of the European Academy of Allergology and Clinical Immunology (89) comprehensively discussed the use of IgG4 testing against foods in allergy, and got to the clear conclusion that it cannot be recommended as a diagnostic tool. This opinion has been also expressed by the American Academy of Allergy Asthma and Immunology (90). Since then, various studies have been conducted to further investigate the role of IgG antibodies in allergy diagnosis. Among 150 hen's egg-allergic children, neither IgG nor IgG4 measurements added any valuable information to the diagnostic procedure of hen's egg allergy (91), thus supporting the position that neither IgG nor IgG4 assays should be included in the diagnostic routine for allergy testing. Similarly, no diagnostic value of IgG and IgA antibodies could be found for cow's milk allergic patients (92). This unanimity against IgG antibodies and subclasses in allergy diagnosis does not rule out the hypothesis of potential other roles of this serological parameter such as e.g. a predictive value. In the early 1990s, an observational study from the Netherlands showed that high IgG1 levels to food allergens were related to the development of allergy to airborne allergens later in life (93). About 50% of the children with a high IgG1 anti-food score developed an IgE response to grass pollen and/or cat dander, which suggested a predictive value of IgG antibodies to food allergens. Although a cross-sectional approach by the same group could confirm this trend (94), a final prospective study on 397 children was not able to reproduce the results and described the determination of allergen-specific IgG levels as not very useful for the identification of patients at risk in clinical practice (95). A randomized double-blind allergy prevention trial from Finland also reported on an increased risk of egg allergy in relation to elevated serum IgA and IgG levels to ovalbumin, but could not confirm this trend for other allergens or as a valid predictive tool (96). Thus, although IgG antibodies, especially the subclass IgG4, are certainly of importance in allergy and tolerance induction, they are nowadays still not of value for clinical practice.

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