

Allergen-specific immunotherapy

Regulatory T cells or allergen-specific IgG?

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IgE-mediated allergies, such as allergic rhino-conjunctivitis and asthma, have become highly prevalent and affect up to one third of the population in westernized countries.¹⁻⁴ The gold standard treatment is subcutaneous allergen-specific immunotherapy (SIT), the administration of gradually increasing quantities of an allergen vaccine to an allergic subject, which is an effective treatment for allergic rhinitis, conjunctivitis, asthma and allergic reactions from stinging insects.⁵⁻⁷ SIT has a high overall success rate of around 80% in hay fever and more than 90% in allergy to insect venom. Also, SIT confers long term benefit even after discontinuation,^{8,9} interrupts expansion of allergic sensitization¹⁰ and progression from allergic rhinitis to asthma.¹¹ From a medical perspective, SIT is therefore superior to symptomatic treatment. The disadvantages of SIT are, however, its long duration over 3–5 years, and the requirement for up to 80 allergen injections. Also, allergen injections frequently cause allergic side effects. The latter range from swelling and itching at the site of injection to urticaria or asthma due to systemic allergen distribution, but also include a risk of anaphylaxis and fatalities.¹²⁻¹⁴ Because of these short comings, less than 5% of allergy sufferers chose to undergo SIT.

The current form of SIT was introduced already nearly a century ago in 1911 by Noon and Freeman,^{15,16} and one has to wonder why, with all the progress in science and immunology, SIT could not significantly be improved over the last century. The main reason for this lack of progress may be due to the fact, that

the immunological mechanism of SIT, and especially the mechanism ultimately responsible for amelioration of allergy symptoms, is still unclear.

Back in 1911, when SIT was introduced, the assumed immunological mechanism was easier to understand. At that time it was thought that the mucosal symptoms of hay fever were caused by “pollen toxins”, and consequently Noon and Freeman aimed at vaccinating against these toxins by “prophylactic inoculation against hay fever”. The understanding of SIT became more complicated after the discovery of IgE as the mediator of the allergic symptoms in the mid 1960ies.^{17,18} As knowledge of T cell function was still largely lacking, induction of allergen-specific blocking IgG antibodies were thought to be the protective mechanism of SIT.¹⁹⁻²¹ With the better understanding of T cell function, first Th1 cells counteracting the Th2 allergic response became the postulated mechanism of SIT.²²⁻²⁴ Today, as immunological interest focuses on regulatory T cells (Tregs), the latter are thought to be largely responsible for the effects of SIT.²⁵⁻²⁷

In the following we will briefly and provocatively discuss whether the concept of Noon and Freeman has become obsolete in the advent of Tregs. It should be pointed out that we are not discussing whether or not any or all the above immunomodulation happens, but just which mechanism in the end protects against allergen induced symptoms. It is well possible that Tregs play an important role in the production of allergen specific IgG4, so that both Tregs and IgGs are important, but here we will discuss how

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protection against an acute allergen challenge occurs.

Mechanism of Desensitization: Regulatory T Cells

SIT has been found to induce allergen-specific Treg, and these are thought to exert the therapeutic effect via suppression of Th2 cells,²⁸ suppression of allergen-specific IgE and/or upregulation of IgG4 and IgA.²⁹ Suppression of mast cells and eosinophils has also been postulated to be an important function of Tregs.³⁰ The problem with assuming Tregs as the key effector cells in SIT, however, lies within the fact that allergic reactions, such as rhinoconjunctivitis upon allergen exposure, occur within seconds to minutes. IgE molecules on mast cells within tissues are cross-linked by the allergen, causing the release of a vast number of mediators, including histamine. These factors induce the local symptoms of swelling and itching. How could regulatory T cells interfere with this process? With all likelihood, they cannot. For regulatory T cells (Tregs) to become activated, they need to interact with specific peptides presented on MHC class II molecules on dendritic cells or other antigen presenting cells (APCs), such as macrophages or B cells. Hence, allergens need to be taken up APCs, processed and presented to Tregs. This may activate Tregs and allow them to suppress activation of mast cells. Either by release of immune modulating cytokines or, even more time consuming, by direct suppression of mast cells by cellular contact. From everything we know about antigen-processing and T cell activation, this process is far too slow to occur within seconds to minutes. Hence, allergen-specific suppression of mast cell activation by Tregs seems largely impossible for kinetic reasons. Nevertheless, it may be conceivable that Tregs do not suppress mast cell activation in an allergen-specific fashion. Indeed, Tregs may foster a “suppressive” environment within tissues, inhibiting mast cell activation.^{30,31} However, this process would not be allergen-specific and this notion is in direct contradiction with clinical experience that desensitization is allergen-specific: desensitization against grass pollen does not simultaneously

desensitization against cat or house dust-mite allergy; therefore the name “allergen-specific immunotherapy or SIT”.

Asthmatic reactions may occur in two phases, an early phase triggered within minutes upon allergen-exposure and a late phase, peaking around 12–24 hours later. The first phase is mainly mediated by IgE antibodies while the second phase is mediated by Th2 cells. As discussed above for the local IgE-mediated so called type I allergic reaction, it seems highly unlikely that the early phase of the asthmatic reaction is modulated by Tregs. This is different for the late, Th2-dependent asthmatic reaction. In this case, allergen-specific Tregs may indeed be able to suppress the activation of Th2 cells, by the release of cytokines such as IL-10 or TGF β . Hence, induction of Tregs may be an important component for a therapy of asthma.

Mechanism of Desensitization: Antibodies

As discussed above, Tregs are unlikely to be responsible for the reduction of rhinoconjunctivitis symptoms upon allergen-specific immunotherapy. Allergen-specific antibodies induced by the therapy may, however, be able to inhibit mast cell activation. Doubt has been cast on the original assumption that antibodies are the protective effector mechanism in SIT since it is difficult to find a correlation of IgG4 antibody levels and clinical outcome of SIT.^{32,33} However, it is difficult to find this correlation because (i) in clinical routine IgG4 are measured against allergen extract, e.g., a grass pollen extract, which includes numerous proteins and allergens against which the patient is not allergic. Thus, an increase in anti-allergen-IgG4 will be masked by an increase of IgG4 against the many proteins the patient is not allergic against (ii) the affinity of IgGs is usually not measured,^{34,35} (iii) the blocking activity of IgG³⁶ may not correlate simply with the levels of IgG,^{32,33} and (iv) the IgG/IgE ratio in the patient is important.³⁷

We have recently shown in mice that allergen-specific IgG antibodies induced by immunotherapy were able to fully block systemic as well as local mast cell activation.³⁸ Adoptive transfer of

allergen-specific IgG into allergic mice was similarly able to alleviate allergic symptoms. In marked contrast, depletion of Tregs had no influence on the efficacy of the allergen-specific immunotherapy. Two mechanisms accounted for IgG-induced inhibition of mast cell activation. On the one hand, polyclonal IgG antibodies were able to “neutralize” allergens and prevent them from engaging to mast cell bound IgE. In addition and almost equally efficient, IgG antibodies inhibited mast cell activation via engagement of the inhibitory Fc γ RIIb expressed by the mast cell. There was an interesting and presumably important difference between local allergen exposure in the skin versus systemic allergen challenge. In the former case, neutralization by polyclonal IgG was sufficient for blocking the allergic reaction. In contrast, polyclonal IgG antibodies were not able to block the anaphylactic reaction in allergic mice exposed to an intravenous allergen challenge in the absence of Fc γ RIIb. Hence, while in the skin, both allergen-neutralization and engagement of the inhibitory Fc γ RIIb are able to block mast cell activation, systemic anaphylactic reactions are inefficiently blocked in the absence of inhibitory receptors. Whether this is due to the kinetics of allergen-binding by specific antibodies or a more prominent involvement of basophils in systemic allergic reactions remains to be established.

Conclusion

Allergen-specific immunotherapy remains the only disease modifying long-term therapy for allergies. While a reduction in rhinoconjunctivitis symptoms is best documented, there is accumulating evidence that the progression of allergy into asthma may also be inhibited by the therapy. Available evidence indicates that induction of Tregs is unlikely responsible for alleviation of allergic symptoms while a beneficial role of Tregs in blocking the progression to asthma seems plausible but remains to be established. In contrast to an important role for Tregs in the therapy of rhinoconjunctivitis, there is strong evidence that allergen-specific IgG antibodies are able to block both local as well as systemic allergic responses. Thus, the

original concept of Noon and Freeman, the inventors of SIT, that “prophylactic inoculation” with pollen extract were a vaccine inducing antibodies against “pollen toxins”, may after all not be that far off. Also other toxins exert their effects by blocking or over stimulating physiological pathways or receptors, and it is not at all wrong to say that “pollen toxins” irritate mucosal surfaces, the mechanism being crosslinking of IgE. Also, it is now becoming clearer how the IgG induced by SIT protect against allergy symptoms. For blocking of the systemic allergic response, engagement of the inhibitory FcγRIIb is required while simple allergen neutralization by specific IgG antibodies is sufficient to halt the local allergen-specific response. These considerations have important consequences for the design of novel allergen-specific immunotherapies, since they high-light an important role for antibodies. This indicates that immunotherapy should use immunogens that carry at least some native allergen-epitopes and that an approach based on short peptides may be of limited value, since such peptides will fail to induce IgG antibodies cross-reacting with the native allergen.

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