

Efficacy and Safety of Allergen-Specific Immunotherapy in Rhinitis, Rhinoconjunctivitis, and Bee/Wasp Venom Allergies

G. Senti

P. Johansen

J. Martinez Gomez

B. M. Prinz Varicka

T. M. Kündig

Department of Dermatology, University Hospital of Zurich,
Zurich, Switzerland

The prevalence of allergic diseases, such as rhinoconjunctivitis, is increasing worldwide, particularly in Westernized countries, where more than 30% of the population is affected. Insect venom allergy is also very common, affecting up to 5% of the population. Allergen-specific immunotherapy is the only immunomodulatory treatment that may alter the natural course of allergic disease, for example by preventing the development of asthma in rhinitic patients. Nonetheless, the risk-benefit ratio for subcutaneous immunotherapy has changed little from when it was first developed a century ago. However, the rapid evolution of new developments, including new methods of administration and new forms of antigen to stimulate the immune system, now offers improvements in both the safety and the efficacy of specific immunotherapy. These developments include the sublingual administration of the relevant antigens, which has a superior safety profile than the original subcutaneous route. This may enable higher dosages to be used over shorter treatment periods, with a lower risk of anaphylactic reactions. Improvements in the purity, specificity, and immunogenicity of the antigens, often as a result of advances in biotechnology, coupled with the development of new adjuvants, may further increase the efficacy of this form of treatment. This review describes and discusses these new developments in the context of the many recent advances in our understanding of the mechanisms by which immunotherapy appears to act.

Keywords: bee/wasp venom, allergies, efficacy, safety, allergoids, peptides, adjuvant, sublingual therapy, corticosteroids, antihistamines, IgE, IgG, anaphylaxis, anti-IgE antibody, recombinant allergens

Address correspondence to PD Dr. T. Kündig, Department of Dermatology, Gloriastr. 31, 8091 Zurich, Switzerland. E-mail: thomas.kuendig@usz.ch

INTRODUCTION

The apparent prevalence and severity of allergic rhinitis and rhinoconjunctivitis continues to increase around the world, particularly in countries with Westernized lifestyles [1,2]. The cumulative prevalence rate varies both within and among countries, with studies indicating a 20% rate in the United States—affecting up to 40 million people—and an average 23% prevalence in a population-based survey in Belgium, France, Germany, Italy, Spain, and the United Kingdom [3]. Many affected patients experience disruptive nasal and ocular symptoms, despite their use of effective pharmacotherapy, such as with nasal corticosteroids and nonsedating antihistamines [2]. Also very common is the sensitivity to insect venoms, with anaphylactic reactions occurring in 0.5% to 5% of the general population. This results in more than 500,000 hospital emergency-room visits and at least 40 deaths caused by anaphylactic reactions every year in the United States alone.

Allergen-specific immunotherapy (SIT) is the only immunomodulatory treatment that is currently available. It is the only treatment that has been shown to modify the natural course of allergic diseases, and it may also prevent the development of asthma in patients with allergic rhinitis [2,4–6]. Long-term effectiveness, persisting after cessation of treatment, has been demonstrated [2]. These benefits arguably justify the long-term commitment, in terms of time and money, and the potential risk of adverse reactions currently associated with immunotherapy [7].

Although progress has been made recently in the understanding of its immunological basis, the overall risk-benefit ratio for immunotherapy does not appear to have improved significantly as a result [7]. This may be due to variations in clinical practice, errors in dosing, labeling, and patient identification, the absence of standardized extracts for many important allergens, or the administration of injections to inappropriate candidates, especially patients with poorly controlled asthma [6–8].

However, a number of recent developments indicate the potential for significant improvements in both the safety and efficacy of immunotherapy. These include use of sublingual immunotherapy (SLIT) [8,9], administration of synthetic peptides [10,11], recombinant and engineered allergens [12,13], and the use of novel adjuvants [14]. Other new developments include the use of “rush” or “ultrarush” immunotherapy and immunotherapy in combination with the recently licensed anti-IgE antibody omalizumab [15]. Some of the evidence regarding these new options is reviewed in the context of existing practice.

THE EVOLUTION OF IMMUNOTHERAPY

Since the publication in 1911 of work by Noon and Freeman at St. Mary's Hospital, London [16], most research on immunotherapy has involved subcutaneous injection of allergens [5]. Noon and Freeman treated patients with allergic rhinoconjunctivitis with subcutaneous injections of increasing amounts of aqueous extracts of grass pollen, starting with very low doses and administering the maximum dose just before the start of the pollen season. Improvement of symptoms occurred in approximately 75% of the patients. The dose increment is necessary to minimize the risk of potentially life-threatening anaphylactic reactions. These can occur as a result of the interaction of the allergen with preformed IgE on mast cells and basophils, leading to widespread mediator release and, potentially, cardiovascular collapse. This potential risk led to immunotherapy becoming the exclusive domain of allergy specialists and specially trained staff. The original type of aqueous extracts, which contain a mixture of molecules from pollen, mold, dust mite, or other allergenic substances, are still widely used in the United States. In Europe, purified and modified allergens and depot adjuvants are used to reduce this risk. To form purified extracts, unwanted low-molecular-weight material is removed, as most true allergens have a molecular weight >10 kDa [5].

Modified allergens were developed to reduce the risk of side effects by reducing their capacity to elicit IgE-mediated responses during immunotherapy without reducing their immunogenicity [6]. This has been achieved by use of formaldehyde, glutaraldehyde, tyrosine, alginate, and aluminum salts to form so-called allergoids or depot preparations from the original allergen [5,6,14]. These methods became widespread in Europe during the 1970s. Depot preparations were developed to release allergen more slowly at the injection site, to reduce the risk of side effects, but also to heighten the immune response. Aluminum salts such as aluminum hydroxide are still widely used as adjuvants for antigens and allergens. However, safety concerns over use of injected aluminum have led to the development of other adjuvants: for example, monophosphoryl lipid A (MPL), a lipopolysaccharide (LPS) derived from bacterial cell walls [5].

LPS is recognized as a pathogen-associated potential threat by Toll-like receptor 4 (TLR4) on antigen-presenting cells of the immune system, especially dendritic cells, which are the most efficient antigen-presenting cells [17]. Immature dendritic cells are located in the epithelium of the skin and respiratory tract, where they ingest and process antigens. The presence of antigen stimulates these cells to

differentiate and migrate to a lymph node. Here they present the antigen via a class II histocompatibility molecules on the dendritic cell surface to naive CD4⁺ T cells, resulting in the formation and maturation of so-called Th1 or Th2 phenotype T cells, which mutually counteract each other. Hence, Th1 cells have the effect of downregulating the production of Th2 cells and the cytokines they produce, such as IL-4, which otherwise stimulate production of IgE. This switch from a Th2 to a more Th1 allergen-specific immune response is therefore believed to be a major component of the mechanism by which immunotherapy gradually desensitizes the individual to a specific antigen, along with production of blocking antibodies [2,5,6,18].

MPL-adsorbed tyrosine-adsorbed antigens, such as grass and birch pollen, have already shown promising results in European clinical trials [19]. Their major benefit appears to be a reduced course of treatment of only four injections prior to the pollen season each year, compared to the standard lengthy series of gradually increasing doses.

Recent work with T cells has revealed a cell-surface receptor—the Notch receptor—which is activated by a Notch-binding protein present on antigen-presenting cells [20]. The combination of T cell antigen recognition and Notch-pathway signaling appears to switch off the unwanted Th2-type immune response to antigen by release of immunosuppressive cytokines. The potential for antigens formulated with Notch-binding proteins as agonists is currently being assessed as a new approach to the treatment of allergy, but clinical data are not yet available [5].

Other new approaches to immunotherapy include use of recombinant technology to produce pure, reproducible, easily standardized molecules from the antigens to which a particular individual is sensitive and the development of bioengineered muteins [5,6,21]. These *muteins* are modified allergen molecules that retain the ability to stimulate Th1 T-cell responses without reacting with specific IgE antibodies. They are derived from allergenic proteins with the replacement of one or more amino acids from their IgE antibody combining sites.

Another important development is immunotherapy by alternative routes of allergen delivery—oral, sublingual, nasal, and bronchial. Interest in these alternatives increased after 1986, when the British Committee for the Safety of Medicines reported 26 fatalities caused by subcutaneous immunotherapy [22]. Nasal administration has been shown to be effective, but its use may be limited by local side-effects: nasal discharge, blocking, and sneezing [23]. In 1999, a position paper by a group of experts at the World Health Organization (WHO) concluded that the sublingual route in particular was a viable alternative to the subcutaneous route [24].

CLINICAL EFFICACY

Subcutaneous Immunotherapy

Subcutaneous immunotherapy has been shown to be a highly effective treatment for seasonal allergic rhinitis. It can result in a 50% reduction in symptom scores and an 80% reduction in medication requirements.

A systematic review of the available literature by WHO experts concluded that immunotherapy was effective in the treatment of allergy to inhalant allergens, such as pollen, dust mites, cat hairs, molds, and to insect venoms [6]. The only studies accepted as evidence were double-blind, placebo-controlled randomized studies, published in English as a full paper in a peer-reviewed journal. Patients had to be selected according to well-defined clinical criteria and a specific allergy diagnosis. The allergen involved had to be clearly defined and standardized.

Using these strict criteria, efficacy was confirmed in the treatment of allergic rhinitis due to grass, ragweed, *Parietaria*, mountain cedar, and coconut pollen in adults and children and in the treatment of allergic conjunctivitis due to grass and ragweed pollen [6]. It had a beneficial effect on bronchial symptoms and/or decreased the need for asthma medications in patients with allergic asthma due to pollen. Immunotherapy appears to be more effective in patients with single rather than multiple allergies.

The WHO suggested that immunotherapy is likely to be more rapidly effective in the treatment of pollen-induced rhinoconjunctivitis than perennial allergy because of the self-limiting nature of exposure to pollen resulting from the short pollination season of most plants. Long-term exposure to perennial allergens such as dust mite and molds may cause permanent inflammatory changes to airway mucosa and even remodeling of lung tissue in patients with chronic asthma, which immunotherapy cannot reverse. However, in patients with bronchial symptoms to cat allergens, immunotherapy resulted in improvement in symptoms and a reduction in the need for medication, possibly due to the fact that cat allergy is mediated mostly by Fel d1. In contrast, dust-mite allergy can be mediated by dozens of allergens from different *Dermatophagoides* species.

The WHO reviewers also reported high efficacy of immunotherapy in the treatment of insect venom-sensitive patients with efficacies of more than 80–95%. They and others state that Hymenoptera-venom immunotherapy is the only effective prophylactic treatment for insect sting-induced anaphylaxis [6,7].

Immunotherapy using purified venoms is an effective treatment for most venom allergic patients, although patients sensitized to honey-bee venom may be less well protected than those allergic to wasp or hornet stings. Most patients remain protected against further stings after venom immunotherapy lasting 3 to 5 years, even if they remain skin test positive [6]. Venom immunotherapy is recommended for patients at high risk for severe or systemic sting reactions, to be continued for at least 3 to 5 years in most cases [6,25]. Only 25% of patients become skin test negative after this period of treatment, but after 7 to 10 years, this may increase to 60–70% [25]. If patients become skin test negative, it is considered safe to discontinue venom immunotherapy [6]. Patients with a history of a severe reaction before immunotherapy and who have systemic allergic side-effects during immunotherapy are more likely to develop systemic reactions to stings after discontinuation [6].

After many years of debate, the long-term efficacy of specific immunotherapy was confirmed, in a landmark study by Durham [2]. This showed that 3 to 4 years of immunotherapy for grass pollen allergy induced clinical remission and a persisting reduction in immunological reactivity [2]. All of the patients involved had severe hay fever that was inadequately controlled by topical nasal corticosteroids and non-sedating antihistamines. The patients were initially followed over a 4-year period, with a placebo-treated control group. The grass-pollen vaccine used was highly effective, reducing symptoms scores and the need for rescue medication after the first year of treatment. Decreased sensitivity of the conjunctiva and skin and inhibition of the late skin response to allergen were also observed.

Clinical improvement was maintained over the next 3 years by continuing immunotherapy. To assess the effect of discontinuation of immunotherapy, Durham et al. followed the same group of patients for a subsequent 3-year period in a further placebo-controlled study [2]. A third matched group of patients who had never received immunotherapy was used as a control for the natural history of the disease in this study. Outcome measures included symptom scores, use of rescue medication, immediate sensitivity of the conjunctiva, and early and late skin responses to grass-pollen extract. Immunologic response was determined by assessing T-cell profile and cytokine (IL-4) production in skin specimens 24 hours after intradermal grass-pollen challenge. Total hay fever symptoms, use of rescue medication, and the visual analogue scale (VAS) for both the continuing immunotherapy group and the discontinuation group remained low, with no significant differences in any of these scores between the two groups throughout the 3-year period [2]. Symptom and rescue-medication

scores for both groups were markedly lower than those of the control group. In both the treatment and the discontinuation groups, the late skin response was virtually absent, and there were fewer infiltrating T cells and markedly fewer cells containing IL-4 mRNA than in the control group over the study period. The decrease in the number of cells expressing IL-4 mRNA suggests that persistent suppression of Th2 responses may be responsible for the sustained clinical improvement in these patients, as reflected by an inhibition of the late response. However, a tendency for immediate sensitivity to grass pollen to return was observed after 3 years in the discontinuation group. This did not result in the return of symptoms, suggesting that late responses are more relevant to the clinical expression of hay fever than early responses.

Sublingual Immunotherapy

The efficacy of sublingual immunotherapy (SLIT) has been assessed in a number of studies in recent years. The methodology involved is that an allergen extract in soluble tablet or drop form is kept under the tongue for a few minutes and then swallowed [8,9]. This is termed the “sublingual swallow” method. An alternative “sublingual spit” method is not widely used and has not yet been proved effective. The allergen doses used in recent studies range from 3 to 500 times those used in subcutaneous immunotherapy [9,23].

The WHO position paper in 1998 stated that only four studies demonstrated the clinical effectiveness of sublingual-swallow immunotherapy [6,26]. Two of these involved grass pollen as the allergen, the others used *Parietaria* and *D. pteronyssinus*. Symptomatic improvement was obtained and a reduction in reactivity during allergen-specific nasal or bronchial challenge was also described. However, the WHO stated that more studies were needed to identify the most appropriate patients, the optimal therapeutic target dose, and the effectiveness of this method [6,24,26].

Another review paper comparing the clinical efficacy and safety of subcutaneous and sublingual approaches to immunotherapy also concluded that the sublingual route appears to be a viable alternative but that further large-scale, controlled, randomized studies are needed to identify suitable candidates [4].

More recent work, however, has confirmed the efficacy of SLIT in both adults and children [4,8,23,24]. A systematic review and meta-analysis assessed 22 randomized, double-blind, placebo-controlled trials involving 979 patients with allergic rhinitis [23]. These trials used house dust mite, grass pollen, *Parietaria*, olive, and ragweed as

allergens. The overall analysis revealed significant reductions in both rhinitis symptoms ($p = 0.002$) and medication requirements ($p = 0.00003$) after SLIT. Subgroup analyses indicated similar benefits in the perennial allergy patients (dust mite and cat), but these results just failed to reach statistical significance, possibly due to the smaller number of patients involved. Similarly, the treatment effect in children did not reach significance perhaps because of the small number of children enrolled in the studies. The two studies that directly compared subcutaneous with sublingual immunotherapy reported similar improvements in symptoms and medication requirements for the two routes of administration [23].

Another review of SLIT concluded that the clinical efficacy in allergic rhinitis, in terms of reduction in symptom scores and medication requirements, ranged from 20% to 60% for grass, mites, birch, *Parietaria*, olive, cypress, and mixed tree pollens [8]. In mite-allergic patients, duration of treatment appears to be crucial. SLIT treatment for a minimum of 2 years has been shown to give positive results both in trials and in a general practice setting [8,27]. Shorter periods may result in clinical improvement that does not reach statistical significance. This is in line with observations from subcutaneous immunotherapy, which is also known to be less effective in mite-allergic patients than in those allergic to pollen.

SLIT has also been shown to reduce symptoms and use of β_2 -agonists and systemic steroids in patients with allergic asthma [8,24,28]. Improvements in lung function, bronchial hyperreactivity, and quality of life (QoL) have also been noted [24,29]. At least one study indicates that the efficacy of SLIT in allergic asthma is dose dependent [30]. Only limited data are available for a long-lasting effect of SLIT in asthma. However, one recently published study in 35 children treated for 4–5 years with SLIT indicates that there was a significant difference versus baseline for the presence of asthma as long as 5 years after discontinuation of treatment [8,31]. No such effect was apparent in a control group treated only with pharmacotherapy.

Other Approaches

Peptides containing T-cell epitopes (i.e., a unique molecular sequence that triggers a T cell immune response) but lacking IgE reactivity have been assessed for their value in immunotherapy [5,10,11]. This approach appears most likely to succeed when there is only one major or very few allergens in the source material, as in bee venom and cat hair [5].

The most convincing data has derived from studies using the epitope from the major bee venom allergen—phospholipase A₂ (PLA₂). Three

short peptides that stimulated T-cell clones from allergic and nonallergic subjects were used in patients who were allergic to bee venom [32]. These subjects were subsequently able to tolerate an injection of PLA₂ equivalent to one bee sting, and some subjects were able to tolerate a bee sting [5,32].

Other studies, using the peptides derived from the major allergen in cat hair—Fel d 1—have shown some efficacy but have caused late-phase adverse events, resulting in the FDA putting further trials on hold [5,18].

A number of recombinant allergens have been shown to have allergenic activity suitable for use in diagnostics such as skin testing and *in vitro* tests. These include derivatives of cat, mite, cockroach, grass, ragweed, birch, and peanut [18]. However, as yet there is no published evidence of their efficacy in clinical practice as allergens for immunotherapy. The same is true for the use of allergens genetically modified to reduce their IgE allergenicity. The theoretical advantages of such molecules may be outweighed by the practicalities of obtaining licenses for such molecules as pharmaceutical products. The likely cost of such products is also likely to limit their use on the scale needed to cope with the increasing prevalence of allergic conditions in the general population.

Immunotherapy in combination with immunostimulatory oligodeoxyribonucleotides (ODN) is another new approach that shows promise. Early studies have shown that a conjugate of ragweed allergen (Amb a 1) and immunostimulatory ODN sequences was 100-fold less allergenic than native allergen and had an equivalent immunogenic effect when used in immunotherapy for ragweed-allergic patients [33]. Patients experienced improvements in symptoms, medication use, QoL, and VAS scores during an initial ragweed season and continued through a subsequent season.

Use of omalizumab, an anti-IgE antibody, to reduce IgE levels close to zero has been shown to benefit patients with allergic rhinitis and asthma as a treatment in its own [15,34]. When used in addition to immunotherapy, it gave additional therapeutic benefits to children with seasonal allergic rhinitis. Omalizumab may also allow more rapid and higher doses of allergen to be used in rush immunotherapy and may allow the use of immunotherapy in high-risk patients, such as those with poorly controlled asthma.

SAFETY

Several surveys of health care professionals involved in allergy treatment have confirmed that avoidable errors in patient and allergen

identification and lack of adequate supervision after immunotherapy injections are largely responsible for the fatalities first highlighted by a British report in 1986 [22]. In the United States, between 1990 and 2001, a total of 20 deaths were reported, 19 due to immunotherapy and 1 to skin-prick testing [27]. As in other surveys, most deaths were in patients with asthma—often poorly controlled. Recommendations to avoid further fatalities have since been issued. These include avoiding skin testing and immunotherapy in patients with suboptimally controlled asthma, assessing respiratory function in all asthma patients prior to injection, extending the postinjection observation period, and providing high-risk patients with self-injectable adrenaline (epinephrine in United States). Immunotherapy should only be performed in suitable premises with well-trained staff, increasing identity checks, and only allowing one patient at a time in the injecting room.

Other measures to improve safety include the increasing use of sublingual immunotherapy. No serious systemic reactions or fatalities have ever been reported in any trials or postmarketing surveillance studies over a 15-year period with SLIT [8,9]. With subcutaneous immunotherapy, systemic reactions occur in 0.5–2.9% of patients, and in one study of children with severe asthma, 5.6% developed anaphylactic reactions [8]. The most frequent adverse event with SLIT is oral or sublingual itch after taking a dose. This is usually mild and self-limiting. These mild adverse reactions with SLIT tend to occur more frequently with lower dose levels, but systemic reactions occur rarely and are not dose-dependent [9]. A high rate of compliance—96.8% for mite allergen, 97.6% for pollen—was reported in one study with SLIT self-administered at home [27]. This was attributed to the low rate of side effects with this method.

Due to the low risk of adverse events with SLIT, rush immunotherapy may be carried out safely. It is possible that an increasing dose or build-up phase may be reduced or even omitted with this method.

The safety benefits of recombinant and engineered allergens remain to be fully determined, but early results with the Amb a 1 and immunostimulatory ODN sequences have demonstrated less allergenicity and good tolerability, with no serious adverse events reported [33].

CONCLUSIONS

Subcutaneous allergen-specific immunotherapy is now clearly established as an efficacious form of treatment for allergy, with the potential to modify the natural course of allergic rhinitis and possibly prevent the development of asthma in children. It is widely used around the world for a wide range of allergic conditions, including

rhinoconjunctivitis, conjunctivitis, asthma, and food allergies. For patients in whom pharmacotherapy fails to provide adequate symptom control, immunotherapy provides an effective alternative treatment option. And, for patients with insect-venom allergy, immunotherapy may be the only viable prophylactic treatment option.

Use of new technologies appears to offer the possibility of a reduction in the duration of therapy and the number of injections needed and, at the same time, improvements in safety. Research into the sublingual method is producing encouraging results, and this approach is well accepted by patients, aiding compliance. The convenience and safety profile are clearly superior to that for subcutaneous immunotherapy. If immunomodulators or improved adjuvants can be used to increase efficacy with no loss of safety, this approach may eventually lead to injections becoming unnecessary and could even be used in a primary care setting rather than specialist allergy clinics. In the long-term future, therefore, immunotherapy may become suitable for more general, widespread use, particularly in children. This could help to slow or even reverse the increasing prevalence of allergic disease seen around the world.

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