Impact of allergen immunotherapy in allergic asthma

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Although traditional pharmacological approaches improve outcomes in disease management for allergic asthma, these fail to modify the underlying immune responses. Allergen immunotherapy remains the only etiological therapy for the treatment of respiratory allergies for which clinical efficacy has been demonstrated through several well-controlled studies. In this review, we examine evidence from the past 5 years regarding the impact of allergen immunotherapy on allergic asthma to inform practitioners and stimulate further discussion and research.

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Allergen immunotherapy (AIT) is a well-recognized treatment for allergic disease that builds immune tolerance through the administration of specific allergens. It is the only known therapy to alter the natural history of respiratory allergy by targeting allergen-activated immune cells such as T helper 2 cells (Th2) and regulatory T cells (T_{reg}) [1]. Ever since Leonard Noon and John Freeman published their work on immunotherapy in 1911 [2], it has been studied extensively under various names (e.g., allergen or allergy immunotherapy, specific or allergen-specific immunotherapy) and allergens have been administered through different routes, but primarily via subcutaneous immunotherapy (SCIT) and sublingual immunotherapy (SLIT). Not only has AIT been known for its impact on disease modification, but it has also been suggested that AIT might prevent progression of allergic diseases [3]. AIT has demonstrated clinical efficacy in alleviating symptoms and reducing medication scores in allergic asthma by triggering a complex cascade of cellular and molecular events and altering a patient's immune responses to various allergens [4–6]. In contrast to traditional pharmacotherapy, AIT requires precise diagnosis of the disease-causing allergens to determine the best treatment plan [7]. There have been multiple reports and meta-analyses on the efficacy of AIT. AIT is used as an add-on to allergic asthma management [8]. Here, we review publications from the last 5 years on AIT with a focus on allergic asthma and present the latest findings regarding the mechanisms, clinical efficacy, safety and adjuvant use.

Mechanism of AIT

AIT causes a myriad of cellular and molecular events to take place, and these can be broadly classified into four groups.

Rapid desensitization of mast cells & basophils

After the initial exposure to therapeutic allergen doses, a decrease in mast cell and basophil activity is observed, and these cells become less responsive to environmental proteins, even in the presence of specific IgE. This process usually takes place over several hours [9]. Although the exact molecular mechanisms for this rapid desensitization process remain unclear, it is possible that the release of inflammatory mediators in quantities lower than the threshold for systemic anaphylaxis alters the subsequent threshold of activation of mast cells and basophils [10,11]. The selective suppression of basophils mediated by the histamine receptor 2 (HR2) is likely crucial for the early induction of allergen tolerance as demonstrated in a study of patients undergoing venom immunotherapy. Rapid upregulation of HR2 within 6 h of the build-up phase was observed and HR2 was noted to strongly suppress

Figure 1. Overview of immunological effects during allergen immunotherapy.

The balance between T helper 2 cells and regulatory T cells is crucial for the development or suppression of allergic inflammation. Blue arrows represent the regulatory effects of regulatory T cells which counteract the proinflammatory and activating effects of the T helper 2 cells as represented by the pink arrows.

FcεRI-induced activation of and mediated release of chemokines by basophils [12]. Evidence showed mast cells can contribute to innate host resistance to venoms during responses to an initial exposure. Mast cells, IgE antibodies and FcεRI can increase acquired resistance [13].

Activation of T_{reg} & B_{reg} cells

Peripheral T-cell tolerance is generally considered to be essential for normal immune system function, and for successful immunotherapy of allergic disorders [14]. T_{regs} maintain immunological unresponsiveness and contribute to allergic disease management by suppressing Th2-type immune responses [4]. The balance between Th2 and T_{reg} cells is crucial for the development or suppression of allergic inflammation (Figure 1). Antigen-specific T_{reg} and B_{reg} abundance increases during both natural allergen exposure and AIT, and when activated, both cell types secrete IL-10 which is essential for the induction of T-cell tolerance [15,16]. Increased IL-10 secretion is a key factor in the induction of a tolerant state though there are many other contributing factors [17,18]. Some of these additional suppressive factors include TGF-β, cytotoxic T lymphocyte antigen 4 and programmed death 1, all of which are crucial for peripheral tolerance to allergens [16]. Similarly, IL-10 secreting B_{regs} have been found to express high surface levels of CD25 and CD71, and low surface levels of CD73 [18]. These cells control excessive inflammation and are potent suppressors of antigen-specific CD4⁺ T-cell proliferation through the secretion of IL-10 [19].

T_{reg} & B_{reg} cells induce B cell class switching

Serum specific IgE levels often transiently increase during AIT, then gradually decrease over months or years [20]. IL-10 induces the production of antigen-specific IgG₄ which competes with IgE for allergen binding on the surfaces of mast cells and basophils [21]. Serum levels of specific IgG₄ increase during AIT, possibly blocking the allergen from reaching effector-cell bound IgE and thus preventing the activation of mast cells and basophils [22]. IL-10 and TGF- β secreted by T_{reg} and B_{reg} cells stimulate B cells to undergo class switching, potentially suppressing total and specific IgE and inducing the production of the protective antibodies IgG₄ and IgA₂ [15,21,23,24].

Suppression of late-phase effector cells (eosinophils & T-cells)

Successful AIT results in a diminished late-phase response (LPR), which is distinct from the immediate response to allergen provocation. The LPR involves the recruitment, activation and persistence of eosinophils and T cells at sites of allergen exposure, in contrast to the immediate response which is mast cell mediated [9]. Suppression of the LPR has been postulated to be relevant to the efficacy of AIT and its effect on chronic allergic inflammation [25]. This hypothesis is supported by studies that demonstrate how reduced plasma levels of markers of eosinophil activation and chemotactic factors for eosinophils and neutrophils correlate with clinical improvement [26,27]. During AIT, a decrease in bronchial, nasal and conjunctival hyper-reactivity to nonspecific stimuli, which may be a marker of underlying mucosal inflammation, is observed [26]. Plasma Endothelin-1, a bronchoconstrictor and proinflammatory peptide, is also significantly decreased in asthmatic children after immunotherapy [28]. A study reported that successful AIT could induce allergen-specific CD4⁺ T cells to expand and shift toward an 'anergic' Th2 phenotype, which is rarely present in either pretreatment participants or healthy controls [29].

Asthma & AIT

Asthma is characterized by airway hyperresponsiveness and in its later stages may lead to airway remodeling. Symptoms of asthma include recurrent breathlessness, wheezing, cough and chest discomfort. Asthma is a major public health problem with a high socio-economic impact. The report by the World Health Organization (WHO) estimates that approximately 235 million people suffer from asthma, and this number is projected to increase by an additional 100 million by 2025 [30].

Asthma is the most common noncommunicable disease among children. Although the prevalence of asthma is lower in the elderly population (~5%), the disease is often underdiagnosed, and is associated with significant mortality [31]. Asthma predominantly affects boys before puberty but in adults has a higher prevalence in women. A recent study showed that IL-4-induced M2 gene expression enhanced by estrogen served as a contributing factor to the sex-difference in asthma prevalence among adults [32].

Both genetic predisposition and environmental exposures are known to be risk factors for asthma. AIT is known to be effective in reducing the risk of asthma onset, and has been progressively applied for the treatment to allergic asthma [33] in combination with pharmacological treatment [34,35]. AIT has been reported to exert a beneficial effect when allergic asthma and rhinitis are concurrent; of note, a substantial share of the current data regarding AIT in asthma was presented in clinical trials designed to target patients with allergic rhinitis whose asthma was a comorbidity [36]. Therefore, measurements of asthma-related outcomes were oftentimes sought out secondarily [34].

SCIT & SLIT

SCIT and SLIT are the two major types of AIT in clinical practice. SCIT introduces allergens (house dust mites [HDM], pollens and animal dander) through repeated subcutaneous administration with a gradual up dosing regimen. This has been relatively well-studied compared with SLIT. SLIT is the other popular AIT method that has been studied since the mid-1980s and was approved as a treatment for adult allergic disorders in 1998. Durham *et al.* reported a grass pollen-AIT trial in which 106 patients with moderate to severe seasonal allergic rhinitis were randomized to receive treatment with SLIT, SCIT or placebo for 2 years. Results indicated that SCIT was more effective than SLIT in reducing total nasal symptom scores, however, the SLIT-treated group had a greater reduction in the use of seasonal rescue medication than the SCIT-treated group [37].

Efficacy

Both of the major AIT regimens have been shown to reduce symptoms with comparable effectiveness to that of pharmacologic medications [2]. The build-up phase required in SCIT is usually shortened or eliminated in SLIT. Significant decrease in the use of symptom-targeting drugs has been observed with SLIT particularly with

HDM [38]. It is reported that the long-lasting effects of SCIT and SLIT are maintained at least 7–12 years after discontinuation of treatment [2].

A recent review of SCIT and SLIT in pediatric allergic asthma stated that when compared with SLIT, SCIT demonstrated better effectiveness and had an earlier onset of action in asthmatic children. The authors examined 42 studies of SCIT and found a significant reduction of symptoms on treatment for HDM and pollen, but not for animal dander or allergenic mixtures. While wheezing responded better to SLIT, SCIT appeared to be more effective in reducing coughing. SLIT has been found to be generally effective in treatment of patients with HDM and grass pollen allergies [39] but studies are lacking for other allergens.

Dhami *et al.* synthesized data by random-effects meta-analyses from 98 AIT studies of allergic asthma to assess the evidence of AIT effectiveness. Reduction in short-term symptom scores was observed with a standardized mean difference (SMD) of -1.11 (95% CI: -1.66, -0.56), and reduction in short-term medication scores with a SMD of -1.21 (95% CI: -1.87, -0.54), although this may be subject to publication bias. Evidence was found supporting the efficacy of SCIT in reducing allergen-specific airway hyperactivity [40]. The 2017 GINA report recommends SLIT as an add-on option for selected adult patients who are HDM-sensitized and have exacerbations in asthma and allergic rhinitis despite Step 3 or Step 4 medium-dose inhaled corticosteroid (ICS) treatment, provided FEV1 is 70% predicted.

Disease-modifying effects

As the only treatment known to alter the natural course of allergic diseases, AIT has been shown to aid in the prevention of asthma onset and allergic rhinitis progression [34]. Back in 1992, the well-known preventive allergy treatment study was initiated to determine whether SCIT is effective in preventing asthma. In this study, 147 patients with grass and pollen allergy underwent 3 years of treatment. The results indicated that SCIT could reduce the development of asthma in children with allergic rhinitis [41]. Further, 5-year and 10-year follow-up reports of the study, which evaluated the long-term and preventive effects 2 and 7 years beyond cessation of SCIT, respectively, confirmed that long-term preventive effects of asthma was sustained [42,43].

Another report published in 2018 presented a retrospective analysis of the long-term efficacy of grass pollen SLIT tablets. 2,851 SLIT-treated and 71,275 control patients with allergic rhinitis were selected from the database for multiple regression analysis. After treatment was completed, significantly fewer asthma onsets, slower progression to asthma and decreases in asthma medication were observed in the treatment group compared with the control group [44].

To take advantage of the immunomodulatory properties of AIT, the European Academy of Allergy and Clinical Immunology (EAACI) published evidence-based guidelines on AIT for clinical practitioners. The guidelines recommended a 3 years course of AIT (SCIT or SLIT) for children and adolescents with moderate-to-severe allergic rhinitis who are allergic to grass/birch pollen, with the goal of preventing asthma onsets. The EAACI also noted the unmet need for more randomized, double-blind, placebo-controlled clinical trials for assessing the preventive effects of AIT [45].

Dose

The effective dose is allergen-dependent with both SCIT and SLIT and has largely not been established or standardized [2,46]. **Table 1** summarizes recent AIT studies in which HDM induced allergic asthma is the major focus. To achieve a similar level of clinical efficacy, the cumulative amount of allergen in 1 year could be as much as 200-times greater with SLIT compared with SCIT [47]. In a randomized, placebo-controlled, double-blind, double-dummy trial assessing the clinical efficacy of SLIT versus SCIT, birch pollen hay fever adults were treated for 2 years with SLIT, SCIT or placebo. The cumulative amounts of allergen administered were: SLIT 4717 µg of Bet v 1 and SCIT 27 µg of Bet v 1 in the first year; SLIT 11,182 µg versus SCIT 51 µg in the second year [48].

SCIT trials tend to have a clearer definition of effective and ineffective doses [46]. While SCIT has a sharp dose-response curve, SLIT has a broad range of effective doses [46]. To define the optimal SLIT dose, a 2015 review evaluated 17 randomized controlled trials and proposed 300 index of reactivity (IR) once-daily as the optimal dose to achieve best efficacy and tolerability for allergic rhinitis with or without HDM-associated moderate, persistent allergic asthma [47]. Casale *et al.* analyzed the safety profile of the 5-grass pollen tablet and concluded the 300IR SLIT is a safer alternative to SCIT for patients with intermittent asthma [49]. Jin *et al.* summarized the dosing regimens (which induced long-term effects) of common SLIT tablets used in patients with allergic rhinitis or allergic rhinoconjunctivitis due to grass pollen, ragweed or HDM. Effective daily maintenance dosing regimens

Table 1. Dose in allergen immunotherapy trials for house dust mite-related allergic asthma.

Form of AIT	Major allergen content	Population	Dose	Study (year)	Ref.
SCIT (build-up) SLIT tablet (maintenance)	HDM/ <i>Dermatophagoides pteronyssinus</i> and <i>Dermatophagoides farinae</i>	60 children, HDM-sensitized, with mild-to-moderate asthma	<i>Der p 1</i> and <i>Der f 1</i> , respectively SLIT: build-up: 1.5 and 1.5 µg maintenance: 52.8 and 52.8 µg SCIT build-up: 16.2 and 22.9 µg maintenance: 44.12 and 62.1 µg SCIT plus SLIT build-up: 16.2 and 22.9 µg maintenance: 43.2 and 43.2 µg	Keles <i>et al.</i> (2011)	[104]
SLIT drop SCIT	HDM/ <i>Dermatophagoides pteronyssinus</i> and <i>Dermatophagoides farinae</i>	32 children, monosensitized to HDM, with persistent rhinitis and asthma,	SLIT: Initiation at 1 drop of 10 TU/ml Up to 28 drop of 100 TU/ml SCIT: Initiation at 0.2–0.8 ml of 50 TU/ml Up to 0.2–0.8 ml of 5000 TU/ml Maintenance dose ratio SLIT/SCIT = 4.2	Yukselen <i>et al.</i> (2012)	[105]
SLIT drop	HDM/ <i>Dermatophagoides pteronyssinus</i> and <i>Dermatophagoides farinae</i>	484 patients, aged 16–50, with mild or moderate, persistent HDM-induced asthma for ≥ 1 year	300 IR	Wang <i>et al.</i> (2014)	[88]
SCIT	HDM/ <i>Dermatophagoides pteronyssinus</i>	90 asthmatic children, aged 5–14 years, with a mild-to-moderate HDM allergy, with or without allergic rhinitis	Initiation at 20 U/ml, Up to 100,000 U/ml	Hui <i>et al.</i> (2014)	[106]
SLIT tablet	HDM/ <i>Dermatophagoides pteronyssinus</i> and <i>Dermatophagoides farinae</i>	604 patients, 14 years or older, with HDM allergic rhinitis and mild-to-moderate asthma	6 SQ-HDM	Mosbech <i>et al.</i> (2014)	[107]
SLIT tablet	HDM/ <i>Dermatophagoides pteronyssinus</i> and <i>Dermatophagoides farinae</i>	834 patients, aged 17–83, with asthma not well controlled by ICS or combination products	6 SQ-HDM 12 SQ-HDM	Virchow <i>et al.</i> (2016)	[66]

AIT: Allergen immunotherapy; HDM: House dust mite; SCIT: Subcutaneous immunotherapy; SLIT: Sublingual immunotherapy.

include 2800 BAU of single-grass tablet (Grastek®), 300 IR of 5-grass tablet (Oralair®), five drops of 25 BU/ml grass liquid extract, 300 IR HDM tablet (Actair®) or five drops of 10,000 RAST units/ml HDM liquid extract [50]. Durham *et al.* evaluated pooled data from randomized, double-blind, placebo-controlled SLIT trials in patients with seasonal allergic rhinitis and perennial allergic rhinitis. The doses used in these analyses were 2800 bioequivalent allergen units (*Phleum pratense*, 75,000 SQ-T) for timothy grass SLIT-tablets, 12 units of *Ambrosia artemisiifolia* major allergen 1 (Amb a 1-U) for ragweed SLIT-tablets and 12 SQ-HDM for HDM SLIT-tablets [51]. Patients with dust mite- and timothy grass-induced allergic rhinitis undergoing dual SLIT self-administered daily doses of 15 µg of glycerated allergenic extracts of timothy grass and 20 µg of HDM. The cumulative dose of allergen after 12 months was 5475 and 7300 µg for timothy grass and HDM, respectively [52].

Effects on bronchial hyper-responsiveness

Bronchial hyper-responsiveness (BHR) is primarily recognized as the defining feature of asthma, but it has also been documented in various other pulmonary diseases including chronic obstructive pulmonary disease. Bronchial allergen provocation (BAP), a clinically established method to test allergic airway response to specific allergens, is recommended to assess the effect of different drugs on the success of AIT [53,54]. A negative methacholine challenge/BAP result rules out the diagnosis of asthma [55]. Changes in the early asthmatic response and the late

asthmatic response can be determined by BAP with the use specific allergens at a constant dose [56]. Buslau *et al.* reported considerable discordance in asthmatic response and BAP diagnosis and recommended to use levels of allergen specific-IgE as a better predictor of allergen-induced asthma in patients with allergic rhinitis [57].

AIT has a greater impact on allergen specific BHR than on nonspecific BHR. The 2010 Cochrane review of SCIT for asthma assessed the impact of SCIT on BHR with data from 19 studies that reported allergen specific BHR and 18 studies that reported nonspecific BHR. While there was an overall reduction in nonspecific BHR after SCIT and treated patients were significantly less likely to develop increased nonspecific BHR, SCIT introduced significant reductions in allergen specific BHR [54]. One controlled study showed that after 1 year of HDM-SCIT, the allergen specific BHR of patients in the treatment group significantly improved, and BAP results clearly differentiated the responders from nonresponders [58]. Ibero *et al.* reported significant improvement of specific BHR in HDM-allergic asthmatic children after 4 months of HDM vaccine treatment [59]. Similar improvement was also reported in children with birch and grass pollen allergy who underwent one cycle of monophosphoryl-a-adjuvanted SCIT and achieved specific bronchial tolerance [60]. In another randomized controlled trial where HDM-allergic asthmatic adults were treated with SQ-standardized HDM SCIT or placebo, nonspecific BHR was reduced similarly in both groups after 1 year [61].

Lung function

The inconclusiveness of AIT's impact on lung function has been repeatedly reported in multiple trials [30]. In the 2017 review, Dhami *et al.* mentioned that the association between beneficial effects of AIT and improvements in lung function or asthma control could not be established [40]. It has been demonstrated that HDM allergy can impact pulmonary function tests in asthmatic patients by the simultaneous presence of HDM-associated allergic rhinitis and asthma [62]. Therefore, by extrapolation, effective treatment of the associated allergy should lead to an improvement in lung function. Additionally, lung function tests are usually weighed as a secondary measurement, as normal results of lung function tests would not be inconsistent with a diagnosis of asthma, given that by definition, asthma patients have variable levels of bronchoconstriction [6].

Steroid-sparing effects

Treatment with ICS, which alleviate asthma symptoms and prevent asthma exacerbations, is a standard practice for asthma control [61]. The risk of systemic side effects associated with long-term ICS use warrants further studies of new treatments for asthmatic patients.

The steroid-sparing effect of AIT has been demonstrated in multiple studies. A 2010 randomized controlled trial reported a steroid-sparing effect with mite allergoid SCIT in children with mite-induced allergic asthma. The mean daily dose of fluticasone propionate in the treatment group 2 years post-SCIT dropped to 151.1 µg, compared with 330.3 µg at baseline [63].

Another study examined 90 mono- or poly-sensitized children with asthma who underwent allergen-specific SLIT between 2010 and 2013, and showed that the treatment resulted in 93.6, 83.3 and 73.7% of retained-avoidance of ICS in mono-allergen, pollen-mixture and two simultaneous-allergen group, respectively [64]. The reduction in the need for ICS treatment was also observed in adult patients with HDM-allergic asthma undergoing a 3 years SQ-standardized HDM SCIT [61].

Effects on asthma exacerbation

Asthma exacerbation refers to the progressive worsening of asthma symptoms that could lead to deterioration in lung function, increase in asthma medications, and even mortality. New treatment options and preventive measures for asthma exacerbation are in urgent demand [65]. A double-blind, randomized, placebo-controlled trial by Virchow *et al.* evaluated efficacy and safety of HDM-SLIT tablet by measuring time to asthma exacerbation during the reduction period of ICS treatment. 834 HDM-allergic patients with moderate or severe asthma were treated with placebo, or HDM-SLIT tablet alone, or HDM-SLIT tablet in combination with ICS. The study reported significantly lower risks of asthma exacerbations, alleviation of asthma symptoms and a significant increase in allergen-specific IgG4, in patients treated with HDM SLIT. The time to first moderate or severe asthma exacerbation during ICS reduction of the treated patients was also significantly improved [66].

IgE & IgG4

Increases in IgE levels are often seen when patients with allergic rhinitis and allergic asthma are naturally exposed to specific allergens. Allergen-specific IgE concentrations also rise soon after AIT initiation and they then gradually decrease over months or years to the pretreatment levels or lower [20,67]. Nevertheless, the decrease in serum IgE concentrations is not considered causative to AIT-induced unresponsiveness to specific allergen, as its correlation with clinical improvement is insufficient [9]. Other subclasses of antibodies including IgG, and particularly IgG4 continuously increase during treatment and increased antivenom IgG levels correlate with treatment-induced protection in venom-allergic patients [68]. IgG and IgG4 compete with IgE to bind allergen, and block allergen-IgE complex formation and its subsequent binding to FcγRIIb on B cells, thereby impeding allergen-specific TH2 responses, such as the activation of mast cells and basophils, suppression of histamine release by basophils [22,69,70]. A study by Wood *et al.* found that the administration of SCIT for cockroach allergen was immunologically more active than SLIT, especially with regard to IgG4 levels and blocking antibody responses [71]. The EAACI position paper on biomarkers of AIT for allergic asthma and allergic rhinoconjunctivitis published in 2017 recommended the use of allergen-specific IgG4 as a biomarker. Specific IgE/Total IgE ratio and serum inhibitory activity for IgE (IgE-facilitated allergen binding) are suggested as potential surrogate candidate biomarkers [70].

Safety

One of the major risks of SCIT is the development of systemic reactions, including anaphylaxis. Previous assessment of systemic reactions in SCIT suggested that it is possible to identify subjects who are prone to exhibit systemic reactions to SCIT at the initial evaluation based on the patterns of response to allergen skin tests. The risk to experience SCIT-related systemic reactions is almost six-times higher for 'highly allergic' patients who showed a pattern of more than 33% 3+ and 4+ skin test responses [72].

Through an assessment of systemic reactions in SCIT recorded on 28.9 million injection visits from 2008 to 2013, Epstein *et al.* identified uncontrolled asthma to be a major risk factor for serious or even fatal adverse reactions. Avoiding SCIT among patients based on asthma severity could significantly reduce the risk of severe reactions. Adjusting allergen dose during peak pollen season could potentially decrease the risk [73].

SLIT is generally considered safer than SCIT: no fatalities have been reported, and episodes of anaphylaxis with SLIT are rare. Despite this, certain risks in the form of local adverse events remain common. A clinical commentary review published in 2016 assessed SLIT-associated local side effects by reviewing large SLIT trials from 2006 to 2015. While SLIT tablets have been shown to be safe with occasional mild local side effects, SLIT with inhalant allergens are more frequently associated with systemic adverse events. Contraindications to initiate AIT include uncontrolled asthma, local lesions, or gastrointestinal disorders [74].

Eosinophilic esophagitis (EoE) has been reported as a complication of SLIT for respiratory allergies in adults. One recent review assessing four reported cases found that patients with respiratory allergies developed EoE following either monoallergen or multiallergen SLIT, and in each case the symptoms of EoE resolved after SLIT cessation [75]. Recently, a follow-up paper affirmed the correlation between aeroallergen SLIT and EoE [76]. It was conjectured that the gradual introduction of allergen during SLIT might cause chronic stimulation in atopic patients [75].

The World Allergy Organization position paper 2013 update on SLIT summarized relevant trials and meta-analyses published between 2009 and 2013. Comments were made that a uniform grading system to assess and classify local adverse events that occurred in SLIT was needed [33]. This fit well with a three-grade classification system previously proposed in 2013 that was based on reported data and the MedDRA nomenclature [77]. Seventeen side effects (affecting the mouth/ear and upper and lower gastrointestinal sites) were related to SLIT including itching, swelling, pruritus, ulceration, irritation and pain were tabulated, each of which could happen alone or in combination. The grading system with which patients' local side effects can be assessed is comprised of four levels: Grade 1 – Mild (not troublesome and no symptomatic treatment required), Grade 2 – Moderate (troublesome and symptomatic treatments required), Grade 3 – Severe (discontinuation of SLIT because of local side effects), and Unknown severity. The proposed grading system has been well accepted in recent literature because of its uniformity.

Multiallergen

The effectiveness of multi-allergen extracts in SCIT remains controversial. While the two well-known studies of multi-allergen SCIT published decades ago by Franklin W in 1967 and Johnstone DE in 1968 supported its effectiveness, a double-blind placebo-controlled trial by Adkinson *et al.* in 1997 indicated that it was ineffective.

The definitive evidence is currently lacking to support effectiveness of multi-allergen extracts in SLIT. While the efficacy of mono-allergen SLIT treatment for polysensitized patients has been validated [78], the use of multiple allergens in SLIT has been insufficiently studied for either efficacy or safety and, at present, evidence remains conflicting [5,79]. One review [33] cited case reports in which anaphylaxis was associated with multiallergen SLIT, but also presented contradicting results from other studies which found no such association.

Omalizumab as an adjuvant of AIT

Progress has been made in studying the use of adjuvants in the context of AIT. Omalizumab (Xolair, Genentech/Novartis) is a monoclonal anti-IgE antibody that has been used to reduce sensitivity to allergens. It specifically binds to IgE molecule at the same epitope whereby FcεRI binds. The anti-inflammatory and protective effect of omalizumab has been reported and well covered in the treatment of asthma alone.

Trischler *et al.* presented a smaller trial to investigate the time course of the early allergic reaction (EAR) and the late allergic reaction (LAR) under the effects of omalizumab. Ten patients with well controlled allergic asthma and a significant response to allergen challenge were administered omalizumab and BAP was repeated along the course of the study to determine the EAR and LAR. Results showed significant reduction of EAR after 4 weeks, and LAR after 1 week, which indicate the early onset of the protective effects by omalizumab [53].

An article published in 2014 summarized the use of omalizumab in SCIT. While it is generally accepted that the use of omalizumab could improve SCIT safety, the authors concluded that the beneficial effects with co-administration or pretreatment of omalizumab with SCIT was more evident in accelerated rush up dosing protocols for highly sensitized patients than in slower protocols in more stable patients [80].

Pre-treatment by omalizumab prior to immunotherapy initiation in children and young adults with severe asthma has been reported to increase tolerability to therapy, decrease side effects and amplify the efficacy of SCIT [39,81].

Despite the small number of subjects ($n = 6$), sustained tolerance to SCIT was observed after the withdrawal of omalizumab in this study. Further studies are necessary to determine the optimal length of omalizumab treatment [81].

Results from multiple clinical trials support the idea that the addition of omalizumab to SCIT protocols reduces the risk of systemic allergic reactions and symptoms of asthma more than conventional SCIT alone. However, it was also reported in grass-allergic patients that, this reduction in asthmatic symptoms was not sustained after omalizumab was discontinued [82].

A randomized, double-blind, placebo-controlled trial published in 2013 inspected the efficacy, safety and the long-lasting effect of SCIT combined with omalizumab. 140 patients with seasonal allergic rhinitis and comorbid seasonal allergic asthma were enrolled and randomly assigned to treatment with depigmented allergoid vaccine with placebo, or the vaccine in combination with omalizumab for 18 weeks. Outcomes of the study include better symptom severity reduction and slight Improvement of lung function in the group whose treatment involved omalizumab [83].

Other adjuvants

Several immunoregulatory cytokines, including IL-4, IL-13, IL-25 and IL-33, play important roles in the regulation of type 2 immunity and contribute to the development of asthma and allergic airway inflammation [24]. Anti-IL-4 monoclonal antibodies may enhance the induction of tolerance as IL-4 directly inhibits the FOXP3 promoter through IL-4-induced GATA3 [84].

Administration of anti-IL-25 in mice led to a significant reduction in IL-5 and IL-13 production, eosinophil infiltration, serum IgE secretion and prevented airway hyperresponsiveness [85]. It was reported that anti-IL-33 effectively inhibits airway hyperresponsiveness and inflammation in an HDM-induced asthmatic murine model [86]. It is likely that interleukin antagonists will be further explored in the future as adjunctive therapies with AIT [87].

Future perspective

Limitations & shortcomings

Although HDM-SLIT proves to be safe and appears to be effective in moderate, persistent asthma [88,89], studies of AIT in severe persistent asthma are scarce [33]. Severe unstable or uncontrolled asthma is a major risk factor for adverse events and systemic allergic reactions including severe bronchospasm, and thereby often a contraindication for AIT [34]. The WHO suggested that a forced expiratory volume in one second (FEV1) <75% in patients

with asthma should be a clear contraindication for SCIT. It has been proposed that, when asthma is the only manifestation of respiratory allergy, AIT should be conducted in combination with other therapies [90].

Despite more than 200 articles published between 2000 and 2016, there is a lack of head-to-head comparison of SCIT and SLIT trials on the effectiveness in asthma symptom alleviation, and the consensus on the optimal end-points of AIT as not been reached [91]. There is urgent need of reliable predictive biomarker of AIT efficacy [92], and the most favorable duration of AIT treatment is yet to be determined [35]. Many allergen formulations for AIT remain unstandardized and the optimal maintenance dose undetermined. While AIT could require considerable patient time, as the allergen administration regimen could take 3–5 years to complete, the earliest age for safe AIT initiation has yet to be determined [6].

Although SLIT is generally believed to be safer than SCIT, the safety profile of SLIT remains incomplete with potential associations with EoE. Risk factors for the occurrence of adverse effects in SLIT demand further investigation. Whether SLIT can be a safer alternative when patients withdraw from SCIT because of systemic reactions is yet to be answered.

In addition, allergen extract-based AIT is known to provide only partial protection. Variations in immunogenicity and potency of each individual allergen and the vast diversity of compositions in natural allergen extracts are current barriers in AIT efficacy and AIT vaccine development [93]. Present vaccines may fail to meet the need of patients' individual sensitization profiles and result in unsuccessful treatment. Treatment with certain important allergens with low immunogenicity could result in nonresponding allergic patients [94]. It is also noteworthy that systemic late-phase side effects could also be induced by IgE-independent allergen-specific T-cell activation and peptides which contains non-IgE-reactive T-cell epitopes [95]. SCIT with allergoids made from natural allergen extracts have been reported to induce late-phase side effects [96,97], which could contribute to failure of treatment. Overall, further investigation and innovation on forms of AIT is required.

Nanoparticles

There have been exciting recent advances in the use of nanoparticles (NPs) to improve the safety of SCIT. Administration of NPs with encapsulated allergens or DNA as the cargo has been shown to reduce the risk of systemic adverse reactions [92,98]. The use of NPs could potentially enhance the efficiency of allergen uptake in SCIT by elongating allergen presentation or by bypassing and/or targeting specific cell-populations [92].

Recent reports summarized the use of biodegradable NPs in SCIT including synthetic and natural polymers, liposomes, virus-like particles; and nonbiodegradable nanoparticles, such as carbon- or metal-based NPs. Results from murine studies showed that administration of poloxamine nanospheres loaded with HDM-coding plasmid reduced airway hyperreactivity and decreased the production of inflammatory cytokines in bronchoalveolar lavage fluid, but administration of carbon nanotubes led to the opposite effect by aggravating aforementioned symptoms. [98].

While most studies on the role of NPs in AIT were conducted in murine models, clinical trials which presented improvement in SCIT efficacy are also available. In one clinical trial in which virus-like particles encapsulating CpG were administered with HDM extract, significant reduction in allergic asthma symptoms was observed [92,99].

Antibodies produced to allergens

There are a number of novel ways to inhibit airway responses to allergens, including a new antibody to Feld1. A single prophylactic SC administration of the high-affinity monoclonal antibody binding Feld 1 epitopes demonstrated efficacy in preclinical models of cat-induced rhinoconjunctivitis. This represents a potential new approach to rational drug design to improve asthma outcomes with immunotherapy [100].

Recombinant allergy vaccines

Recombinant hypoallergenic allergen derivatives designed to inhibit IgE are emerging adjuvants to AIT. A recombinant B cell epitope-based grass pollen allergy BM32 has been studied in a SCIT trial and proved to be safe among patients with grass pollen-induced rhinitis and controlled asthma [101]. Long-term effect of BM32 is yet to be evaluated, and further investigation is needed to see if it can be safely administered to patients with severe asthma. [7,102]. A TAT-fused recombinant allergen has been shown to enhance SLIT efficacy in murine models, potentially by promoting T_{reg}-mediated immunomodulation [103].

An investigational DNA vaccine, ASP4070, includes a short DNA sequence encoding the endogenous human protein LAMP along with allergenic sequences. ASP4070, developed by Astellas Pharma to treat patients with Japanese red cedar pollen allergy has completed Phase I testing in the USA and Japan is currently in Phase II

dose response trials (ClinicalTrials.gov Identifier NCT03101267). It is designed to sensitize individuals by skewing IgE/Th2 allergenic response to an IgG/Th1 pathway.

Conclusion

Our knowledge of AIT has been expanding at an accelerated pace. In this review, our goal was to present the latest findings on AIT and their impact on allergic asthma and to aid clinical practitioners as well as researchers in understanding AIT's mechanism, efficacy and safety, impact on lung function, disease-modifying and steroid-sparing effects, dose, the use of adjuvants, its limitation and potential future directions. Generally, AIT has been shown to be effective in reducing symptoms in patients with allergic asthma, and has been recommended by national and international guidelines as add on therapy to pharmacotherapy, or for consideration as an alternative option for patients who are refractory to pharmacotherapy treatment [31], although close monitoring of patients may be required. Further research on AIT should be conducted to address urgent unmet needs including discovery of effective biomarkers, innovation on forms of AIT and investigation of AIT strategies toward chronic persistent asthma, as we enter the age of precision medicine.

Executive summary

Mechanism of allergen immunotherapy

- The myriad of cellular and molecular events underlying allergen immunotherapy (AIT) include rapid desensitization of mast cells and basophils, activation of T_{reg} and B_{reg} which induce B cell class switching, and suppression of late-phase effector cells (eosinophils and T-cells).

AIT in asthma

- As an add-on approach to pharmacological treatment, AIT has demonstrated a beneficial effect to allergic asthma control.
- Current evidence favors subcutaneous immunotherapy (SCIT) over sublingual immunotherapy (SLIT) on the efficacy in symptom reduction.
- The cumulative dose of SLIT could be as much as 200-times more than dose of SCIT.
- AIT is very effective in reducing specific bronchial hyper-responsiveness, but not very effective in nonspecific bronchial hyper-responsiveness.
- Impact of AIT on lung function is still inconclusive.
- AIT has demonstrated disease-modifying and steroid-sparing effects.
- The use of AIT in addition to asthma medication could improve time to asthma exacerbation during inhaled corticosteroids reduction.
- The use of multiple allergens in AIT remains insufficiently studied.
- IL antagonists can be further explored as adjunctive therapies with AIT.
- Pretreatment with or addition of omalizumab to SCIT protocols improves the safety.

Future perspective

- Unmet needs: safety profile of SLIT is incomplete, especially with severe asthma. Whether SLIT can be a safer alternative to SCIT is yet unknown, due to the lack of head-to-head comparison of SCIT and SLIT trials. Predictive biomarkers of AIT efficacy, optimal duration of AIT treatment, optimal end point and earliest initiation age needs to be determined. Further investigation of vaccine development and innovation on forms of AIT is required.
- The use of nanoparticles in AIT and combination of recombinant allergy vaccines with AIT are emerging in clinical studies.

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