Check for updates

Accepted Article

PROFESSOR MAMIDIPUDI THIRUMALA KRISHNA (Orcid ID : 0000-0003-2109-5777)

Article type : Unsolicited Review

Peptide Allergen Specific Immunotherapy for Allergic Airway Diseases – *state of the art*

David C Wraith[#] and Mamidipudi T Krishna[#*]

[#]Institute of Immunology and Immunotherapy, University of Birmingham, UK

[*]Department of Allergy and Immunology, University Hospitals Birmingham NHS Foundation Trust, UK

Running title: *Peptide immunotherapy for allergic diseases*

Word count: 4,412; Tables: 1; Figures: 1

Funding: None

COI: MTK received grants from FSA, GCRF and NIHR for research outside submitted work. MTK received funds from ALK Abello to attend an international conference. MTKs department has received an educational grant from Thermofisher, ALK Abello, MEDA and

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1111/cea.13840

This article is protected by copyright. All rights reserved

Accepted Article

other pharmaceutical companies for annual PracticAllergy course. The authors received a grant from MRC CiC for immunological validation of Der p 1 peptides for treatment of house dust mite allergy. DW serves as Chief Scientific Officer for Apitope Intl NV on a consultative basis; has sat on scientific advisory boards for Actelion Pharma, and Zealand Pharma; received travel funding from Apitope Intl NV; is a senior editor for Immunotherapy; holds patents for peptides, tolerisation-inducing composition, FVIII peptides and their use in tolerising haemophiliacs, composition, disease markers, tolerogenic peptides from myelin basic protein, peptide selection method; has consulted for Peptide Therapeutics Ltd., Teva, GSK Bio, Hoffman La Roche, Novartis, DTI, and the Food Standards Agency; received research support within the past 3 years from Apitope International NV, UCB Celltech, MRC, the Immune Tolerance Network, the Helmsley Trust, Diabetes UK and Wellcome Trust; holds stock and stock options with Apitope Int. NV; and was an expert witness for Geron.

Address for correspondence

Professor M T Krishna,
Department of Allergy and Immunology
Birmingham Heartlands Hospital
University Hospitals Birmingham NHS Foundation Trust
Bordesley Green East
Birmingham B9 5SS, UK
Tel: +44 1214241807
Fax: +44 1214243229
Email: mtkrishna@yahoo.com

ABSTRACT

Allergen-specific immunotherapy (AIT) is the only means of altering the natural immunological course of allergic diseases and achieving long term remission. Pharmacological measures are able to suppress the immune response and/or ameliorate the symptoms but there is a risk of relapse soon after these measures are withdrawn. Current AIT approaches depend on the administration of intact allergens, often comprising crude extracts of the allergen. We propose that the challenges arising from

This article is protected by copyright. All rights reserved

current approaches, including the risk of serious side effects, burdensome duration of treatment, poor compliance and high cost, are overcome by application of peptides based on $CD4^+$ T cell epitopes rather than whole allergens. Here we describe evolving approaches, summarise clinical trials involving peptide AIT in allergic rhinitis and asthma, discuss the putative mechanisms involved in their action, address gaps in evidence and propose future directions for research and clinical development.

Key words: allergen-specific immunotherapy, rhinitis, asthma, peptides, epitopes, T cells

Accepted Article

This article is protected by copyright. All rights reserved

Accepted Article

Introduction and a historical tribute

The concept of allergen-specific immunotherapy (AIT) was described over a hundred years ago for treatment of grass pollen-induced hay fever. Bostock in 1819 delineated hay fever as a seasonal illness characterised by airway catarrh, and Dunbar in 1903 associated it with 'pollen toxin'[1]. Dunbar described the muco-cutaneous symptoms in hay fever and that injection of 'pollen toxin' in animals induced production of neutralising antibodies. In a landmark publication in The Lancet in 1911, Noon elegantly described that prophylactic pre-seasonal inoculation of 'pollen toxin' induced 'active immunity' against hay fever[1]. The concept of allergen challenge, then described as 'sensitiveness', and how this was altered following repeated inoculation of 'pollen toxin' was ingeniously employed to obtain an objective read out of therapeutic benefit. A standardised pollen extract was manufactured by Dunbar's method that involved extraction in distilled water, by repeated thawing and freezing[1]. The potency of different pollens was compared with a conjunctival challenge procedure and it was established that *Phleum pratense* was most potent among different grass species in England. Noon developed a protocol of incremental subcutaneous inoculation of 'pollen toxin' at regular intervals and discovered enhancement of 'resistance' as evidenced by a 'conjunctival challenge[1]. Noon administered a relatively small starting dose as 1/3rd of the minimal dose that elicited a conjunctival response to challenge.

This work was carried forward by Freeman[2] who inoculated 20 patients with pollen extract and described therapeutic efficacy in 16 patients. Frankland and Augustin conducted the first 'controlled' clinical trial for hay fever and seasonal asthma and showed efficacy of 'crude' and 'purified pollen extracts'[3].

The fundamental principle underpinning AIT has not changed since it was first described in 1911, but great strides have been made with respect to standardisation, purification and optimisation of treatment protocols in order to maintain safety and maximise therapeutic benefit. The aim of AIT is to reduce allergic mucosal inflammation by induction of immune tolerance, thereby alleviating symptoms and improving health-related quality of life. Administration of

This article is protected by copyright. All rights reserved

Accepted Article

crude allergen extracts for AIT is associated with a risk of provoking an immediate (type-1) hypersensitivity reaction due to recognition of antigen/s or allergen by specific IgE on the surface of mast cells and basophils. Hence, efforts have focussed on development of novel approaches involving administration of modified allergens (recombinant allergens or peptides) to reduce/circumvent IgE binding, i.e., reduce allergenicity but maintain immunogenicity.

An ideal AIT product should meet the following criteria: cost-effective, minimal allergenicity and maximal immunogenicity and tolerogenicity, needing few doses or a short treatment course with ease of administration (preferably self-administration unsupervised), providing long-term therapeutic efficacy and importantly carrying minimal side effects in terms of provoking local and systemic allergic reactions.

Current approaches in clinical practice

Treatment modalities: The two main modalities currently employed worldwide for AIT in allergic rhinoconjunctivits (ARC) include subcutaneous injection immunotherapy (SCIT) and sublingual immunotherapy (SLIT). As per current British and European guidelines, these treatments are offered to patients with moderate-severe ARC that is either unresponsive or partially responsive to standard pharmacotherapy[4,5]. An important recent development has been the recommendation by the European Academy of Allergy and Clinical Immunology (EAACI) and Global Initiative for Asthma (GINA 2020; https://ginasthma.org/wp-content/uploads/2020/06/GINA-2020-report_20_06_04-1-wms.pdf) regarding use of AIT in house dust mite-driven allergic asthma[6]. HDM SLIT tablets have been recommended for adults as an 'add-on' or adjunct therapy in those with controlled or partially controlled disease[6] with an aim to reduce acute attacks. HDM SLIT drops have been similarly recommended for children with well controlled disease in order to reduce medication use and better control of asthma symptoms. Similarly, HDM SCIT has been recommended for adults with well controlled disease for better symptomatic control and reduction of medication usage[6]. It has been acknowledged by the authors of EAACI guidelines that the quality of evidence, however, is fairly limited at present with respect to use of AIT to treat

This article is protected by copyright. All rights reserved

Accepted Article

chronic asthma. Well-designed studies are needed to delineate the role for AIT in the context of mild-moderate asthma endotypes[7] (Type-2 'high' and non-Type-2 or Type-2 'low') based on predominant underlying inflammatory pathways, although it is likely to be more effective in Type-2 'high'.

Safety aspects: Safety remains an important concern, as uncontrolled asthma is an important risk factor for fatal and near fatal systemic allergic reactions to AIT. Hence, patient selection is paramount, and AIT is restricted to specialist use only in a secondary care setting. As regards UK practice, use of AIT in ARC is a relative contraindication in patients with concomitant mild-moderate well controlled asthma as per The British Society for Allergy and Clinical Immunology (BSACI) guidelines and an absolute contraindication in those with uncontrolled, severe or brittle asthma[5]. Furthermore, AIT is not recommended for the treatment of chronic asthma *per se* in the UK practice.

Several clinical trials with different AIT products have highlighted safety and efficacy (short-term and long-term) of SCIT and SLIT, particularly for grass pollen, birch pollen, ragweed and house dust mite[8-12]. They involve administration of a standardised intact whole allergen extract, although modified allergens (allergoids and polymerised extracts) have also been shown to be safe and effective. SCIT formulations include aqueous and depot (aluminium hydroxide or tyrosine to enhance immunogenicity) preparations. SLIT is available as dissolving tablets or drops. Systematic review and meta-analysis of SCIT and SLIT for ARC have shown that they have a modest/moderate therapeutic benefit[11,12].

Whilst mild oropharyngeal reactions are relatively frequent and commonly associated with SLIT, systemic reactions and anaphylaxis are very rare[4,5]. SCIT is associated with the risk of systemic reactions and anaphylaxis, and rarely fatal anaphylaxis could occur[4,5,13-15]. The SCIT national surveillance project from N America reported 7 fatalities between 2004-2017 out of 54.7million injection visits. SCIT associated systemic reactions occurred in 0.1% of injection visits, involving varying severity with greater risks associated with accelerated (rush and ultra-rush) protocols[15]. Hence, preference is to employ a conventional up dosing

This article is protected by copyright. All rights reserved

Accepted Article

protocol involving 12 weekly visits, particularly in the UK, where most whole allergen extracts for SCIT are currently unlicensed[5].

Practical aspects: There are recognised logistic issues with SCIT and SLIT[4,5]. SCIT should be administered under specialist supervision and in secondary care with access to critical care should anaphylaxis occur. Hence, there are overhead expenses attached to administration of SCIT. It involves considerable time commitment for patients with multiple visits and with a waiting time for an hour following each injection. SLIT on the other hand, is initiated in hospital and patients are trained for self-administration. Standard recommended duration of both treatments is 3 years. Those failing to show therapeutic response can be identified early and treatment may be withdrawn. Compliance to treatment has been highlighted as a concern in a real-life setting. A study from The Netherlands in 6486 patients (2789 SCIT and 3690 SLIT)[16] reported an overall 18% compliance with respect to completion of the 3year course of standard treatment (median 1.7 and 0.6 years respectively for SCIT and SLIT). One of the predictors for discontinuation was the prescriber, with patients more likely to persist with treatment with a general practitioner than with a specialist.

Other AIT modalities: Other approaches that have shown some promise include AIT with adjuvants (TLR-4 agonist, monophosphoryl lipid A; TLR-9 agonist, bacterial DNA oligonucleotides containing a CpG motif), combination of AIT with omalizumab, recombinant bet v 1, recombinant B cell epitope-based vaccine, comprising a recombinant hybrid grass allergen mix conjugated with a hepatitis B domain surface[17-23]. Other routes of administration have also been attempted including intradermal, epicutaneous and intralymphatic[24-26] routes. There have been several studies during the last 2 decades investigating efficacy and safety of short and long contiguous overlapping peptides (COPs) targeting dominant T cell epitopes of major allergens as an alternative to employing intact or whole allergens for AIT. Long COPs represent fragments of overlapping peptides covering the entire sequence of respective allergen, thereby preserving the relevant peptides for T cell recognition, but carrying an advantage of lacking conformation of the whole molecule to prevent IgE binding on the surface of mast

This article is protected by copyright. All rights reserved

Accepted Article

cells of basophils. Similarly, short peptides, generally comprising single T cell epitopes, are usually 8-20 amino acids. Selection of the correct sequence and optimising the length of peptide is critical to safety, success and cost of peptide AIT.

The main aim of this review is to critically appraise AIT involving peptide-based treatment for ARC. We will review clinical trials that have investigated the therapeutic efficacy and safety of peptide AIT for ARC, challenges associated with this modality, and describe putative mechanisms and future directions.

Rationale behind peptide immunotherapy

Peptide AIT in allergic airways disease rests on two fundamental principles[27,28], (a) a pivotal role for T cells in orchestrating chronic allergic mucosal inflammation in ARC and asthma and (b) circumventing IgE mediated recognition and cross-linking with allergen on the surface of mast cells and basophils, the mechanism underpinning systemic type-1 hypersensitivity reactions to whole allergen AIT.

Predefined and well mapped short peptides (SPs) or long COPs of major allergens representing dominant T cell epitopes, with the ability to bind to a vast array of HLA class II alleles, can be administered at regular intervals, either intradermally or subcutaneously to induce allergen-specific T cell tolerance[27,28]. This concept has been also been tested with some success in the context of autoimmune diseases where there are recognised target antigens such as in autoimmune liver diseases, rheumatoid arthritis, type I diabetes, multiple sclerosis and Graves' disease[28-34]. Targeting pathogenic T cells in organ-specific autoimmune diseases with T cell epitopes induces antigen-specific regulatory T cells and this strategy is under evaluation in clinical trials.

From a logistical and cost viewpoint, short peptides can be manufactured in a standardised fashion with low production costs and are relatively stable in a lyophilised form at room temperature[35]. Also, cost of production of peptide-based therapeutics is low since mass production of high purity product is achievable. Therefore, as soon as development costs are recovered, we predict that treatments for common allergic disorders will be inexpensive.

This article is protected by copyright. All rights reserved

Accepted Article

Analysis of clinical trials and studies involving peptide AIT for allergic airway diseases

Framework for peptide AIT: Phase 1-3 clinical trials have been conducted to investigate the efficacy and safety of peptide AIT for allergic airway diseases. These have mainly focussed on major allergens including birch pollen[36,37], grass pollen[38-44] or cat[45-56]. The main approach taken in these studies has been to administer SPs or COPs over 4-6 weeks, either starting at a relatively low dose with structured escalation to reach a target cumulative dose or by delivering a fixed pre-determined dose administered weekly or fortnightly. Most studies have investigated a dose-response relationship employing at least 2 doses with injections administered either intradermally or subcutaneously. Efficacy was measured using a standardised clinical scoring system of combined nasal/bronchial and ocular symptom and medication scores, night time nasal symptom scores, health-related quality of life scores, lung function (in some cases also airway hyper-responsiveness), early and late phase skin test response, measurement of biomarkers such as allergen-specific IgE (sIgE), allergen-specific IgG_4 and facilitated IgE binding assays.

Therapeutic response to peptide AIT: Peptide AIT has been shown to improve clinical responses to allergen challenge either following a natural exposure or during/after a controlled exposure in an Environmental Exposure Chamber (EEC) alongside alteration in immunological parameters with a significant increase in $sIgG_4$ and reduction in $sIgE/IgG_4$ ratio. Recent studies have also shown a sustained clinical benefit at year 1-3 following a relatively short course of therapy, alongside persistent immunological signals as described above, *albeit* with a downward trend from immediate post-treatment metrics[37,40].

Ellis et al[39] reported a phenomenon of a bell-shaped pharmacological response rather than a linear dose response association with grass pollen peptides, highlighting the importance of dose titration in early clinical trials to inform robust design of phase III trials and beyond. Whilst a cumulative dose of 48nmol of grass

This article is protected by copyright. All rights reserved

Accepted Article

peptides (4 or 8 injections) induced a significant therapeutic benefit, a similar (or expected dose-response benefit) response was not detected with a higher dose of 96nmol. This is in contrast to SCIT clinical trials involving an alum preparation of *Phleum pretense* which showed a linear dose-response relationship, i.e. a maintenance dose of 100,000 SQ.U was superior to 10,000 SQ.U of whole allergen extract in terms of therapeutic efficacy and associated with more frequent local and systemic treatment-emergent allergic reactions[8].

Safety aspects: Whilst recent studies with grass[39,41,42] and cat[55] peptides have been shown to be relatively safe with respect to grade 3 or 4 systemic reactions, as per the World Allergy Organisation (WAO) grading system, others, particularly those involving grass pollen[38], cat[47,52] and birch pollen[36] have reported relatively more severe early and delayed (>3hrs) systemic allergic reactions. Specifically, lower airway symptoms with a decline in FEV_1 (>30%) has been reported, and this has been attributed to a MHC restricted allergen-specific T cell activation[57]. It has been suggested that the late asthmatic response to peptides improves with repeated dosing[52], but this needs confirmation. Local injection site reactions have been less frequent and relatively less severe.

Putting evidence into context: In summary, peptide AIT is a promising option as an immunomodulatory and disease modifying treatment for ARC and asthma, but further work is clearly required in well characterised patients to demonstrate its long-term efficacy and safety. Current recommendations with whole allergen extracts of SLIT and SCIT are that 3 years of treatment is recommended to achieve long term efficacy[4,5]. Whilst there is preliminary and promising evidence that a short course of peptide AIT for 4-6 weeks confers sustained[37,40,48] clinical benefit at year 1-3, without additional treatment, further studies are clearly warranted to determine long-term efficacy with optimisation of dosing regimens, route of administration (intradermal vs subcutaneous), duration of therapy and safety specifically in asthma. Further research is also needed in children in order to determine the role for peptide AIT not only in established allergic airways disease, but also with respect to prevention of asthma and newer sensitisations as reported with whole allergen AIT[58]. Further research to investigate the efficacy

This article is protected by copyright. All rights reserved

Accepted Article

and safety of peptide AIT in house dust mite-related ARC and asthma are also needed. Importantly, it would be crucial to have a proportionate representation of Black, Asian and Minority Ethnic population in peptide AIT clinical trials to be able to maintain credibility and generalisability of findings at a global level.

Table-1 summarises key studies in peptide AIT undertaken during the last 3 decades and includes details regarding study design, efficacy and safety. Notably, AIT with Fel d 1 – derived peptides showed a stable treatment effect when the clinical response was measured employing an EEC. The significance of this effect however was challenged in a phase 3 study (ClinicalTrials.gov Identifier: NCT01620762; unpublished) in which primary outcome was measured by a combined score [CS = Total Rhinoconjunctivitis Symptom Score (TRSS) + Rescue Medication Score (RMS)] involving natural cat allergen exposure over a 3week period as opposed to challenge in a an EEC, and no significant differences were detected between the treatment and placebo groups. The reason for this study not reaching its desired primary end point is not clear. It is plausible that differences in study methodology, specifically method of cat allergen challenge (natural exposure vs EEC) and psychosomatic factors might potentially impact in cat allergy clinical trials. This needs due consideration in future peptide AIT clinical trials which should include a combined clinical cum biomarker approach, as successfully shown in recent studies involving grass pollen peptide AIT.

Putative mechanisms underpinning peptide AIT in allergic airways disease and lessons learnt from peptide immunotherapy in autoimmune diseases

Lessons regarding induction of immune tolerance from whole allergen AIT studies:

The fundamental question relating to effective AIT is the type of immune modulation required to produce effective tolerance towards the allergen. On the one hand, there is evidence that administration of conventional whole allergen-based immunotherapy leads to generation of allergen-specific blocking antibodies i.e. IgG_2 and especially IgG_4 isotypes that compete for binding antigen and block crosslinking of IgE on mast cells. For example, Zhao and colleagues have shown that the IgE-blocking factor induced by AIT correlates with IgG_4 antibodies and a

This article is protected by copyright. All rights reserved

Accepted Article

decrease of symptoms in HDM-allergic children[59]. Furthermore, Shamji and colleagues have shown that grass pollen SCIT leads to serum IgG_4 inhibitory antibodies that prevent IgE facilitated allergen binding by B cells and hence presentation to T cells[60]. On the other hand, there is evidence that AIT leads to the deletion, induction of immunological paralysis (anergy) or immune modulation (alteration of cytokine secretion with respect to dampening of Th2 and skewing towards a Th1 or Tr1 phenotype) of allergen specific T cells[61].

The key question is which form and mode of delivery of the antigenic component of an allergen is safe and effective in eliciting either the induction of blocking antibodies or modulation of allergen-specific $CD4^+$ T cells. In other words, should AIT involve active immunisation against the allergen or the induction of tolerance mechanisms designed to reduce the immune response to the antigen. One of the main barriers in our current understanding of AIT is that there are no reliable immunological correlates or biomarkers that determine therapeutic efficacy or indeed those that can accurately predict response to treatment at a patient level. The majority of patients remain sensitised to the respective allergen despite deriving clinical benefit post AIT and this phenomenon is also well recognised in the context of hymenoptera venom immunotherapy (VIT).

Peptides vs whole allergen: The advantage of using peptides rather than intact antigens is that peptides generally do not fold into the conformation found in the native antigen. As a result, it is most unlikely that a peptide would crosslink surface bound IgE antibodies even if the peptide retained a low affinity B cell epitope. Furthermore, this is even less likely with short linear peptides that generally form a random coil state.

The aims for a safe, effective and durable impact of AIT are to:

1. Induce immunological tolerance by administering a preparation that limits the risk of crosslinking IgE and hence causing anaphylaxis
2. Develop a treatment strategy that induces effective and long lasting tolerance within months rather than years

This article is protected by copyright. All rights reserved

3. Reduce the levels of Th2 cells specific for the allergen
4. Increase levels of both Foxp3$^+$ Tregs and IL-10 secreting Tr1 cells responding to the allergen
5. Increase the ratio of IgG4:IgE secreting B cells so as to increase levels of blocking antibodies

The only way to prevent IgE binding to a desensitising agent is to disrupt the conformation of the allergen and its associated B cell epitopes. This can be achieved using allergen fragments generated by enzymatic digestion of the allergen or synthesis of either COPs[36,37,62] or the design of short synthetic peptides (SPs) representing dominant T cell epitopes of the allergen[63]. Here we will discuss the pros and cons of the latter two approaches.

COPs vs SPs: COPs contain all of the CD4$^+$ T cell epitopes within the specific allergen and, therefore, have the desired immunomodulatory effect irrespective of individual variation in HLA type of the patient. This is more difficult with SPs; however, the promiscuous peptide binding properties of HLA-DR molecules means that pan-DR binding peptides can be designed that engage allergen-specific T cells in most individuals. The fundamental principle between COPs and SPs is, however, completely different. COPs are designed to be immunogenic, to induce immune modulation with induction of IL-10 producing T cells and an increase in the ratio of IgG_4:IgE. Importantly, it is known that IL-10 producing T cells such as Tr1 cells promote B cells to produce IgG_4[64]. IgG_4 has two important properties: first, it binds both complement and FcR poorly and hence does not promote inflammation; secondly, IgG_4 is functionally monovalent since its heavy and light chains can undergo half-Ig exchange[65]. Currently, COPs are administered with alum as adjuvant and as a result, there is a theoretical risk of both late phase reaction and anti-drug antibody development with repeated administration. Nevertheless, COP treatment of birch pollen allergy has led to clinical benefit for two seasons after a single pre-seasonal course with demonstrable immunological signals[37].

There is increasing evidence that SPs based on the CD4$^+$ T cell epitopes of allergens can induce tolerance and mediate suppression of the allergic response. The pioneers of this approach in allergy were Kay and Larche working with peptides from Fel d 1. They

This article is protected by copyright. All rights reserved

combined seven T cell epitopes from the allergen that was safe to administer and reduced the immune response to the antigen in allergic individuals. A short treatment with Fel d 1 SPs led to reduction in rhinoconjunctivitis symptoms that persisted for 2 years from the start of the treatment[48]. Campbell and colleagues confirmed that this approach generates IL-10$^+$ regulatory T cells capable of 'linked' suppression of the response to distinct T cell epitopes within the same allergen[66]. Similar observations have been made in SP-AIT studies of bee venom[67], grass allergen[39,40] and peanut allergy[68]. The induction of IL-10 secreting T cells in these SP-AIT studies is important since these Tr1 cells are known to promote IgG_4 production. Therefore, the induction of blocking antibodies, the aim of active immunisation with whole allergens or COPs, can also be achieved by tolerance induction with SPs.

Experience from peptide antigen immunotherapy in autoimmune diseases: The clinical trials summarised above are reminiscent of similar studies of SP administration in autoimmune diseases. SPs have been designed for a range of autoimmune diseases and have led to recent clinical trials in relapsing MS[69] and Graves' disease[70]. A phase 1b clinical trial of intradermal immunotherapy with SPs from a myelin antigen in saline solution showed a significant decrease in new/persisting T1 gadolinium-enhanced lesions from baseline to week 16, returning to baseline values at week 48[69]. Similarly, in Graves' disease, 7/10 mild to moderate hyperthyroid patients showed improvement in free thyroid hormone levels during the course of treatment. Importantly, no unexpected safety signals arose from administration of these SPs. It is interesting to note that a short course of treatment with SPs in ARC has been shown to induce long-term suppression of symptoms[37,40] whereas the use of SPs produces only short-term benefit in autoimmune disease[69,70]. We propose that this is due to the continued exposure of allergic patients to strong antigens and the generation of memory B cells producing blocking antibodies. Effective use of SPs for autoimmune diseases will require repeated administration to maintain suppression of the relatively weak response to their self-antigens.

Mechanisms associated with peptide antigen immunotherapy: The Wraith laboratory has used experimental models to reveal the detailed mechanism of antigen-specific immunotherapy with SPs (Figure 1). Not all T cell epitopes induce tolerance and it

This article is protected by copyright. All rights reserved

transpires that peptides must bind directly to MHC II on antigen presenting cells (APCs) to induce tolerance[71]. Recent work has shown that these antigen processing independent T cell epitopes (apitopes) bind preferentially to steady-state dendritic cells (DC) in lymphoid organs following subcutaneous injection (unpublished). Steady-state DC express low levels of costimulatory molecules and hence presentation of T cell epitopes by them is tolerogenic[72]. T cells responding to SPs presented by steady-state DC become anergic[73] and upregulate expression of inhibitory receptors (CTLA-4, TIM3, TIGIT and LAG3) and the transcription factors, MAF and NFIL3, that dictate IL-10 production[74]. The resulting Tr1-like cells suppress costimulatory molecule expression on neighbouring APC in an IL-10 dependent manner[75] and hence mediate both linked and bystander suppression[76]. Immune regulation following immunotherapy with SPs is reinforced by the generation of myeloid-derived suppressor cells (MDSC)[77] and IL-10 secreting Breg cells[29,78]. Recent work has shown that the generation of immunoregulatory Tr1 cells is governed by epigenetic priming of genes characteristic of a tolerogenic gene signature[79]. It seems likely that the mechanisms leading to the generation of Foxp3 and Tr1 cells are similar for allergens and self-antigens: we have much to learn from the parallel development of antigen-specific immunotherapeutic approaches in the fields of allergy and autoimmune diseases.

Future development of peptide immunotherapy for treatment of allergy and asthma

The time required to achieve effective tolerance by AIT for allergic airway diseases is currently months to years. Based on initial results of peptide therapy, however, it appears that tolerance to allergens is achievable with a short course of peptide AIT. Even so, there are likely to be combination therapies that could enhance AIT without disrupting tolerance induction. While the use of steroids can inhibit T cell activation, previous studies have shown that combined dexamethasone and vitamin D3 promotes Tr1 cell differentiation[80]. In addition, antibodies targeting effector arms of the allergic response, including anti-IgE or anti-IL-5, could be given while concurrently inducing tolerance by peptide therapy. Furthermore, it may now be possible to reduce levels of plasma cells using anti-CD38 antibodies, as tested recently in systemic lupus erythematosus[81].

This article is protected by copyright. All rights reserved

As mentioned above, COPs and SPs depend on fundamentally different mechanisms of active immunisation versus tolerisation respectively. It is possible that the effect of COPs could be improved by combination with virosomes. Anergis, the company producing COPs, has announced improved efficacy of their treatment when combined with virosomes from Mymetics, in an unpublished pre-clinical study. A number of groups are investigating different ways to deliver SPs for treatment of autoimmune diseases[82]. These include presentation by tolerogenic DCs, generated *in vitro* and then transferred back to the patient; delivery on red blood cells or nanoparticles designed to target the tolerogenic environment of the liver; combination with immunosuppressive drugs in nanoparticles or delivery on MHC coated nanoparticles. Although each of these approaches is tried and tested in pre-clinical models of autoimmune disease, none have yet been designed to treat allergic diseases. Any one of these approaches could improve on currently available approaches.

At present, we know little about the mechanism of action (MoA) of novel AIT approaches being test in clinical trials. We need to define the optimal conditions for induction of tolerance, immune correlates of effective desensitisation, kinetics of tolerance, longevity of disease suppression and molecular basis of effective immunotherapy. We now have the tools allowing us to focus on antigen specific cells isolated from patients[83] using peptide-MHC multimers and high throughput transcriptional profiling to define MoA. These tools enable us to address the critical questions listed above. Furthermore, we must use precision medicine approaches to define why each patient does or does not respond to treatment and hence allow better patient stratification.

In conclusion, we are now beginning to understand the MoA of conventional and novel approaches to AIT. We propose that peptide AIT will greatly enhance the safety and ultimately compliance levels in those treated and hence improve management of this rapidly increasing group of diseases. A number of distinct approaches are in the pipeline and when proven to be both safe and effective these will greatly improve the armamentarium of the allergist.

Figure 1: legend

This article is protected by copyright. All rights reserved

Induction of IL-10 secreting Tr1 cells by SPs. Soluble peptides administered without adjuvants are presented to naïve or effector T cells by antigen presenting cell (APC), including steady-state dendritic cell (DC). When encountering MHC and antigen on these steady state APC, where levels of costimulatory molecules are limited, T cells differentiate into IL-10 secreting Tr1 cells. The resulting Tr1 cells suppress costimulatory molecule expression mediate linked and bystander suppression of the response of other T cells and promote differentiation of myeloid derived suppressor cells (MDSC) and IL-10 secreting Breg cells. Furthermore, Tr1 cells induce class switching to IgG_4 in B cells.

Accepted Article

This article is protected by copyright. All rights reserved

REFERENCES

1. Noon L CB, C. Prophylactic inoculation against hay fever. *The Lancet* 1911; **1911**: 1572-3.
2. Freeman J. Further observations on the treatment of hay fever by hypodermic inoculation of pollen vaccine. *The Lancet* 1911; **1911**: 814-7.
3. Frankland AW, Augustin R. Prophylaxis of summer hay-fever and asthma: a controlled trial comparing crude grass-pollen extracts with the isolated main protein component. *Lancet* 1954; **266**(6821): 1055-7.
4. Roberts G, Pfaar O, Akdis CA, et al. EAACI Guidelines on Allergen Immunotherapy: Allergic rhinoconjunctivitis. *Allergy* 2018; **73**(4): 765-98.
5. Walker SM, Durham SR, Till SJ, et al. Immunotherapy for allergic rhinitis. *Clin Exp Allergy* 2011; **41**(9): 1177-200.
6. Agache I, Lau S, Akdis CA, et al. EAACI Guidelines on Allergen Immunotherapy: House dust mite-driven allergic asthma. *Allergy* 2019; **74**(5): 855-73.
7. Breiteneder H, Peng YQ, Agache I, et al. Biomarkers for diagnosis and prediction of therapy responses in allergic diseases and asthma. *Allergy* 2020.
8. Frew AJ, Powell RJ, Corrigan CJ, Durham SR. Efficacy and safety of specific immunotherapy with SQ allergen extract in treatment-resistant seasonal allergic rhinoconjunctivitis. *J Allergy Clin Immunol* 2006; **117**(2): 319-25.
9. Durham SR, Emminger W, Kapp A, et al. Long-term clinical efficacy in grass pollen-induced rhinoconjunctivitis after treatment with SQ-standardized grass allergy immunotherapy tablet. *J Allergy Clin Immunol* 2010; **125**(1): 131-8 e1-7.
10. Durham SR, Walker SM, Varga EM, et al. Long-term clinical efficacy of grass-pollen immunotherapy. *N Engl J Med* 1999; **341**(7): 468-75.
11. Dhami S, Kakourou A, Asamoah F, et al. Allergen immunotherapy for allergic asthma: A systematic review and meta-analysis. *Allergy* 2017; **72**(12): 1825-48.
12. Dhami S, Nurmatov U, Arasi S, et al. Allergen immunotherapy for allergic rhinoconjunctivitis: A systematic review and meta-analysis. *Allergy* 2017; **72**(11): 1597-631.
13. Rajakulasingam RK, Farah N, Huber PAJ, et al. Practice and safety of allergen-specific immunotherapy for allergic rhinitis in the UK national health service: A report of "real world" clinical practice. *Clin Exp Allergy* 2018; **48**(1): 89-92.
14. Bernstein DI, Wanner M, Borish L, Liss GM. Twelve-year survey of fatal reactions to allergen injections and skin testing: 1990-2001. *J Allergy Clin Immunol* 2004; **113**(6): 1129-36.

Accepted Article

This article is protected by copyright. All rights reserved

15. James C, Bernstein DI. Allergen immunotherapy: an updated review of safety. *Curr Opin Allergy Clin Immunol* 2017; **17**(1): 55-9.

16. Kiel MA, Roder E, Gerth van Wijk R, Al MJ, Hop WC, Rutten-van Molken MP. Real-life compliance and persistence among users of subcutaneous and sublingual allergen immunotherapy. *J Allergy Clin Immunol* 2013; **132**(2): 353-60 e2.

17. Drachenberg KJ, Wheeler AW, Stuebner P, Horak F. A well-tolerated grass pollen-specific allergy vaccine containing a novel adjuvant, monophosphoryl lipid A, reduces allergic symptoms after only four preseasonal injections. *Allergy* 2001; **56**(6): 498-505.

18. DuBuske LM, Frew AJ, Horak F, et al. Ultrashort-specific immunotherapy successfully treats seasonal allergic rhinoconjunctivitis to grass pollen. *Allergy Asthma Proc* 2011; **32**(3): 239-47.

19. Creticos PS, Schroeder JT, Hamilton RG, et al. Immunotherapy with a ragweed-toll-like receptor 9 agonist vaccine for allergic rhinitis. *N Engl J Med* 2006; **355**(14): 1445-55.

20. Rolinck-Werninghaus C, Hamelmann E, Keil T, et al. The co-seasonal application of anti-IgE after preseasonal specific immunotherapy decreases ocular and nasal symptom scores and rescue medication use in grass pollen allergic children. *Allergy* 2004; **59**(9): 973-9.

21. Casale TB, Busse WW, Kline JN, et al. Omalizumab pretreatment decreases acute reactions after rush immunotherapy for ragweed-induced seasonal allergic rhinitis. *J Allergy Clin Immunol* 2006; **117**(1): 134-40.

22. Pauli G, Larsen TH, Rak S, et al. Efficacy of recombinant birch pollen vaccine for the treatment of birch-allergic rhinoconjunctivitis. *J Allergy Clin Immunol* 2008; **122**(5): 951-60.

23. Zieglmayer P, Focke-Tejkl M, Schmutz R, et al. Mechanisms, safety and efficacy of a B cell epitope-based vaccine for immunotherapy of grass pollen allergy. *EBioMedicine* 2016; **11**: 43-57.

24. Senti G, Prinz Vavricka BM, Erdmann I, et al. Intralymphatic allergen administration renders specific immunotherapy faster and safer: a randomized controlled trial. *Proc Natl Acad Sci U S A* 2008; **105**(46): 17908-12.

25. Slovick A, Douiri A, Muir R, et al. Intradermal grass pollen immunotherapy increases TH2 and IgE responses and worsens respiratory allergic symptoms. *J Allergy Clin Immunol* 2017; **139**(6): 1830-9 e13.

26. Senti G, von Moos S, Tay F, et al. Epicutaneous allergen-specific immunotherapy ameliorates grass pollen-induced rhinoconjunctivitis: A double-blind, placebo-controlled dose escalation study. *J Allergy Clin Immunol* 2012; **129**(1): 128-35.

27. Larche M. Update on the current status of peptide immunotherapy. *J Allergy Clin Immunol* 2007; **119**(4): 906-9.

This article is protected by copyright. All rights reserved

28. Larche M, Wraith DC. Peptide-based therapeutic vaccines for allergic and autoimmune diseases. *Nat Med* 2005; **11**(4 Suppl): S69-76.

29. Wraith DC. Antigen-specific immunotherapy. . *Nature* 2016; **530**: 422-3.

30. Richardson N, Ng STH, Wraith DC. Antigen-Specific Immunotherapy for Treatment of Autoimmune Liver Diseases. *Front Immunol* 2020; **11**: 1586.

31. Pearce SHS, Dayan C, Wraith DC, et al. Antigen-Specific Immunotherapy with Thyrotropin Receptor Peptides in Graves' Hyperthyroidism: A Phase I Study. *Thyroid* 2019; **29**(7): 1003-11.

32. Chataway J, Martin K, Barrell K, et al. Effects of ATX-MS-1467 immunotherapy over 16 weeks in relapsing multiple sclerosis. *Neurology* 2018; **90**(11): e955-e62.

33. Burkhart C, Liu GY, Anderton SM, Metzler B, Wraith DC. Peptide-induced T cell regulation of experimental autoimmune encephalomyelitis: a role for IL-10. *Int Immunol* 1999; **11**(10): 1625-34.

34. Tian J, Atkinson MA, Clare-Salzler M, et al. Nasal administration of glutamate decarboxylase (GAD65) peptides induces Th2 responses and prevents murine insulin-dependent diabetes. *J Exp Med* 1996; **183**(4): 1561-7.

35. Moldaver D, Larche M. Immunotherapy with peptides. *Allergy* 2011; **66**(6): 784-91.

36. Spertini F, DellaCorte G, Kettner A, et al. Efficacy of 2 months of allergen-specific immunotherapy with Bet v 1-derived contiguous overlapping peptides in patients with allergic rhinoconjunctivitis: Results of a phase IIb study. *J Allergy Clin Immunol* 2016; **138**(1): 162-8.

37. Kettner A, DellaCorte G, de Blay F, et al. Benefit of Bet v 1 contiguous overlapping peptide immunotherapy persists during first follow-up season. *J Allergy Clin Immunol* 2018; **142**(2): 678-80 e7.

38. Shamji MH, Ceuppens J, Bachert C, et al. Lolium perenne peptides for treatment of grass pollen allergy: A randomized, double-blind, placebo-controlled clinical trial. *J Allergy Clin Immunol* 2018; **141**(1): 448-51.

39. Ellis AK, Frankish CW, O'Hehir RE, et al. Treatment with grass allergen peptides improves symptoms of grass pollen-induced allergic rhinoconjunctivitis. *J Allergy Clin Immunol* 2017; **140**(2): 486-96.

40. Ellis AK, Frankish CW, Armstrong K, et al. Persistence of the clinical effect of grass allergen peptide immunotherapy after the second and third grass pollen seasons. *J Allergy Clin Immunol* 2020; **145**(2): 610-8 e9.

41. Mosges R, Kasche EM, Raskopf E, et al. A randomized, double-blind, placebo-controlled, dose-finding trial with Lolium perenne peptide immunotherapy. *Allergy* 2018; **73**(4): 896-904.

42. Mosges R, Koch AF, Raskopf E, et al. Lolium perenne peptide immunotherapy is well tolerated and elicits a protective B-cell response in seasonal allergic rhinitis patients. *Allergy* 2018; **73**(6): 1254-62.

Accepted Article

This article is protected by copyright. All rights reserved

43. Sharif H, Singh I, Kouser L, et al. Immunologic mechanisms of a short-course of Lolium perenne peptide immunotherapy: A randomized, double-blind, placebo-controlled trial. *J Allergy Clin Immunol* 2019; **144**(3): 738-49.

44. Mosges R, Bachert C, Panzner P, et al. Short course of grass allergen peptides immunotherapy over 3 weeks reduces seasonal symptoms in allergic rhinoconjunctivitis with/without asthma: A randomized, multicenter, double-blind, placebo-controlled trial. *Allergy* 2018; **73**(9): 1842-50.

45. Worm M, Lee HH, Kleine-Tebbe J, et al. Development and preliminary clinical evaluation of a peptide immunotherapy vaccine for cat allergy. *J Allergy Clin Immunol* 2011; **127**(1): 89-97, e1-14.

46. Oldfield WL, Kay AB, Larche M. Allergen-derived T cell peptide-induced late asthmatic reactions precede the induction of antigen-specific hyporesponsiveness in atopic allergic asthmatic subjects. *J Immunol* 2001; **167**(3): 1734-9.

47. Oldfield WL, Larche M, Kay AB. Effect of T-cell peptides derived from Fel d 1 on allergic reactions and cytokine production in patients sensitive to cats: a randomised controlled trial. *Lancet* 2002; **360**(9326): 47-53.

48. Couroux P, Patel D, Armstrong K, Larche M, Hafner RP. Fel d 1-derived synthetic peptide immuno-regulatory epitopes show a long-term treatment effect in cat allergic subjects. *Clin Exp Allergy* 2015; **45**(5): 974-81.

49. Norman PS, Ohman JL, Jr., Long AA, et al. Treatment of cat allergy with T-cell reactive peptides. *Am J Respir Crit Care Med* 1996; **154**(6 Pt 1): 1623-8.

50. Smith TR, Alexander C, Kay AB, Larche M, Robinson DS. Cat allergen peptide immunotherapy reduces CD4(+) T cell responses to cat allergen but does not alter suppression by CD4(+) CD25(+) T cells: a double-blind placebo-controlled study. *Allergy* 2004; **59**(10): 1097-101.

51. Pene J, Desroches A, Paradis L, et al. Immunotherapy with Fel d 1 peptides decreases IL-4 release by peripheral blood T cells of patients allergic to cats. *J Allergy Clin Immunol* 1998; **102**(4 Pt 1): 571-8.

52. Maguire P, Nicodemus C, Robinson D, Aaronson D, Umetsu DT. The safety and efficacy of ALLERVAX CAT in cat allergic patients. *Clin Immunol* 1999; **93**(3): 222-31.

53. Alexander C, Tarzi M, Larche M, Kay AB. The effect of Fel d 1-derived T-cell peptides on upper and lower airway outcome measurements in cat-allergic subjects. *Allergy* 2005; **60**(10): 1269-74.

54. Alexander C, Ying S, Kay AB, Larche M. Fel d 1-derived T cell peptide therapy induces recruitment of CD4+ CD25+; CD4+ interferon-gamma+ T helper type 1 cells to sites of allergen-induced late-phase skin reactions in cat-allergic subjects. *Clin Exp Allergy* 2005; **35**(1): 52-8.

Accepted Article

This article is protected by copyright. All rights reserved

55. Patel D, Couroux P, Hickey P, et al. Fel d 1-derived peptide antigen desensitization shows a persistent treatment effect 1 year after the start of dosing: a randomized, placebo-controlled study. *J Allergy Clin Immunol* 2013; **131**(1): 103-9 e1-7.

56. Simons FE, Imada M, Li Y, Watson WT, HayGlass KT. Fel d 1 peptides: effect on skin tests and cytokine synthesis in cat-allergic human subjects. *Int Immunol* 1996; **8**(12): 1937-45.

57. Haselden BM, Kay AB, Larche M. Immunoglobulin E-independent major histocompatibility complex-restricted T cell peptide epitope-induced late asthmatic reactions. *J Exp Med* 1999; **189**(12): 1885-94.

58. Halken S, Larenas-Linnemann D, Roberts G, et al. EAACI guidelines on allergen immunotherapy: Prevention of allergy. *Pediatr Allergy Immunol* 2017; **28**(8): 728-45.

59. Zhao D, Lai X, Tian M, et al. The Functional IgE-Blocking Factor Induced by Allergen-Specific Immunotherapy Correlates with IgG4 Antibodies and a Decrease of Symptoms in House Dust Mite-Allergic Children. *Int Arch Allergy Immunol* 2016; **169**(2): 113-20.

60. Shamji MH, Kappen J, Abubakar-Waziri H, et al. Nasal allergen-neutralizing IgG4 antibodies block IgE-mediated responses: Novel biomarker of subcutaneous grass pollen immunotherapy. *J Allergy Clin Immunol* 2019; **143**(3): 1067-76.

61. Wambre E. Effect of allergen-specific immunotherapy on CD4+ T cells. *Curr Opin Allergy Clin Immunol* 2015; **15**(6): 581-7.

62. Fellrath JM, Kettner A, Dufour N, et al. Allergen-specific T-cell tolerance induction with allergen-derived long synthetic peptides: results of a phase I trial. *J Allergy Clin Immunol* 2003; **111**(4): 854-61.

63. Prickett SR, Rolland JM, O'Hehir RE. Immunoregulatory T cell epitope peptides: the new frontier in allergy therapy. *Clin Exp Allergy* 2015; **45**(6): 1015-26.

64. Satoguina JS, Adjobimey T, Arndts K, et al. Tr1 and naturally occurring regulatory T cells induce IgG4 in B cells through GITR/GITR-L interaction, IL-10 and TGF-beta. *Eur J Immunol* 2008; **38**(11): 3101-13.

65. Aalberse RC, Schuurman J. IgG4 breaking the rules. *Immunology* 2002; **105**(1): 9-19.

66. Campbell JD, Buckland KF, McMillan SJ, et al. Peptide immunotherapy in allergic asthma generates IL-10-dependent immunological tolerance associated with linked epitope suppression. *J Exp Med* 2009; **206**(7): 1535-47.

67. Muller U, Akdis CA, Fricker M, et al. Successful immunotherapy with T-cell epitope peptides of bee venom phospholipase A2 induces specific T-cell anergy in patients allergic to bee venom. *J Allergy Clin Immunol* 1998; **101**(6 Pt 1): 747-54.

68. Prickett SR, Voskamp AL, Phan T, et al. Ara h 1 CD4+ T cell epitope-based peptides: candidates for a peanut allergy therapeutic. *Clin Exp Allergy* 2013; **43**(6): 684-97.

Accepted Article

This article is protected by copyright. All rights reserved

69. Chataway J, Martin K, Barrell K, et al. Effects of ATX-MS-1467 immunotherapy over 16 weeks in relapsing multiple sclerosis. *Neurology* 2018.

70. Pearce SHS, Dayan C, Wraith DC, et al. Antigen-Specific Immunotherapy with Thyrotropin Receptor Peptides in Graves' Hyperthyroidism: A Phase I Study. *Thyroid* 2019.

71. Anderton SM, Viner NJ, Matharu P, Lowrey PA, Wraith DC. Influence of a dominant cryptic epitope on autoimmune T cell tolerance. *Nat Immunol* 2002; **3**(2): 175-81.

72. Steinman RM, Hawiger D, Nussenzweig MC. Tolerogenic dendritic cells. *Annu Rev Immunol* 2003; **21**: 685-711.

73. Sundstedt A, O'Neill EJ, Nicolson KS, Wraith DC. Role for IL-10 in suppression mediated by peptide-induced regulatory T cells in vivo. *J Immunol* 2003; **170**(3): 1240-8.

74. Burton BR, Britton GJ, Fang H, et al. Sequential transcriptional changes dictate safe and effective antigen-specific immunotherapy. *Nature communications* 2014; **5**: 4741.

75. Gabrysova L, Nicolson KS, Streeter HB, et al. Negative feedback control of the autoimmune response through antigen-induced differentiation of IL-10-secreting Th1 cells. *Journal of Experimental Medicine* 2009; **206**(8): 1755-67.

76. Anderton SM, Wraith DC. Hierarchy in the ability of T cell epitopes to induce peripheral tolerance to antigens from myelin. *Eur J Immunol* 1998; **28**(4): 1251-61.

77. Wegner A, Verhagen J, Wraith DC. Myeloid-derived suppressor cells mediate tolerance induction in autoimmune disease. *Immunology* 2017; **151**(1): 26-42.

78. Clemente-Casares X, Blanco J, Ambalavanan P, et al. Expanding antigen-specific regulatory networks to treat autoimmunity. *Nature* 2016; **530**(7591): 434-40.

79. Bevington SL, Ng STH, Britton GJ, Keane P, Wraith DC, Cockerill PN. Chromatin Priming Renders T Cell Tolerance-Associated Genes Sensitive to Activation below the Signaling Threshold for Immune Response Genes. *Cell Rep* 2020; **31**(10): 107748.

80. Barrat FJ, Cua DJ, Boonstra A, et al. In vitro generation of interleukin 10-producing regulatory CD4(+) T cells is induced by immunosuppressive drugs and inhibited by T helper type 1 (Th1)- and Th2-inducing cytokines. *J Exp Med* 2002; **195**(5): 603-16.

81. Ostendorf L, Burns M, Durek P, et al. Targeting CD38 with Daratumumab in Refractory Systemic Lupus Erythematosus. *N Engl J Med* 2020; **383**(12): 1149-55.

82. Wraith DC. Designing antigens for the prevention and treatment of autoimmune diseases. *Current Opinion in Chemical Engineering* 2018; **19**: 35-42.

83. Ryan JF, Hovde R, Glanville J, et al. Successful immunotherapy induces previously unidentified allergen-specific CD4+ T-cell subsets. *Proc Natl Acad Sci U S A* 2016; **113**(9): E1286-95.

This article is protected by copyright. All rights reserved

Accepted Article

TABLE-1: CLINICAL TRIALS* INVOLVING PEPTIDE IMMUNOTHERAPY FOR ALLERGIC RHINO-CONJUNCTIVITIS

Study	Allergen	Protocol	Key observations	Comments
BIRCH POLLEN PEPTIDES				
Spertini et al[36], 2016	Birch pollen (Bet v 1)	• Double blind randomised placebo controlled (DBRPC) parallel group phase-IIb; Betv1 long contiguous overlapping peptides (COP; 25 or 50 and 100mcg in aluminium hydroxide, subcutaneous injections) • 23 centres in 7 European countries • Placebo n=79; 50mcg n=79 and 100mcg n=82; white adults with seasonal allergic rhino-conjunctivitis • Pre-seasonal administration of 5 injections • Half dose on day 1 • 50mcg or 100mcg on days 8, 15, 29 and	• 50 and 100mcg arms effective, with improvement in rhinitis combined symptoms and medication scores (RSMS) and mini rhino-conjunctivitis quality-of-life questionnaire (RQLQ) • 20-fold increase in allergen-specific IgG4 in active groups with no significant differences between active groups • Significance increase in Betv1 specific IgE in placebo but not in active group • Short-term benefit shown with an immunological response	• Local injection site reactions related to alum were common • 6.5% of patients reported decrement in FEV1 of ≥30% >3hrs after injection, none graded as serious requiring hospitalisation • Mild late respiratory reactions occurred in 55%, 35% and 13% of patients in 100mcg, 50mcg and placebo arms • Other mild non-respiratory side effects

This article is protected by copyright. All rights reserved

Accepted Article

		57 • Inclusion criteria: 18-55years with moderate to severe rhino-conjunctivitis to birch pollen; positive skin prick test to birch pollen; positive specific IgE Bet v1 • Exclusion criteria: FEV1<80% of predicted; treatment for chronic asthma; perennial allergies; positive skin prick test to Bet v1 COPs; pregnancy or lactation; immunosuppressive treatment or allergen-specific immunotherapy during last 5years		reported • 3 serious adverse reactions in 50 and 100mcg groups, were reported (urticaria, oropharyngeal angioedema, conjunctivitis), treated with antihistamines, oral corticosteroids, inhaler and intramuscular epinephrine • Long-term studies needed, and better characterisation of those who are at risk of delayed reactions needed
Kettner et al[37], 2018; **follow on**	Birch pollen (Bet v 1)	• No further treatment offered prior to season-2; 240 participants (82%) returned for follow up	• Persistent significant improvement in RSMS in 50mcg group	• Persistent clinical benefit for two seasons after a single pre-seasonal

This article is protected by copyright. All rights reserved

Accepted Article

study of the above (Spertini et al, 2016)		• Inclusion and exclusion criteria: see above under parent study	• Persistent but a trend seen in 100mcg group with RSMS • Persistent improvement in mini RQLQ and night-time nasal symptom scores in 50mcg and 100mcg groups • Bet v 1-specific IgG4 levels dropped but remained persistently elevated in 50mcg and 100mcg groups compared to placebo and pre-treatment levels	course with demonstrable immunological signals
GRASS POLLEN PEPTIDES				
Ellis et a[139], 2017	Rye grass pollen (Cyn d 1, Lol p 5, Dag g 5, Hol l 5, Phl p 5)	• Multi-centre DBRPC study; pre-seasonal intradermal administration • Adults with a minimum 2year history of seasonal allergic rhino-conjunctivitis • • 4 groups: 1. 8x6 nmol, 2week intervals (n=71) 2. 4x12 nmol, 4week intervals (n=70) 3. 8x12 nmol, 2week intervals, (n=71) 4. Placebo, (n=70)	• Significantly greater reduction total rhino-conjunctivitis symptom scores in 8x6nmol group vs placebo	• 2 serious adverse events but not related to active treatment • No case of grade ≥3 hypersensitivity reaction as per World Allergy Organization (WAO) grading system, no case of anaphylaxis or treatment-induced

This article is protected by copyright. All rights reserved

Accepted Article

		• Baseline and post-treatment challenge in an environmental exposure chamber (EEC) • Inclusion criteria: 18-65years; minimum 2year history of grass pollen-induced allergic rhinoconjunctivitis; positive skin prick test to rye grass pollen; environmental chamber challenge: minimum TRSS and TSS of 10/24 and 6/12 respectively • Exclusion criteria: grass pollen induced asthma; anaphylaxis to grass pollen; FEV1<80% of predicted; allergen-specific immunotherapy in last 12months or grass pollen immunotherapy during last 10years; current or planned pregnancy; acute/chronic sinusitis; patients in whom epinephrine is contraindicated		asthma • Mild self-remitting local injection site reactions more frequent in active arm • No immune marker analysis carried out
Ellis et al[40], 2020; **Follow on to parent study (above**	Rye grass pollen (Cyn d 1, Lol p 5, Dag g 5, Hol l 5,	• N=122 and N=85 participants followed up in seasons of year 2 and 3 respectively • No further treatment offered	There was a non-statistical but a trend to significance with greater improvement in TRSS compared to baseline in 8x6 nmol group vs placebo group in years 2 and 3 off treatment	• Indicates persistent benefit at year 3 after a short course of treatment • No immune marker analysis carried out

This article is protected by copyright. All rights reserved

Accepted Article

Ellis et al, 2017)	Phl p 5)	• Primary endpoint: change in mean total rhino-conjunctivitis symptom score (TRSS) from baseline to follow up post-treatment challenge • Inclusion criteria: patients randomised in the previous study; completed all treatments and post-treatment challenge; mean baseline TRSS of ≥8/24 in previous study • Exclusion criteria: participants who were unblinded regarding treatment allocation in parent study; grass pollen-induced asthma since previous study; uncontrolled asthma or asthma requiring step-2 Global Initiative for Asthma (GINA) treatment or greater; FEV1<80% of predicted; allergen-specific immunotherapy since parent study completion; acute/chronic sinusitis; current or planned pregnancy; in whom epinephrine is contraindicated		
Mosges et al[42], 2018	Grass pollen	• Open label dose escalation study in adult patients (n=65) with moderate-	• Significant improvement in CPT, with 70% becoming non-reactive at	• No serious adverse events (SAEs) or

This article is protected by copyright. All rights reserved

Accepted Article

	(*Lolium perenne*)	severe grass pollen-induced seasonal allergic rhino-conjunctivitis • Purified *lolium perenne* extracts denatured and enzymatically hydrolysed to generate 1,000-10,000 Da *lolium perenne* peptides (LPP) • 12 incremental subcutaneous injections over 6weeks, starting at 10mcg, cumulative 490mcg • Baseline and post-treatment (week8) conjunctival provocation test (CPT) and blood tests for sIgE, IgG4 blocking abs • Inclusion criteria_ moderate-severe grass-pollen induced seasonal allergic rhinoconjunctivitis for 2years; positive skin prick test and specific IgE to grass pollen • Exclusion criteria: grass pollen immunotherapy during last 5years; ongoing immunotherapy with other	week8 • 12fold increase in sIgG4 abs at week8, 55% reduction in sIgE/sIgG4 ratio at week8 • Significant decrease in IgE facilitated allergen binding at week8	anaphylaxis • N=2 grade-1 and n=4 grade-2 reactions • Relatively small injection site reactions (mean wheal and flare <0.6cms and 2.5 cms respectively)

This article is protected by copyright. All rights reserved

Accepted Article

		allergens; chronic asthma or emphysema with FEV1 <80% of predicted; uncontrolled asthma; severe autoimmune disease; pregnancy; antihistamines or corticosteroids in 3weeks preceding the trial; those on beta blockers or ACE inhibitors, anti-IgE treatment, anti-leukotrienes or mast cell stabilisers		
Mosges et al[41], 2018	Grass pollen (*Lolium perenne*)	• Multi-centre DBRPC phase IIb parallel group study involving LPP devoid of adjuvant as an active arm • Subcutaneous escalating doses administered over 4weeks • Groups (LPP dose) 1. Cumulative dose 70mcg, n=50 2. Cumulative dose 170 mcg, n=49 3. Cumulative dose 370 mcg, n=53 4. Placebo, n=46 • 21% of patients had well controlled asthma	• Significant improvement in CPT post treatment compared to placebo arm • Significant improvement in facilitated allergen binding in a dose related fashion • Significant increase in sIgG4 abs post-treatment compared to placebo arm	• No cases of anaphylaxis or grade-3 or grade 4 allergic reactions • Mean wheal diameter at injection sites up to 0.8cms in active arm

This article is protected by copyright. All rights reserved

Accepted Article

		• Assessments: Baseline and post-treatment (week8) CPT and blood tests for sIgE, sIgG4 blocking abs and facilitated IgE binding assays • Inclusion criteria: 18-70years; at least 2year history of grass pollen-induced seasonal allergic rhinoconjunctivitis needing therapy; positive skin prick test to grass pollen and positive specific IgE to recombinant phleum pratense (phl p 1/5); positive conjunctival provocation test • Exclusion criteria: same as in above study and the following: fever; ongoing malignancy; PEF <70% of predicted; co-sensitisations to rag weed and mugwort with wheal diameter and/or sIgE exceeding grass pollen; positive serology to hepatitis B or C, HIV-1/2 and use of immunosuppressive treatment (oral, nasal and topical steroids) directly prior to the study		

This article is protected by copyright. All rights reserved

Accepted Article

Mosges et al[44], 2018	Grass pollen (*Lolium perenne*)	• DBRCPC parallel group phase III study; 57 centres in Europe • 1:2 randomisation, placebo = 182; active arm (LPP 170mcg) = 372; 8 subcutaneous injections over 3weeks • Inclusion criteria: Adults with moderate-severe grass pollen seasonal allergic rhinoconjunctivitis as per ARIA criteria during the last 2 years; positive skin prick test and serum specific IgE and conjunctival provocation test to grass pollen; seasonal asthma patients were included • Exclusion criteria: previous grass pollen immunotherapy; perennial allergic rhinitis; underlying systemic diseases; previous history of anaphylaxis; allergy to excipients in the vaccine; contraindication to epinephrine; FEV1 <80% of predicted and PEFR <70% of predicted	• Combined symptom and medication scores improved both over peak and during entire season by 15.5% and 17.9% respectively in LPP group in comparison to placebo • Reduced reactivity to conjunctival provocation test in LPP group • Improvement in QOL in LPP group	• Early systemic reactions: (<30minutes after injections): 10.1% vs 2.3%, LPP vs placebo; all except 3 were WAO grade 1 or 2; 1 grade-2, grade-3 and grade-4 (epinephrine given) WAO een in patients with asthma in LPP group • Systemic reactions: (after 30minutes): 21.2% vs 4, LPP vs placebo; all grade-1 or -2 WAO criteria

This article is protected by copyright. All rights reserved

Accepted Article

Sharif et al[43], 2019 **Sub-study of above study by Mosges et a, 2018**	Grass pollen (*Lolium perenne*)	• Protocol and selection criteria as above • Placebo = 11; LPP group = 21	• Combined symptom and medication score improved by 35% and 54% in LPP group in comparison to placebo during the peak period and entire pollen season respectively • Significant decrease in CD63 and CD203c^{bright}CRTH2 positive basophils in LPP group but not in placebo group • Suppression of seasonal increase in grass pollen specific IgE in LPP but not in placebo group • Significant reduction of IL-4^{+} Th2 cells and IL-4^{+} and IL-21^{+} follicular Th cells and dual IL-4^{+}IL-21^{+} follicular Th cells in LPP but not in placebo group • Induction of Foxp3^{+}, follicular T	Pre-seasonal LPP immunotherapy: • suppressed seasonal basophil activation • induced production of Foxp3 regulatory T cells • regulatory B cells and IgG_4 blocking abs

This article is protected by copyright. All rights reserved

Accepted Article

			and IL-10^+ regulatory B cells in all patients in LPP group with neutralising IgG_4 blocking abs	
Shamji et al[38], 2018	Grass pollen (*Lolium perenne*)	• DBRCPC parallel group study • 1:1:1 (n=27; grass pollen induced seasonal allergic rhino-conjunctivitis) receiving 5 escalating subcutaneous injections of placebo, LPP or LPP/recombinant DnaK (bacterial homologue of heat shock protein 70 family, an adjuvant) • Inclusion criteria: 18-50years; grass pollen-induced moderate-severe allergic rhinoconjunctivitis as per Allergic Rhinitis and Its Impact on Asthma (ARIA) classification for at least 2 previous years; positive skin prick test and specific IgE to grass pollen Exclusion criteria: None specifically stated	• Immunological signals: 1. Induction of sIgG4 abs persisting for 24weeks post-treatment 2. Significant reduction in IgE binding in LPP arm but not in LPP/DnaK or placebo	• LPP and LPP/DnaK was safe and well tolerated • 16 adverse reactions in LPP group and 12 in LPP/DnaK group; all mild-moderate and self-remitting • No alterations in haematological and biochemical parameters • First patient (at 50mcg) developed anaphylaxis after first LPP injection, and dose regimen was altered for rest of the study with starting dose at 5mcg
CAT PEPTIDES				

This article is protected by copyright. All rights reserved

Accepted Article

Patel et al[55], 2013	Cat (Fel d 1)	• RDBPCS, adults with cat allergy ± mild well controlled asthma • N=202; intradermal injections • 3 Groups (treatment for 3months) 1. Placebo 2. 8x3 nmol 2 weeks apart 3. 4x6 nmol 4 weeks apart • Baseline challenge in EEC for 3hrs/day for 4days • 18-22 weeks, post-treatment challenge in EEC for 3hrs/day54weeks, post-treatment challenge in EEC for 3hrs/day • Endpoint – change in total rhinoconjunctivitis symptom score • Inclusion criteria: 18-65years; history of cat-induced rhinoconjunctivitis and/or step-1 GINA asthma treatment for at least 1year	• Clinical benefit of 4x6 nmol 4 weeks apart superior to placebo and 8x3nmol arm • Benefit persistent at 1year after start of treatment	• No serious treatment-emergent adverse reactions • No decrement in FEV1 >30% in any arm of the study • Good safety profile • Clinical benefit seen 1year after start of treatment

This article is protected by copyright. All rights reserved

Accepted Article

		• Exclusion criteria: Persistent asthma or those using inhaled corticosteroids or leukotriene modifiers for asthma		
Couroux et al[48], 2015 **Follow on of above study (Patel et al 2013)**	Cat (Fel d 1)	• 2 years follow on of a previous DBRPC parallel group study • Adult patients • Challenges in an EEC pre and post treatment • Groups 1. 8 doses of 3nmol (n=17) 2. 4 doses of 6nmol (n=12) of synthetic peptides of immunoregulatory epitopes 3. placebo n= 22 • intradermal administration over 3months (SPIRES)	• Mean reduction in total rhino-conjunctivitis symptom score of 3.85 units in 4x6nmol group	• No serious adverse events during 2year follow up • A clinically meaningful reduction rhino-conjunctivitis score seen after 2years in 4x6nmol group • Evidence of long-term benefit at 2years • Larger multi-centre studies needed for further confirmation

This article is protected by copyright. All rights reserved

Accepted Article

		• Inclusion criteria: Participants who completed all visits in parent study were invited • Exclusion criteria: None specifically stated		
Worm et al[45], 2011	Cat (Fel d 1)	• RDBPCS, adults with cat allergy ± controlled asthma • Single dose intradermal or subcutaneous dose (0.03-12nmol) safety study • Late phase skin test response assessed 3weeks after treatment • N=40 placebo or Tolerommune CAT by intradermal route • N=48 placebo or Tolerommune CAT by subcutaneous route • Inclusion criteria: 18-65years; cat-induced allergic rhinoconjunctivitis with or without controlled asthma (GINA	• Maximum suppression of late phase skin test response was seen with 3nmol	• No serious adverse events • 2 subjects in the active group developed late asthmatic reaction with a 25-29% decline in FEV1, requiring treatment • Relatively mild reactions -naso-pharyngitis, cough and headache, more in subcutaneous group • Local injection site reactions seen in both groups

This article is protected by copyright. All rights reserved

Accepted Article

		2006 classification 1); positive skin prick test and/or positive serum specific IgE to cats for at least 1 year • Exclusion criteria: None specifically stated		
Smith et al[50], 2004	Cat (Fel d 1)	• RDBPCS in adult patient with cat allergic rhinitis and asthma • N=8 in active and n=8 in placebo arms • 12 overlapping Fel d 1 peptides • Escalating intradermal dose of 5, 10, 25, 50, 100 and 100mcg • Blood samples taken at baseline and post-therapy • Inclusion criteria: 18-55years with history of cat- allergic rhinitis and asthma; positive skin prick test to cat dander; withheld oral and inhaled corticosteroids for 2months prior to the study	• significant reduction in both proliferation and IL-13 production by allergen-stimulated CD4+ T cells in the active arm • $CD4^+CD25^+$ T cells suppressed proliferation and IL-13 production by $CD4^+CD25^-$ T cells in culture before and after treatment but peptide immunotherapy did not affect these responses significantly • Clinical responses and adverse events were not described in this report	• Fel d 1 peptide immunotherapy affects T cell response, but this may be independent of involvement of $CD4^+CD25^+$ T cells

This article is protected by copyright. All rights reserved

Accepted Article

		• Exclusion criteria: None specifically stated		
Oldfield et al[47], 2002	Cat (Fel d 1)	• RDBPCS in patients with cat allergic asthma • Fel d 1 peptides 5mcg, 10mcg, 25mcg and 50mcg (cumulative - 90mcg) at 3-4day intervals • 12 overlapping peptides spanning most of Fel d 1 • n=16 active or placebo (n=8) • Assessments at baseline, 40-weeks and 3-9months • Inclusion criteria: 25-50years; non-smokers; history of cat allergy in the past 12months; FEV1 reversibility with short acting beta-2 agonist of >20%; PC20 (bronchial hyperreactivity) with methacholine with <4g/l methacholine; positive skin prick test to Fel d 1 with late-phase response to intradermal Fel d	• Significant decline in early phase skin test response to allergen and Fel d 1 in the active arm at follow up visit-2 • Significant reduction in late phase skin test response to allergen at follow up visit 1&2 in active group • Significant reduction in late phase skin test response to Feld d 1 at follow up visit 1&2 in active group • PBMC responses to *in vitro* cat allergen: reduced proliferation and production of production of IL-4, IL-13 and IFNν, but not between groups. Increase in IL-10 production in active arm • Subjective improvement in clinical tolerance to cat allergen as per	• 4 out of 16 patients in active group developed late asthmatic reactions to Fel d 1 peptides, but could be desensitised with higher doses • This study provided proof of concept but was inconclusive

This article is protected by copyright. All rights reserved

Accepted Article

		1; no current illness and no clinically significant abnormalities in routine haematology, biochemistry and urine analysis • Exclusion criteria: None specifically stated	visual analogue scale in active arm compared to placebo • No significant changes in PD_{20} allergen and PC_{20} methacholine at the 2 visits between the 2 arms	
Norman et al[49], 1996	Cat (Fel d 1)	• RDBPCS in patient cat allergic asthma • N=95; placebo, 7.5mcg or 75mcg or 750mcg of ALLERVAX CAT • Weekly subcutaneous injections for 4weeks • Assessments at baseline and 6 weeks post-treatment with exposure a room with a live cat • Inclusion criteria: adults with rhinitis or asthma symptoms to cat exposure during the last 12months; positive skin prick test to cat dander; positive cat room challenge – increase in 2	• Significant improvements in nasal and lower airway symptom score in 75mcg and 750mcg group • Dose response effect seen	• No serious adverse events • Relatively mild allergic reactions occurred ≥1 hour post-first dose in 16/24 subjects in 750mcg group • Study demonstrated short-term therapeutic efficacy and safety of Fel d 1 peptide immunotherapy in cat allergy

This article is protected by copyright. All rights reserved

Accepted Article

		symptoms by 2 points or 1 symptom by 2 points and drop in FEV1 by 15% from baseline • Exclusion criteria: contraindication to immunotherapy (betablocker, major medical illness), tricyclic antidepressants, doxepin, astemizole, monoamine oxidase inhibitors during 6weeks prior; women of childbearing age not using adequate contraception; FEV1 <70% predicted; previous immunotherapy to cat or HDM; unstable or severe asthma; previous peptide therapy; cat ownership or regular environmental exposure		
Alexander et al[53], 2005	Cat (Fel d 1)	• Adults with cat allergic asthma with early and late phase response n=16 (8 in each arm, active and placebo); randomly assigned • 12 cat allergic subjects with early asthmatic response studied separately	Double-blind study: • Significant decrease in late phase asthma response at 3-4 weeks in active arm • Significant improvement in quality of life and eye, nasal and lower	• No serious adverse events. Five in active arm of double-blind study and 2 in open label study, developed late asthmatic response, but did not require nebulised

This article is protected by copyright. All rights reserved

Accepted Article

		and openly • Fel d 1, 12 overlapping peptides • Intradermal sequential dose escalations of 1, 5, 10, 25, 50, 100 and 100mcg (total 291mcg) at 14day intervals • Cat allergen-induced nasal and bronchial responses and Qol measures determined at baseline, 4-8weeks and 3-4months post therapy • Inclusion criteria: 18-55years; non-smokers; history of rhinitis and asthma symptoms following cat exposure in the preceding year; positive skin prick test to cat dander; positive late phase intradermal response to cat dander; FEV1 >90% of predicted; no previous history of immunotherapy; no current illness; no clinically significant abnormalities in routine haematology, biochemistry and urine analysis; none received oral, inhaled or nasal	airway symptom scores in active arm Open label study: • Significant improvement in nasal symptoms following cat allergen challenge	treatment, epinephrine or corticosteroids. Two patients required treatment with salbutamol accuhaler • Another proof of concept study demonstrating attenuation of early and late asthmatic response following treatment with Fel d 1 peptide immunotherapy

This article is protected by copyright. All rights reserved

Accepted Article

		corticosteroids for 6months, 2months and 7 days prior respectively • Exclusion criteria: None specifically stated		
Alexander et al[54], 2005	Cat (Fel d 1)	• Open label study • N=8 cat allergic asthmatics • Increasing doses of 11 Fel d 1 peptides, 0.1, 1.0, 5, 10 and 25mcg) administered intradermally at 14day intervals • PC_{20} to histamine and skin biopsies were taken at baseline and post-treatment following intradermal allergen challenge and subjected to immunohistochemistry and *in situ* hybridisation • Inclusion criteria: adults with history of cat allergic asthma; non-smokers; no other significant illness; PC20 histamine <4mg/ml; FEV1 reversibility >20% with short acting beta-2 agonist; discontinued	• Significant improvement in airway hyperresponsiveness and late phase skin test responses • Significant increase in $CD4^+/IFN\nu^+$ and $CD4^+/CD25^+$ cells post-treatment following intradermal challenge with cat allergen	• Proof of concept but inconclusive • 1 subject developed sneezing and cramping abdominal pain after the 3rd injection; epinephrine IM was administered although no change in vital parameters • Small sample size • Short course of Fel d 1 peptide immunotherapy reduces nonspecific bronchial hyperresponsiveness, reduces late-phase skin response to allergen

This article is protected by copyright. All rights reserved

Accepted Article

		antihistamines, leukotriene antagonist and inhaled corticosteroids for 2 days prior, positive skin prick test and serum specific IgE to cat dander • Exclusion criteria: None specifically stated		challenge and is associated with an induction of Th1 immune response as evidenced by influx of Th1 cytokine positive cells
Pene et al[51], 1998	Cat (Fel d 1)	• RDBPCS (part of a large multi-centre European study with ALLERVAX CAT) • Adults with mild cat allergic asthma • Placebo (n=6) or low (n=8; 15-45mcg), medium (n=6; 150-450mcg) or high (n=11; 1500-4500mcg) dose of Fel d 1 • Subcutaneous weekly injections for 6weeks • Assessments at baseline and 6weeks post-therapy • Inclusion criteria: Adults with history of cat allergy with respiratory symptoms on exposure; positive skin prick test and	• No significant difference in $PD_{20}FEV1$ between groups • Significant improvement baseline vs post-treatment in medium and high dose groups • IL-4 production significantly reduced in high dose group	• Proof of concept but inconclusive • Small sample size • Showed some immunological effect in high dose group

This article is protected by copyright. All rights reserved

Accepted Article

		serum specific IgE to cat dander; positive methacholine and cat allergen challenge tests • Exclusion criteria: FEV1 <70% of predicted; >3 asthma attacks per week; cat ownership or routine exposure; any form of immunotherapy in preceding 5years; treatment with systemic or inhaled corticosteroids, sodium cromoglycate, nedocromil sodium ketotifen or theophylline in previous 3months		
Maguire et al[52], 1999	Cat (Fel d 1)	• Multi-centre RDBPCS (part of a large multi-centre N American study with ALLERVAX CAT) • N=133 adult patients chronically exposed to cat or failed cat immunotherapy previously • N=27 (placebo), N=53 (75mcg) and N=53 (750mcg). 8 injections weeks 1-19; 2injections weeks 1, 2, 18 and 19	• Improved clinical tolerance to cat in active group but not placebo • Improvement in FEV1 in those with a reduce value at baseline	• Significantly greater adverse reactions in active group • Most adverse events reported were respiratory in origin and occurred in late phase following injection, improved with treatment • Early reactions – with

This article is protected by copyright. All rights reserved

Accepted Article

		• Dropouts: 11%, 23% and 15% in placebo, 75mcg and 750mcg arms • 40-49% of participants in each group had history of asthma • Pre- and post-treatment evaluation • Inclusion criteria children and adults with history of cat allergy (age range 13-64years); positive skin prick test to Fel d 1 • Exclusion criteria: unstable asthma; FEV1 <70% of predicted; underlying renal, hepatic, pulmonary, cardiovascular or psychiatric disease; women of childbearing age unless using effective and continuous birth control; history of alcohol or drug abuse; experimental drug within last 30days; participated in ALLER-VAX cat trial previously		30mins, mostly respiratory; 2 in placebo, 5 each in active arm • Late phase reactions: >30mins and <24hrs, more frequent in active arm (36% vs placebo (19%). Mostly respiratory and after first injection • Adrenaline requirement: 3 patients required treatment with adrenaline, 1 from 75mcg and 2 from 750mcg groups. *First – 20 minutes after injection 7 (750mcg). Second – 3.5hrs after first injection (750mcg). Third – 2hrs after first injection (75mcg)*

This article is protected by copyright. All rights reserved

Accepted Article

Simons, et al[56] 1996	Cat (Fel d 1)	• RDBPCS • 2 long Fel d 1 peptides (IPC-1 and -2) • N=42 adults with cat-induced rhinitis and/or asthma; 21 in active arm (750mcg of each peptide) and 21 in placebo arm • 4weekly subcutaneous injections • Skin prick tests at baseline and 2, 6 and 24weeks after last injection • Intradermal tests with cat at baseline and 2 weeks after last injection • PBMC cultures stimulated with Fel d 1 at baseline and 6&24weeks after the last injection and measurement of IL-4, -10 and IFNν • Inclusion criteria: adults with history of rhinitis and/or asthma symptoms following cat exposure; positive skin	• No significant changes in skin prick and intradermal tests following treatment • No significant changes *in vitro* T cell responses following treatment	• Adverse reactions within 24hrs more frequent in active arm – rhinitis, worsening of asthma, reduction lung function and pruritis • No clear explanation offered by authors for the contrasting observations in this study compared to other studies with Fel d 1 peptides

This article is protected by copyright. All rights reserved

Accepted Article

		prick test to Feld 1 • Exclusion criteria: cat owners; pregnancy or women of childbearing potential; underlying medical disorders; those who receive allergen immunotherapy in preceding 12months; ever received peptide injections; subjects requiring beta blockers, oral corticosteroids, ketotifen, astemizole, doxepin, tricyclic antidepressants or monoamine oxidase inhibitors,		

*Search strategy: The above studies were selected based on the following search strategy for this narrative review:

Databases used: Pubmed.gov (advanced search) and Cochrane library; dates – 01 Jan'90 to 31 Dec'20; filters – clinical trial; secondary search was based on references cited within selected output.

search terms used:

(a) rhinitis AND immunotherapy AND peptide

(b) asthma AND immunotherapy AND peptide

Outputs:

(a) Pubmed = 129; Cochrane library = 28

(b) *Pubmed =86; Cochrane library = 0*

This article is protected by copyright. All rights reserved

Accepted Article

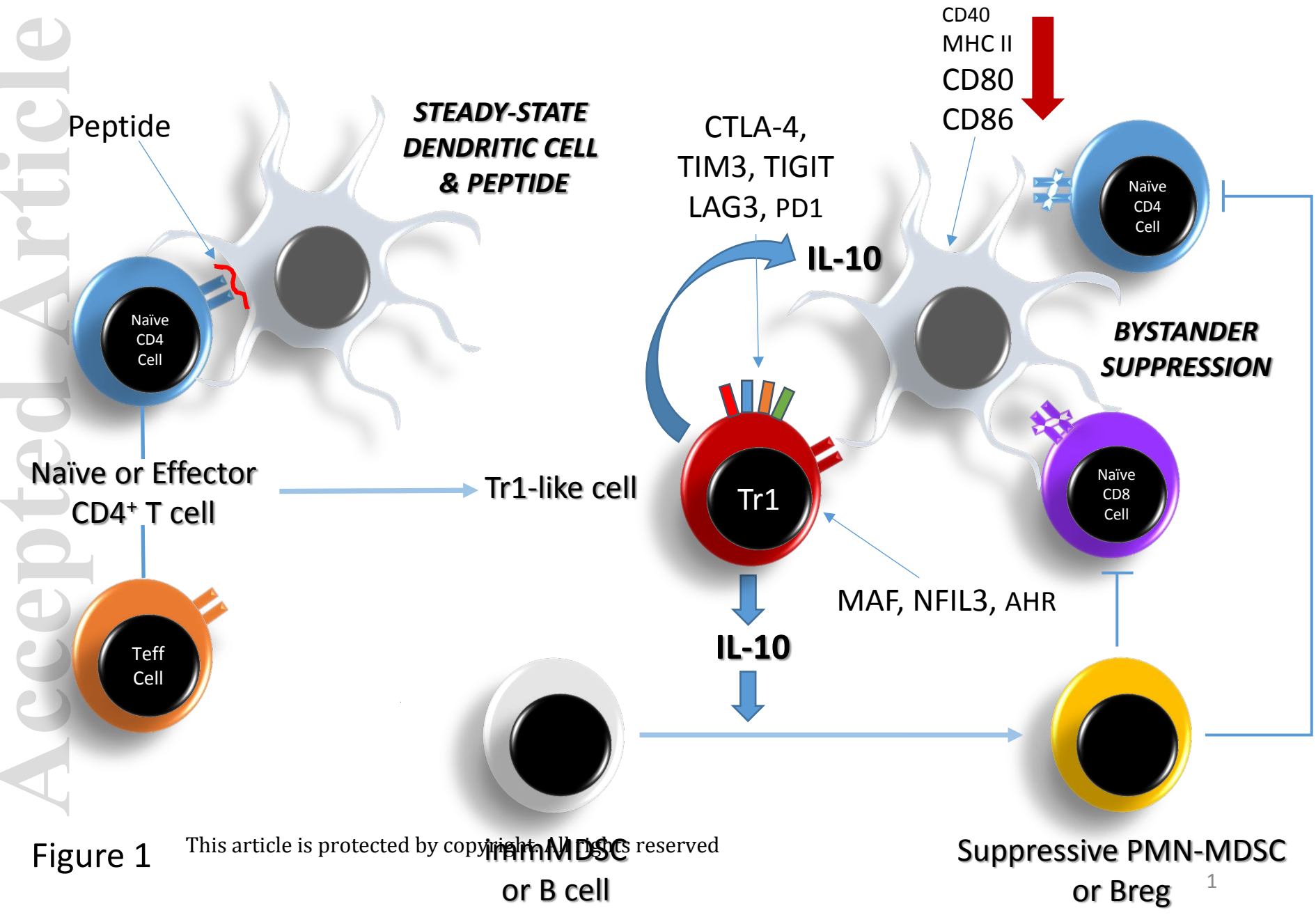


Figure 1

This article is protected by copyright. All rights reserved