

Expert Opinion

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Novel treatments for food allergy

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Food allergy is a major cause of life-threatening hypersensitivity reactions. Currently, the strict avoidance of the allergenic food and ready access to self-injectable adrenaline is the standard of care for food allergy. Based on extensive characterisation of food allergens and a better understanding of the immunological mechanisms underlying allergic disease, promising therapeutic modalities for the treatment and eventual prevention of food allergy are being developed. Novel immunotherapeutic strategies include peptide immunotherapy, traditional Chinese medicine, mutated or homologous protein immunotherapy, DNA immunisation and immunisation with immunostimulatory sequences, which all strive to elicit a decreased T helper cell type 2-like response or tolerance by the immune system in response to a specific food allergen. Other approaches such as the anti-IgE therapy or the Fc γ -Fc ϵ fusion protein aim at preventing the release of mediators by mast cells. It is the combination of these different approaches that would probably offer the best treatment option for food-allergic patients in a not too distant future.

Keywords: allergen-specific immunotherapy, anti-IgE immunotherapy, DNA immunisation, food allergy, immunostimulatory sequences, mutated allergen, peanut hypersensitivity, soybean homologous proteins, T cell-reactive peptide

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1. Introduction

Food allergy occurs in 4 – 6% of young children and 3 – 4% of adults, and is a major cause of life-threatening hypersensitivity reactions [1]. Food allergy is an immune-mediated disease and generally thought to be IgE mediated in most cases [1]. Food-induced anaphylaxis is the most common reason for someone to present to the emergency department for an anaphylactic reaction [2]. Currently, the avoidance of the allergenic food is the only method of preventing further reactions for allergic patients. Even with good educational information, 50% of all allergic patients have accidental ingestions and allergic reactions over a 24-month period [3]. With better characterisation of allergens and an understanding of the immunological mechanism involved in this reaction, investigators have developed several therapeutic modalities potentially applicable to the treatment and eventual prevention of food allergy [4]. Among the therapeutic options currently under investigation, there is peptide immunotherapy, traditional Chinese medicine, mutated protein immunotherapy, homologous protein immunotherapy, DNA immunisation, immunisation with immunostimulatory sequences and anti-IgE therapy. These novel forms of treatment for allergic disease hold promise for the safe and effective treatment of food-allergic individuals and the prevention of food allergy in the future.

Desirable therapeutic strategies for the treatment and prevention of food allergies must be safe, relatively inexpensive and easily administered. Recent advances in the understanding of the immunological mechanisms underlying allergic disease and the better characterisation of food allergens have greatly expanded the potential therapeutic options for future use.

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2. Traditional therapies

Immunotherapy for IgE-mediated disease has been used since its first description nearly a century ago [5]. Although injection immunotherapy has traditionally been employed in the treatment of inhalant allergies, such as allergic rhinitis, it has been successfully used in the treatment of food allergy. Injection immunotherapy was first used in the treatment of food allergy when a young child was successfully desensitised to fish [6]. In patients with allergic rhinitis experiencing oral allergy symptoms with the ingestion of crossreacting allergens in fresh fruits, nuts and vegetables, traditional injection immunotherapy has also been successful in many cases in ameliorating the oral allergy symptoms with minimal adverse reactions [7-10].

Traditional injection immunotherapy for food allergy is currently not recommended because of the allergic side effects of the therapy [11]. Studies have been conducted in a double-blind, placebo-controlled trial of rush immunotherapy for the treatment of anaphylactic hypersensitivity to peanuts [12]. Patients in the treatment group were able to tolerate increased amounts of peanuts in post-treatment food challenges. Unfortunately, an unacceptably high rate of adverse systemic reactions was associated with the rush immunotherapy and maintenance protocols [12,13]. The actual peanut immunotherapy produced so many side effects that it would be impossible to perform on a routine basis.

Although food immunotherapy in the oral allergy syndrome has been generally successful and well tolerated with crossreacting pollen immunotherapy [14], the practice of injection immunotherapy for food allergy has been largely abandoned due to the associated risk of serious systemic reactions. As traditional immunotherapy has been largely impractical for the treatment of most food allergies, several novel therapies are now being explored (Table 1).

3. Novel therapies

Techniques under current investigation for the treatment of food allergy in both animal models and humans include peptide immunotherapy, traditional Chinese medicine, mutated protein immunotherapy, allergen DNA immunisation, vaccination with immunostimulatory DNA sequences and anti-IgE therapy.

3.1 Peptide immunotherapy

Peptide immunotherapy utilises peptide fragments containing T cell-reactive epitopes rather than complete protein molecules. Theoretically, these peptide fragments are unable to crosslink the IgE molecules required to activate mast cells, but do appear to render T cells unresponsive to subsequent allergen exposure [15]. In one study utilising peptide immunotherapy, the investigators injected cat-allergic patients with T cell-reactive peptide fragments containing dominant cat allergen epitopes [16]. Patients tolerated the injections with few, mild adverse reactions. The investigators demonstrated

that cat-allergic patients treated with peptide fragments had statistically significant improvement in nasal, lung and total symptom scores compared with placebo-treated subjects after exposure to cats [16]. A similar approach that has involved larger numbers of peptides from cat has been shown to be effective [17]. Cat-allergic patients treated with intradermal cat peptides developed a significant immune response, as well as clinical improvement in their symptoms. Another preliminary study demonstrated that pepsin-digested peanut allergen containing T-cell epitopes, but no IgE-binding epitopes, induced IFN- γ , a T helper type 1 (T_H1) cytokine, in a concentration-dependent manner [18]. Most recently, T-cell epitopes of the major peanut allergen, Ara h 2, were mapped, laying down the groundwork for designing peptides that could be used in future *in vivo* studies [19]. Although peptide immunotherapy for food allergy has not yet reached clinical trials, studies utilising the peptides of the peanut allergens are encouraging and suggest a potential role for peptide immunotherapy in the future therapy of food allergy.

3.2 Traditional Chinese medicine

Herbal remedies have been used in Asia for centuries for the treatment of allergic diseases [20,21]. They have a favourable safety profile and the advantage of low cost. Recent studies have utilised traditional Chinese medicine in a mouse model of peanut allergy [21,22]. IgE levels were significantly reduced by the herbal formula (food allergy herbal formula [FAHF]-2) treatment and remained significantly lower as long as 5 weeks post therapy. Splenocytes from FAHF-2-treated mice showed significantly reduced IL-4, -5 and -13, and enhanced IFN- γ production to recall peanut stimulation *in vitro*.

3.3 Fusion proteins

The crosslinking of Fc γ receptor (R)-IIb to Fc ϵ RI on human mast cells and basophils by a genetically engineered Fc γ -Fc ϵ protein (GE2) leads to the inhibition of mediator release on Fc ϵ RI challenge [23,24]. The GE2 protein was shown to inhibit cord blood-derived mast cell and peripheral blood basophil mediator release *in vitro* in a dose-dependent fashion, including the inhibition of human IgE reactivity to cat allergens. The mechanism of inhibition in mast cells included alterations in IgE-mediated Ca²⁺ mobilisation, spleen tyrosine kinase phosphorylation and the formation of downstream complexes. The pro-allergic effects of Langerhan's-like dendritic cells and B-cell IgE switching were also inhibited by GE2. *In vivo*, GE2 was shown to block passive cutaneous anaphylaxis driven by human IgE in mice expressing the human Fc ϵ RI and inhibit skin-test reactivity to dust mite antigen in a dose-dependent manner in rhesus monkeys [24]. The balance between positive and negative signalling controls the reactivity of mast cells and basophils, which is critical in the expression of human allergic diseases. This approach using a human Fc γ -Fc ϵ fusion protein to co-aggregate Fc ϵ RI with Fc γ RII holds promise as a new therapeutic platform for the immunomodulation of allergic

Table 1. Potential immunotherapeutic strategies for the treatment of food allergy.

Therapy	Type of allergy	Route	Immunological mechanism	Risk*
Traditional injection immunotherapy	Oral allergy syndrome	Subcutaneous	Immune deviation from T_H2 to T_H1 , increased IgG-blocking antibodies, decreases specific IgE	Safe when performed properly
Peptide immunotherapy	IgE-mediated food allergy	Subcutaneous	Immune deviation from T_H2 to T_H1	Appears to be safe
Traditional Chinese medicine	IgE-mediated food allergy (asthma)	Subcutaneous	Immune deviation from T_H2 to T_H1	Appears to be safe
Fusion proteins	IgE-mediated (all types)	Subcutaneous	Blocks IgE-mediated signalling	Currently unknown
Mutated protein immunotherapy	IgE-mediated food allergy	Subcutaneous, intranasal and oral	Immune deviation from T_H2 to T_H1 (?)	Appears to be safe
Homologous protein immunotherapy	IgE-mediated food allergy	Subcutaneous, intranasal and oral	Immune deviation from T_H2 to T_H1 (?)	Appears to be safe
DNA immunisation	IgE-mediated food allergy	Subcutaneous and oral	Subcutaneous: immune deviation from T_H2 to T_H1 Oral: increase levels of allergen-specific secretory IgA in gut and increased systemic IgG	Unknown long term
Immunostimulatory sequences	IgE-mediated food allergy	Subcutaneous	Immune deviation from T_H2 to T_H1	Appears to be safe
Anti-IgE therapy	IgE-mediated food allergy	Subcutaneous	Depletes IgE, blocks IgE from binding to high-affinity IgE receptor (FcεRI), downregulates IgE receptor production	Appears to be safe; anaphylaxis and malignancy noted, but not increased

*Most of the risk assessment was carried out in animal studies, but some preliminary studies have been carried out in humans.

Ig: Immunoglobulin; $T_H1/2$: T helper cell type 1/2.

diseases and potentially other mast cell/basophil-dependent disease states. Recently, a chimeric human–cat fusion protein (γ -*Feline domesticus* [GFD]) composed of a truncated human IgG FcγI and the major cat allergen *Feline domesticus* allergen (Fel d 1) was engineered [25]. In contrast to GE2, GFD indirectly crosslinks FcγRIIB and FcεRI through antigen-specific IgE and inhibits basophil and mast-cell reactivity in an allergen-specific fashion. Chimeric human Fcγ allergen proteins such as GFD provide another option for antigen-specific immunotherapy that could be given as a safe series of high-dose injections and represent a particularly powerful approach for the treatment of severe food-induced allergy.

3.4 Mutated protein immunotherapy

With greater understanding of the allergic mechanism and better characterisation of allergens, the authors and their collaborators [4,26] are developing techniques that appear to be both safe and effective in the treatment of food allergy. This type of immunotherapy can be achieved through the modification of the primary amino acid sequences of IgE-binding allergenic epitopes of the major allergens present in foods. Specific IgE binding to food allergens occurs to the linear amino acid sequence, compared with

IgE binding to inhaled allergens, which occurs along typical conformational epitopes [26]. By altering the primary amino acid sequence of the IgE epitopes, researchers have been able to substantially alter IgE antibody binding with the major peanut allergens as well as shrimp allergens, ranging from reduced binding to a complete abolishment of binding. The major peanut allergens, Ara h 1, -2 and -3 have had mutations made to their cDNA and then mutated, less allergenic proteins were expressed [27,28]. However, mutating the IgE-binding sites did not alter the T-cell epitopes. Patients with peanut hypersensitivity had dramatically reduced peanut-specific IgE binding to the mutated peanut protein compared with the wild-type peanut protein. In animal studies with peanut-allergic mice, the mutated protein significantly reduced the amount of peanut-specific IgE antibody produced by the peanut-allergic animal, as well as reduced their clinical symptoms on peanut challenge [26,29]. The treated animals had a reduction in T_H2 cytokines over the course of treatment, with data suggesting an increase in T regulatory cells producing IL-10. It was also demonstrated that the use of an adjuvant with a mutated allergen may increase the likelihood of obtaining the desired immunological effect and clinical protection [30,31].

Designing molecules that have reduced or abolished IgE binding, while preserving their ability to stimulate T cells, should result in the availability of safe and effective therapeutics for the treatment of food allergy. Furthermore, theoretically, the alteration of such molecules in foods could substantially reduce the incidence of food-induced anaphylactic reactions in such sensitised patients. However, this approach is very unlikely because of the large numbers of allergens in each food.

3.5 Homologous protein immunotherapy

Like peanut, soybean belongs to the legume family. This taxonomic kinship is also reflected at the molecular level as their respective seed-storage proteins share significant amino acid sequence and three-dimensional structure homologies. The soybean counterparts of the three major peanut allergens, Ara h 1, - 2 and - 3 are the β -conglycinin, (2S)-albumin and glycinin proteins, respectively. These soybean proteins can be viewed as natural mutants of the peanut allergens and used as desensitising agents in allergen-specific immunotherapy. This innovative concept was tested in a model of peanut-allergic mice [32]. After peanut challenge, peanut-allergic mice that received soybean immunotherapy were equally protected as those that underwent peanut immunotherapy, and their clinical symptoms were significantly reduced compared with the placebo-treated mice. Levels of peanut-specific IgG1 were significantly lowered after soybean treatment compared with peanut treatment. The cellular response to peanut was also downregulated in the soybean-treated group, as demonstrated by diminished lymphoproliferative responses and immunodeviation of the remaining T-cell reactivity toward a more $T_H1/0$ -like response associated with the production of higher amounts of peanut-specific IFN- γ and TNF- α . A closer look at the amino acid sequence homologies between Ara h 2 and the soy (2S)-albumin revealed that most of the amino acid discrepancies were located in regions corresponding to the immunodominant IgE-binding epitopes of Ara h 2 whereas regions corresponding to T cell epitopes were more conserved. This related-species homologue naturally harbours features that are sought when engineering intraspecific mutated allergens for immunotherapy.

If soybean immunotherapy can be effectively used in patients allergic to peanuts, the treatment would provide an immediate and safer therapeutic option for patients with peanut hypersensitivity than is currently available.

3.6 DNA immunisation

Another novel approach to the treatment of food allergy currently under investigation is DNA immunisation [33,34], which employs the subcutaneous injection of a plasmid DNA (pDNA) vector encoding a specific allergenic protein. The pDNA sequence is taken up by antigen-presenting cells (APCs). Once inside the cell, the DNA encoding the allergen is transcribed and translated. The allergen is then presumably presented on the surface of the APC in the context of the

major histocompatibility complex (MHC) to T cells. This endogenously produced allergenic protein or protein fragment induces a T_H1 -phenotypic response with the upregulation of IFN- γ , an increase in IgG2a and suppression of allergen-specific IgE production [35,36]. Oral delivery of DNA immunisations has also been described. It has been utilised as an immunoprophylactic strategy to modulate peanut antigen-induced anaphylaxis [36]. In this model, the oral delivery of DNA complexed to chitosan, a biocompatible polysaccharide, also favoured a T_H1 response and suppressed the T_H2 allergic immune response.

3.7 Immunisation with immunostimulatory sequences

Another DNA immunisation technique under investigation is immunisation with pDNA conjugated to immunostimulatory sequences (ISSs). These ISSs contain unmethylated cytosine and guanine dinucleotide repeat motifs. These CpG motifs stimulate APCs and natural killer cells to secrete IFN- γ and IL-12, cytokines that promote immune deviation toward the T_H1 phenotype and away from the allergic T_H2 phenotype [35]. These ISSs stimulate immune deviation to the T_H1 phenotype when administered in several ways. They can be administered with DNA encoding the allergen (DNA immunisation) [37], alone [38] or conjugated with the allergen. Although the majority of these DNA immunisation techniques have been studied in the mouse model, a recent report describes enhanced immune deviation to the T_H1 phenotype and reduced allergenicity after injection, immunotherapy with the major ragweed allergen, Amb a 1, conjugated to ISS in mice and rabbits, as well as in primates [39].

3.8 Anti-immunoglobulin E immunotherapy

In general, the immune response to allergen immunotherapy is allergen specific, making the treatment of multiple allergies complex and expensive. Often, patients are allergic to more than one allergen. Ideally, it would be preferable to utilise a single therapy to combat the entire allergic mechanism, one therapeutic option that would be effective for multiple inhalant allergies as well as food allergies. One such therapy, already in clinical trials, involves the use of anti-IgE antibodies [40]. IgE exists freely, circulating or attached to mast cells and basophils. When two mast cell-bound IgE molecules are crosslinked by antigen, the cell is activated and vasoactive mediators are released. Anti-IgE, a humanised mouse monoclonal IgG antibody, directed against the human IgE molecule, binds to freely circulating IgE and not to mast cell- or basophil-bound IgE. Once circulating IgE is bound by anti-IgE, it is unable to bind to its specific high-affinity receptor (Fc ϵ RI) on mast cells and basophils. These antigen-antibody complexes are then cleared from the circulation by the reticuloendothelial system without activation of complement. Several clinical trials involving patients with allergic rhinitis [41] and allergic asthma [42] have been completed. Statistically significant reductions in medication use, allergic symptoms and airway hyper-reactivity have been described following anti-IgE treatment. Reductions in total serum IgE levels to < 1% of the pretreatment levels were

noted in addition to downregulation of the high-affinity IgE receptor (FcεRI) [43].

A recent study of 84 patients with peanut hypersensitivity treated with anti-IgE has been completed [40]. A significant number of patients receiving the highest dose of anti-IgE monthly for 4 months had a significant decrease in their clinical symptoms following peanut challenge post treatment. Patients received anti-IgE 150, 300 or 450 mg monthly for 4 months. After treatment, the subjects were subjected to a peanut challenge. At baseline, they tolerated about half a peanut and on the highest treatment dose they were able to tolerate almost nine peanuts. One problem with the study is that 25% of the subjects even in the high-dose group had no change in their tolerance to peanuts after treatment.

Although anti-IgE therapy is expensive and requires frequent administration to maintain the IgE-deficient state, it could theoretically be used quite effectively in conjunction with other therapeutic modalities. For example, pretreatment with anti-IgE could potentially prevent the systemic reactions experienced with peanut immunotherapy [12] while achieving the beneficial effects of desensitisation. This promising technique is currently under investigation in patients who have food allergy.

4. Conclusion

Food allergy affects ~ 4 – 6% of children and 3 – 4% of adults [44]. For those patients and the families of patients who have anaphylactic reactions, food allergy can be devastating. Reactions can range from mild urticarial reactions to severe anaphylactic shock and death from allergenic foods consumed unknowingly. The only preventative measure currently available for food allergy is the strict avoidance of the food in question, which is often very difficult. It is likely that immunotherapy will be available in the near future as a safe and potentially effective therapy for the treatment of food allergy.

5. Expert opinion

New therapies currently under investigation should help the physician greatly improve the care available for food-induced allergic reactions while reducing the risk of anaphylaxis in these patients. The use of the anti-IgE therapy in conjunction with other immunotherapy would probably be the best treatment option for food allergy in the coming years.

Bibliography

Papers of special note have been highlighted as either of interest (•) or of considerable interest (••) to readers.

- SAMPSON HA: Food allergy. Part 1: immunopathogenesis and clinical disorders. *J. Allergy Clin. Immunol.* (1999) 103(5 Part 1):717-728.
- SAMPSON HA: Anaphylaxis and emergency treatment. *Pediatrics* (2003) 111(6 Part 3):1601-1608.
- SICHERER SH, BURKS AW, SAMPSON HA: Clinical features of acute allergic reactions to peanut and tree nuts in children. *Pediatrics* (1998) 102(1):e6.
- BURKS W, LEHRER SB, BANNON GA: New approaches for treatment of peanut allergy: chances for a cure. *Clin. Rev. Allergy Immunol.* (2004) 27(3):191-196.
- NOON L, CANTAB BC: Prophylactic inoculation against hay fever. *Lancet* (1911) 177(4580):1572-1573.
- FREEMAN J: 'Rush' inoculation, with special reference to hay fever. *Lancet* (1930) 215(5562):744-747.
- ASERO R: Effects of birch pollen-specific immunotherapy on apple allergy in birch pollen-hypersensitive patients. *Clin. Exp. Allergy* (1998) 28(11):1368-1373.
- ASERO R: Fennel, cucumber, and melon allergy successfully treated with pollen-specific injection immunotherapy. *Ann. Allergy Asthma Immunol.* (2000) 84(4):460-462.
- BOLHAAR ST, TIEMESSEN MM, ZUIDMEER L *et al.*: Efficacy of birch-pollen immunotherapy on cross-reactive food allergy confirmed by skin tests and double-blind food challenges. *Clin. Exp. Allergy* (2004) 34(5):761-769.
- BUCHER X, PICHLER WJ, DAHINDEN CA, HELBLING A: Effect of tree pollen specific, subcutaneous immunotherapy on the oral allergy syndrome to apple and hazelnut. *Allergy* (2004) 59(12):1272-1276.
- BURKS W, BANNON G, LEHRER SB: Classic specific immunotherapy and new perspectives in specific immunotherapy for food allergy. *Allergy* (2001) 56(Suppl. 67):121-124.
- OPPENHEIMER JJ, NELSON HS, BOCK SA, CHRISTENSEN F, LEUNG DY: Treatment of peanut allergy with rush immunotherapy. *J. Allergy Clin. Immunol.* (1992) 90(2):256-262.
- NELSON HS, LAHR J, RULE R, BOCK A, LEUNG D: Treatment of anaphylactic sensitivity to peanuts by immunotherapy with injections of aqueous peanut extract. *J. Allergy Clin. Immunol.* (1997) 99(6 Part 1):744-751.
- Side effects of traditional immunotherapy.
- ASERO R: How long does the effect of birch pollen injection SIT on apple allergy last? *Allergy* (2003) 58(5):435-438.
- BRINER TJ, KUO MC, KEATING KM, ROGERS BL, GREENSTEIN JL: Peripheral T-cell tolerance induced in naive and primed mice by subcutaneous injection of peptides from the major cat allergen Fel d 1. *Proc. Natl. Acad. Sci. USA* (1993) 90(16):7608-7612.
- 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 Part 1):1623-1628.
- 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.
- Concept of peptide immunotherapy.
- HONG SJ, MICHAEL JG, FEHRINGER A, LEUNG DY: Pepsin-digested peanut contains T-cell epitopes but no IgE epitopes. *J. Allergy Clin. Immunol.* (1999) 104(2 Part 1):473-478.
- GLASPOLE IN, DE LEON MP, ROLLAND JM, O'HEHIR RE: Characterization of the T-cell epitopes of a major peanut allergen, Ara h 2. *Allergy* (2005) 60(1):35-40.

20. LI XM, ZHANG TF, HUANG CK *et al.*: Food allergy herbal formula-1 (FAHF-1) blocks peanut-induced anaphylaxis in a murine model. *J. Allergy Clin. Immunol.* (2001) 108(4):639-646.
21. LI XM, SAMPSON HA: Novel approaches to immunotherapy for food allergy. *Clin. Allergy Immunol.* (2004) 18:663-679.
- Comprehensive review of emerging treatments for food allergy.
22. SRIVASTAVA KD, KATTAN JD, ZOU ZM *et al.*: The Chinese herbal medicine formula FAHF-2 completely blocks anaphylactic reactions in a murine model of peanut allergy. *J. Allergy Clin. Immunol.* (2005) 115(1):171-178.
23. ZHU D, KEPLEY CL, ZHANG M, ZHANG K, SAXON A: A novel human immunoglobulin Fc- γ Fc- ϵ bifunctional fusion protein inhibits Fc- ϵ RI-mediated degranulation. *Nat. Med.* (2002) 8(5):518-521.
24. SAXON A, ZHU D, ZHANG K, ALLEN LC, KEPLEY CL: Genetically engineered negative signaling molecules in the immunomodulation of allergic diseases. *Curr. Opin. Allergy Clin. Immunol.* (2004) 4(6):563-568.
25. ZHU D, KEPLEY CL, ZHANG K *et al.*: A chimeric human-cat fusion protein blocks cat-induced allergy. *Nat. Med.* (2005) 11(4):446-449.
26. LI XM, SRIVASTAVA K, GRISHIN A *et al.*: Persistent protective effect of heat-killed *Escherichia coli* producing 'engineered', recombinant peanut proteins in a murine model of peanut allergy. *J. Allergy Clin. Immunol.* (2003) 112(1):159-167.
- Validates the concept of mutated protein immunotherapy in an animal study.
27. STANLEY JS, HELM RM, COCKRELL G, BURKS AW, BANNON GA: Peanut hypersensitivity. IgE binding characteristics of a recombinant Ara h I protein. *Adv. Exp. Med. Biol.* (1996) 409:213-216.
28. STANLEY JS, KING N, BURKS AW *et al.*: Identification and mutational analysis of the immunodominant IgE binding epitopes of the major peanut allergen Ara h 2. *Arch. Biochem. Biophys.* (1997) 342(2):244-253.
- Initial study mutating peanut proteins.
29. LI XM, SEREBRISKY D, LEE SY *et al.*: A murine model of peanut anaphylaxis: T- and B-cell responses to a major peanut allergen mimic human responses. *J. Allergy Clin. Immunol.* (2000) 106(1 Part 1):150-158.
30. LI XM, SRIVASTAVA K, HULEATT JW *et al.*: Engineered recombinant peanut protein and heat-killed *Listeria monocytogenes* coadministration protects against peanut-induced anaphylaxis in a murine model. *J. Immunol.* (2003) 170(6):3289-3295.
31. FRICK OL, TEUBER SS, BUCHANAN BB, MORIGASAKI S, UMETSU DT: Allergen immunotherapy with heat-killed *Listeria monocytogenes* alleviates peanut and food-induced anaphylaxis in dogs. *Allergy* (2005) 60(2):243-250.
32. PONS L, PONNAPPAN U, HALL RA *et al.*: Soy immunotherapy for peanut-allergic mice: modulation of the peanut-allergic response. *J. Allergy Clin. Immunol.* (2004) 114(4):915-921.
33. SLATER JE, COLBERG-POLEY A: A DNA vaccine for allergen immunotherapy using the latex allergen Hev b 5. *Arb. Paul Ehrlich Inst. Bundesamt Sera Impfstoffe Frankfurt. A M* (1997) 91:230-235 (Discussion 235-236).
34. SPIEGELBERG HL, OROZCO EM, ROMAN M, RAZ E: DNA immunization: a novel approach to allergen-specific immunotherapy. *Allergy* (1997) 52(10):964-970.
35. CHU RS, TARGONI OS, KRIEG AM, LEHMANN PV, HARDING CV: CpG oligodeoxynucleotides act as adjuvants that switch on T helper 1 (T_H1) immunity. *J. Exp. Med.* (1997) 186(10):1623-1631.
36. ROY K, MAO HQ, HUANG SK, LEONG KW: Oral gene delivery with chitosan-DNA nanoparticles generates immunologic protection in a murine model of peanut allergy. *Nat. Med.* (1999) 5(4):387-391.
37. HSU CH, CHUA KY, TAO MH *et al.*: Immunoprophylaxis of allergen-induced immunoglobulin E synthesis and airway hyperresponsiveness *in vivo* by genetic immunization. *Nat. Med.* (1996) 2(5):540-544.
38. BOHLE B, JAHN-SCHMID B, MAURER D, KRAFT D, EBNER C: Oligodeoxynucleotides containing CpG motifs induce IL-12, IL-18 and IFN- γ production in cells from allergic individuals and inhibit IgE synthesis *in vitro*. *Eur. J. Immunol.* (1999) 29(7):2344-2353.
39. TIGHE H, TAKABAYASHI K, SCHWARTZ D *et al.*: Conjugation of immunostimulatory DNA to the short ragweed allergen amb a 1 enhances its immunogenicity and reduces its allergenicity. *J. Allergy Clin. Immunol.* (2000) 106(1 Part 1):124-134.
40. LEUNG DY, SAMPSON HA, YUNGINGER JW *et al.*: Effect of anti-IgE therapy in patients with peanut allergy. *N. Engl. J. Med.* (2003) 348(11):986-993.
- Anti-IgE therapy utilised for the first time in peanut-hypersensitive patients significantly increased their threshold of sensitivity to peanut antigens to a level that translated into at least partial protection against most unintended ingestions of peanut.
41. NAYAK A, CASALE T, MILLER SD *et al.*: Tolerability of retreatment with omalizumab, a recombinant humanized monoclonal anti-IgE antibody, during a second ragweed pollen season in patients with seasonal allergic rhinitis. *Allergy Asthma Proc.* (2003) 24(5):323-329.
42. MILGROM H, FICK RB Jr, SU JQ *et al.*: Treatment of allergic asthma with monoclonal anti-IgE antibody. rhuMAb-E25 Study Group. *N. Engl. J. Med.* (1999) 341(26):1966-1973.
43. SAINI SS, MacGLASHAN DW Jr, STERBINSKY SA *et al.*: Down-regulation of human basophil IgE and FC- ϵ RI α surface densities and mediator release by anti-IgE-infusions is reversible *in vitro* and *in vivo*. *J. Immunol.* (1999) 162(9):5624-5630.
44. SAMPSON HA: 9. Food allergy. *J. Allergy Clin. Immunol.* (2003) 111(Suppl. 2):S540-547.

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