

WHO Position Paper

Allergen immunotherapy: therapeutic vaccines for allergic diseases

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American Academy of Allergy, Asthma, and Immunology (AAAAI)
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Japanese Society of Allergology
National Institute of Allergy and Infectious Diseases (NIAID)
World Health Organization (WHO)

endorsed by

American College of Allergy, Asthma, and Immunology (ACAAI)
International Association of Asthmology (Interasma)

Chairmen

J. Bousquet (France)
R. F. Lockey (USA)
H.-J. Mailing (Denmark)

Panel members

E. Alvarez-Cuesta (Spain)
O. W. Canonica (Italy)
M. D. Chapman (USA)
P. J. Creticos (USA)
J. M. Dayer (Switzerland)
& R. Durham (UK)
P. Dcmoly (France)
R. I. Goldstein (USA), NIAID
T. Ishikawa (Japan)
K. Ito (Japan)
D. Kraft (Austria)
P. H. Lambert (Switzerland), WHO
H. Lgiwenstein (Denmark)
U. Muller (Switzerland)
P. S. Norman (USA)
R. F. Reisman (USA)
R. Valenta (Austria)
F. Valovirta (Finland)
H. Vssel (France)

Contents

Preface

Synopsis

Abbreviations

1. Introduction

2. Standardization, storage, and mixing of allergen vaccines

2.1. Introduction

2.2. Allergen standardization

2.3. Allergen vaccines for immunotherapy

3. Mechanisms of immunotherapy

3.1. Introduction

3.2. Serum antibody concentrations

3.3. Effector cells

3.4. Lymphocyte response

4. Efficacy of subcutaneous immunotherapy

4.1. Introduction

4.2. Objectives

4.3. Immunotherapy with Hymenoptera venoms

4.4. Immunotherapy with inhalant allergens

4.5. Meta-analysis of efficacy of immunotherapy in asthma

4.6. Immunotherapy with mixtures of allergen vaccines in unselected allergic asthmatic children

4.7. Long-term efficacy of immunotherapy

4.8. Compliance to immunotherapy

5. Safety of immunotherapy

- 5.1. Introduction
- 5.2. Risk factors based on nonfatal systemic reactions
- 5.3. Risk factors based on fatal reactions
- 5.4. Risk factors for immunotherapy
- 5.5. Precautions for immunotherapy
- 5.6. Equipment recommended for settings where allergen immunotherapy is administered
- 5.7. Conclusion

6. Other routes of immunotherapy

- 6.1. Introduction
- 6.2. Efficacy and safety
- 6.3. Practical aspects
- 6.4. Conclusions

7. Pediatric issues

- 7.1. Introduction
- 7.2. Advantages of immunotherapy in children
- 7.3. Problems of immunotherapy in children

8. Indications

- 8.1. Relative contraindications
- 8.2. Immunotherapy for Hymenoptera venom sensitivity
- 8.3. Subcutaneous immunotherapy for allergic rhinoconjunctivitis and asthma
- 8.4. Other routes for allergen administration
- 8.5. Indications for immunotherapy in children

9. Costs

10. Future strategies for immunotherapy

- 10.1. Therapeutic vaccines of the future
- 10.2. Novel delivery systems
- 10.3. Nonanaphytactic allergens, allergen fragments, or peptides for active immunotherapy
- 10.4. IgE-binding haptens of major allergens for passive saturation of effector cells and induction of blocking antibodies
- 10.5. Plasmid DNA immunization
- 10.6. Allergen-specific antibodies and antibody fragments for passive therapy in the allergic effector organs
- 10.7. Immunotherapy with humanized anti-IgE monoclonal antibodies or IgE mimotopes
- 11. Clinical research needs

References

Preface

Readers may wonder how the idea to publish standard guidelines for immunotherapy began. Jean Bousquet and Richard Lockey met at a meeting on allergen immunotherapy in Chicago, Illinois, in 1995 where a conversation developed concerning the feasibility of writing international guidelines on allergen immunotherapy. The idea was revisited between Drs. Lockey and Bousquet at the 1996 American Academy of Allergy, Asthma, and Immunology meeting in San Francisco, California. Dr. Bousquet agreed to discuss the idea with colleagues at the World Health Organization (WHO) and various allergy, asthma, and immunology societies throughout the world to determine whether or not they collectively would sponsor such guidelines. They agreed to do so and the idea became a reality.

Drs. Hans-Jorgen Mailing, Lockey, and Bousquet were approved as co-chairs by their respective regional organizations to co-chair the project. Various individuals who are experts on allergen immunotherapy, and from various geographic locations throughout the world, were appointed to serve on the committee. A draft document was circulated to members of the committee prior to the first and only meeting, which took place at the WHO headquarters, on 21–29 January 1997, in Geneva, Switzerland. During the three-day meeting, the committee reached a consensus on the information to be included in the statement and how the group would proceed to finalize and publish the document. The committee unanimously agreed to change the historical terminology of “allergen extract” to the new terminology used in the manuscript, “allergen vaccine”. This decision was made since many allergen vaccines used for allergen immunotherapy are no longer crude extracts but are defined in biological units and/or in micrograms of major allergens.

During the ensuing months, many drafts were circulated among committee members to assure that a consensus was reached on the information contained in the manuscript.

The committee members hope that these guidelines will result in a better understanding of the science and rationale for using allergen immunotherapy as well as improve the safety of such therapy. The document also defines new techniques being developed which may result in better efficacy and less risk for allergen immunotherapy as well as recommends areas of additional and necessary research. The co-chairs thank the various organizations which have sponsored this initiative, the financial support of the allergen vaccine pharmaceutical manufacturers and colleagues who worked diligently and for many hours to complete the manuscript.

Jean Bousquet, MD

Richard E Lockey, MD

Hans-Jorgen Mailing, MD

* Please note: The WHO Position Paper is not a standard of care document for allergen immunotherapy for individual countries. Standards of care on allergen immunotherapy are established by the physician members of the major allergy/immunology organizations of each country.

Except for a few papers which were discussed as pre-prints during the Geneva meeting, scientific articles reviewed for data included in this

Synopsis

- Allergen immunotherapy is the administration of gradually increasing quantities of an allergen vaccine to an allergic subject, reaching a dose which is effective in ameliorating the symptoms associated with subsequent exposure to the causative allergen.
- Controlled studies have shown that allergen immunotherapy is an effective treatment for patients with allergic rhinitis/conjunctivitis, allergic asthma, and allergic reactions from stinging insects.
- The treatment of allergic diseases is based on allergen avoidance, pharmacotherapy, allergen immunotherapy, and education of the patient. Immunotherapy, where appropriate, should be used in combination with all forms of therapy with the goal that the allergic patient will become as symptom-free as medically possible.
- Allergen immunotherapy is indicated for patients who have demonstrated evidence of specific IgE antibodies to clinically relevant allergens. The rationale for prescribing allergen immunotherapy depends on the degree to which symptoms can be reduced by medication, the amount and type of medication required to control symptoms, and whether effective allergen avoidance is possible.
- The response to immunotherapy is specific for the antigen administered. Mixtures of allergens unrelated to the patient's sensitivity should not be utilized.
- Physicians should know of local and regional aerobiology and the exposure of the patient in the home and work environments. Only physicians with training in allergology (allergy/ immunology) should prescribe the clinically relevant vaccine for allergen immunotherapy.
- The quality of the allergen vaccine is critical for both diagnosis and treatment. Where possible, standardized vaccines of known potency and shelf life should be utilized for allergen immunotherapy.
- The use of well-characterized and standardized vaccines makes it possible to define an optimal maintenance dose in the range of 5—20 mg of major allergen per injection for a number of primary allergens. Therapeutic efficacy correlates with such doses.
- The major risk of allergen immunotherapy is anaphylaxis. Therefore, allergen immunotherapy should be administered by or under the close supervision of a trained physician who can recognize early symptoms and signs of anaphylaxis and administer appropriate emergency treatment.
- The optimal duration of immunotherapy is still unknown. Many clinicians advise 3—5 years of therapy for patients who have had a good therapeutic response. However, the decision to discontinue allergen immunotherapy should be individualized.
- Several studies suggest that venom immunotherapy may be discontinued after 3—5 years in most patients. However, the decision to discontinue venom immunotherapy should be individualized.

Abbreviations

- AAAAI: American Academy of Allergy, Asthma and Immunology
- ACAAI: American College of Allergy, Asthma and Immunology
- AU: Allergy Unit
- BAU: Bioequivalent Allergy Unit
- BU: Biologic Unit
- CBER: Center for Biologic Evaluation and Research

EAACI: European Academy of Allergy and Clinical Immunology
ESPACI: European Society of Pediatric Allergy and Clinical Immunology
EU: European Union
IAACI: International Association of Allergy and Clinical Immunology
LU: International Unit
IUIS: International Union of Immunological Societies

1. Introduction

Allergen immunotherapy is the practice of administering gradually increasing quantities of an allergen extract to an allergic subject to ameliorate the symptoms associated with the subsequent exposure to the causative allergen. Allergen immunotherapy was introduced to treat “pollinosis” or allergic rhinitis by Noon and Freeman in 1911 (1). Since then, immunotherapy has been used to treat allergic diseases caused by inhalant allergens and is an effective treatment for patients with seasonal or perennial allergic rhinoconjunctivitis and asthma. Hymenoptera venom immunotherapy used for about 20 years is accepted as the standard of care for Hymenoptera sting-induced systemic allergic reactions.

Vaccines are utilized in medicine as immune modifiers. So too, is allergen immunotherapy. Knowledge gained from studies of allergic mechanisms, such as the importance of Th1 and Th2 cells, cytokine regulation of the immune responses, and specific inhibition or ablation of pathogenic immune responses by means of tolerance induction, may be applicable to a variety of allergic and immunologic diseases. This is especially true for autoimmune diseases such as juvenile diabetes mellitus and multiple sclerosis. Thus, the concepts utilized and the scientific data which support the use of allergen immunotherapy to treat allergic diseases are now being applied scientifically for other immunologic diseases. The panel therefore entitled this position statement “Allergen immunotherapy: therapeutic vaccines for allergic diseases” to indicate that vaccines (allergen extracts) which modify or down-regulate the immune response for allergic diseases are part of this broad-based category of therapies presently utilized and being developed to treat other immunologic and infectious diseases.

Immunotherapy is the only treatment that may affect the natural course of allergic diseases, and it also may prevent the development of asthma in patients with allergic rhinitis. New routes of administration of immunotherapy are currently being explored. Nasal, sublingual, or oral immunotherapy, using high doses of allergen vaccines, may prove to be effective, safe, and easy routes of administration. Moreover, in the future, new technologies and additional knowledge of the basic mechanisms of allergic diseases may completely alter the way allergen immunotherapy is utilized.

Guidelines or indications for immunotherapy with inhalant allergens and venoms have been published within the past years by the World Health Organization (WHO) (2, 3), the European Academy of Allergy and Clinical Immunology (EAACI) (4–6), the International Consensus Report on Asthma (7), the Global Strategy for Asthma Management and Prevention (8), the International Consensus Report on Rhinitis (9), the British Society for Allergy and Clinical Immunology (10), the American Academy of Allergy, Asthma, and Immunology (AAAAI), and the American College of Allergy, Asthma, and Immunology (ACAAI) (11).

These reports provide guidelines for a better understanding of and indications for the use of allergen immunotherapy. However, none of them represent a consensus report of representatives from various parts of the world (2, 4, 10), and some reports address specific issues on one of the target organs in relation to asthma (7, 8) or rhinitis (9).

Therefore, physicians and scientists from various parts of the world convened at the WHO headquarters in Geneva, January 27–29, 1997 to review the science of and indications for allergen immunotherapy. New forms of therapy which are being investigated and under development, and may prove to be safer and more effective, also are discussed.

2. Standardization, storage, and mixing of allergen vaccines

2.1. Introduction

Allergenic extracts (vaccines) have been defined. "Allergenic extract" means a preparation of an allergen obtained by extraction of the active constituents from animal or vegetable substances with a suitable menstruum. 'Allergenic product' means a biologic product, including allergenic extracts and others, that is administered to man for diagnosis, prevention and treatment of allergy and allergenic diseases" (12). In the European Pharmacopoeia, "allergen products are pharmaceutical preparations which are derived from vaccines of naturally occurring source materials containing allergens, which are substances that cause and/or provoke allergic (hypersensitivity) disease. The allergenic components are most often of a proteinaceous nature. Allergen products are intended for *in vivo* diagnosis and/or treatment of allergic hypersensitivity diseases attributed to these allergens" (13).

The committee which met in Geneva decided to use the term "allergen vaccine" rather than allergen extract to indicate that vaccines (allergen extracts) modify or downregulate the immune response for allergic diseases and are part of a broad-based category of therapy presently utilized and being developed to treat other immunologic and infectious diseases.

Successful immunotherapy is dependent on the use of high-quality allergen vaccines that are properly standardized and can be manufactured with consistency. The present WI-JO recommendation on allergen standardization is adapted largely from the approved position statements of the American and European allergy societies (14,15). Approaches to standardization in Europe and the USA have differed in the past but common strategies are being developed. Both the European and the US position statements recommend that all allergenic vaccines be standardized for total allergenic potency, biologic activity, and major allergen measurements in mass units. In the USA, allergenic products are regulated through the FDA; in Europe they are regulated by different member states, though general rules are now evolving as part of the guidelines issued by the European Union (16). From a global perspective, this WHO recommendation encourages the adoption of standardization procedures that can be used and interpreted worldwide. Regulatory agencies should work toward common methods for production and standardization of allergen vaccines.

2.2. Allergen standardization

The most common vaccines used in clinical allergy practice are now available as standardized products or are pending standardization. However, there are several dozen vaccines currently being marketed (many of which are used only occasionally) for which it is neither feasible nor economically possible to standardize. It is proposed that the allergen manufacturers introduce vaccines tested for consistency relative to an in-house reference standard (13). This approach is designed to ensure an acceptable level of standardization and quality control in otherwise unstandardized allergen vaccines.

Details of allergen standardization and calibration are described in the EAACI Allergen Standardization and Skin Test Position Paper (14) and recommendations of North American societies of allergy, asthma, and immunology (15).

2.2.1. Raw material

The allergen raw material should be selected from relevant source material. Detailed instructions for harvesting, storage, extraction, and purification have been described by the IUIS (17), in the Nordic Guidelines (18), and by the EU (Production and quality control of allergens, Note for Guidance 111/927/90). Manufacturing of allergen vaccines should be based on Good Manufacturing Practice (GMP) (13).

2.2.2. Methods of allergen standardization

Allergen vaccine standards. The measurement of major allergens for standardization is now a realistic and desirable goal. A key element in this process is the maintenance of reference standards containing known amounts of relevant allergens. Standards for a number of vaccines have been produced as part of the WHO/IUIS/EAACI allergen standardization program (19–23). Several of these standards, e.g., short ragweed, mite (*Dermaphagoides pteronyssinus*), and dog, contain known amounts of the major allergens (24, 25). The allergen content of some CBER reference preparations has also been determined. Standards with defined allergen content are maintained under stable conditions, in approved repositories, such as the FDA, WHO, Central Bureau für Schimmelsvampes (CBS), or the American Type Culture Collection (ATCC) facilities. In the future, it is likely that recombinant allergens will provide primary standards for allergen analysis and form the basis for

development of new diagnostic and therapeutic products to diagnose and treat allergic diseases (26—28).

Current methods of standardization. Standardization is primarily based on *in vivo* and *in vitro* detection of IgE antibodies to allergens. Skin testing makes it possible to define the allergen vaccine in biologic units. Two methods are commonly used (14, 18, 29). Both depend on the availability of allergic patients and the criteria used to select them (30) (Table 1). The inhibition of the binding capacity of IgE antibodies is measured by RAST-inhibition-derived methods (31). The results reflect measurement of total allergenic potency, and such tests are required by the European Pharmacopoeia (13). Accuracy of these tests depends on the availability of appropriate human sera (32), the composition of the serum pool, and the allergen vaccine used as a reference standard (24, 33). However, there is some variability with these *in vivo* and *in vitro* tests, and it may be difficult to compare total allergenic potency of vaccines produced by different manufacturers.

The composition of the vaccine can be determined by methods such as isoelectric focusing, SDS—PAGE electrophoresis, IgE immunoblotting, and CRLF (cross-radio-immunoelectrophoresis) (for review, see Ref 34).

Table 1. Comparison between skin test methods to standardize allergens

	US method	Nordic Council method
Skin test method	Intradermal	Prick test
Dosage increment	Threefold	1 fl-Fold
No. of patients tasted	15	20
Selection of patients	Highly allergic	Allergic
Reaction measured	Frytheme	Wheel
Histamine reference	No	Yes
Calculation	Arithmetic mean	Geometric mean
Units of standardization	Rioequivalent allergy unit (BAU)	Riologic unit(BU)

New technologies. The rapid development of new technologies for both DNA and protein analysis offers opportunities for improved methods of standardization of allergen vaccines. Many important allergens of pollen, dust mites, animal danders, insects, and foods are cloned and expressed as homogeneous recombinant proteins. Some have comparable allergenic activity to natural protein allergens (for review, see Ref. 35). With these new technologies, an allergen vaccine can be characterized in terms of content of major allergen (ng or µg), and the consistency of each lot can be accurately monitored. Such measurements will facilitate objective comparisons of allergen vaccines (24, 25, 33, 36, 37) by focusing on proteins of established allergenic importance; e.g., Fel d 1, Der p 1, Lol p 1, Amb a 1, and Bet v 1(38, 39). It is important that a consistent relationship exists between the amount of allergen measured and the amounts of other allergens present in the allergen vaccine (40). While monoclonal antibody-based assays are preferred because the reagents are available in large quantities in perpetuity and have defined specificity, tests using polyclonal antibodies may be equally useful. At present, for example, measurements of Fel d 1 form the basis of assigning BAU (bioequivalent allergen units) by the FDA to cat hair and cat pelt vaccines. The amount of Fel d 1 correlates with skin test potency in BAU. Major allergen measurements have been shown to correlate with estimates of biologic potency (40,41). In Japan, allergen vaccines are standardized by quantifying the major allergen and by biologic activity. Vaccines are labeled in JAU (Japanese allergen unit) (42).

The production of recombinant human IgE antibody fragments using combinatorial libraries maybe useful for detecting IgF epitopes in allergen vaccines and thus may also contribute to standardization (43).

2.2.3. Units

Allergenic vaccines have been standardized in different ways (30, 44). however, only those which specify total potency and the concentration of individual allergens should be used for allergy diagnosis and immunotherapy (18).

A critical problem in determining total allergenic potency, whether based on biologic assays or on *in vitro* assays, is that the unitage is arbitrary and may be confusing (PNU, AU, BAU, BU, and LU, as well as other company-related arbitrary units). Therefore, allergen vaccines standardized in BALi in the USA cannot be accurately compared to vaccines marketed in BU (skin test potency relative to histamine) or in LU (WHO/IUIS/IAACI reference preparations) (25, 44). In the USA (1991), the FDA introduced the BAU to replace the AU in order to distinguish potency labeling based on the results of skin testing from that derived only from *in vitro* testing. The 1D50 EAL system for determining BALi is advocated by the FDA and is currently being used to establish USA reference allergen vaccines (29, 45, 46).

2.2.4. Recommendations

Vials should be labeled in accordance with the requirements of regulatory authorities. Labeling includes a designation of relevant units obtained by an approved method. The shelf life or the expiration date must appear on the vial. For standardized allergens, the package insert should state the concentrations of selected individual allergens (marker proteins) in biologic or absolute units and or its potency in biologic units as determined by quantitative skin testing. Recommended therapeutic dosage schedules should also be included. These recommendations are necessary so that physicians can compare vaccines from different pharmaceutical sources. Appropriate methods for storage to maintain stability should be stated on the product inserts.

2.3. Allergen vaccines for immunotherapy

“For immunotherapy, allergen products may be either unmodified vaccines or vaccines modified chemically and/or by adsorption onto different carriers” (13).

2.3.1. Aqueous vaccines

The majority of aqueous allergen vaccines used for immunotherapy are heterogeneous mixtures of allergens and non allergenic materials. These allergen vaccines can be standardized and used for both venom (47—49) and inhalant allergen immunotherapy (50—52).

2.3.2. Depot and modified vaccines

Depot and modified allergen vaccines have been developed in an attempt to make immunotherapy more effective and reduce side-effects. The principle of preparing modified vaccines is to reduce or remove allergenicity, e.g., the capacity to induce IgE-mediated reaction. At the same time, it is desirable to preserve or increase the immunogenicity; e.g., the capacity to modulate the immune system and maintain clinical efficacy. The problems of structurally altering allergens are complex and far from clear.

2.3.3. Types of modification

Physical modification includes absorption and inclusion of allergens as depot vaccines. Aluminium (53), calcium phosphate, tyrosine (54, 55), and liposomes (56, 57) are examples of vaccines used.

Chemical modification refers to the so-called allergoids, such as formaldehyde (58, 59), glutaraldehyde (60), and alginate-modified vaccines (61). Several studies have shown that clinical efficacy is retained by using such modified vaccines and that high-molecular weight preparations were found to be safer than aqueous, unmodified vaccines (60, 62). Other examples are nonpolymerized vaccines, e.g., methoxypolyethylene glycol-modified vaccine (63—67), which were found to be less effective than conventional vaccines.

Combinations of modification. Combinations of physically and chemically modified vaccines include tyrosine-adsorbed, glutaraldehyde modified vaccines (68—70) and aluminium hydroxide-adsorbed formaldehyde vaccines.

2.3.4. Standardization and control of modified allergen vaccine

Preparation of modified allergen vaccines should include

- 1) standardization of the allergen vaccine before it is modified
- 2) the reproducibility of the modification process, so that it is possible to determine the allergenic epitopes retained in the final product
- 3) a consistent reproducible product with the same properties.

2.3.5. Mixtures of allergen vaccines

Allergen vaccines for immunotherapy are prescribed by physicians for patients with proven allergic diseases. When a patient has multiple sensitivities due to related and unrelated allergens, vaccines containing mixtures of these allergens may be prescribed. Two problems may occur with allergen mixtures. First, excessive dilution by multiple allergens may result in suboptimal doses of individual allergens. Second, the potency of individual allergens may deteriorate more rapidly when diluted (71) or mixed with other allergen vaccines (72). This may occur because some allergens possess enzymatic activity which can alter the composition of other allergens (73). Pollen or mite vaccines may undergo degradation when mixed with mite, mold, or cockroach vaccines (72, 74). Ragweed pollen vaccine, however, appears to be particularly resistant to protease degradation (72). Glycerol preservation, but not serum albumin, may prevent some degradation by proteases (72). The relative amount of each component of a mixed allergen vaccine should be indicated.

Related allergens may have epitopes in common, resulting in cross-reactivity. Examples of such interrelated vaccines are those derived from *D. farinae* and *D. pteronyssinus*; from temperate grasses such as *Phleum pratense*, *Lolium perenne*, *Poa pratensis*, *Secale cereale*, etc.; from deciduous trees such as *Alnus glutinosa*, *Betula verrucosa*, *Corylus avellana*, etc.; from *Parietaria judaica* and *P. officinalis*, and from *Ambrosia elatior* and *A. trifida*. Therefore, it may not make any practical difference whether a single vaccine or a mixture of these vaccines is used (75–77).

Careful diagnosis may identify a few dominant sensitizing allergens which can be used for immunotherapy, and avoid potential problems which occur when some vaccines are mixed together.

Allergen vaccines should therefore be distributed as either (1) vaccines from a single source material or (2) mixtures of related, cross-reacting allergen vaccines such as grass pollen vaccines, deciduous tree pollen vaccines, related ragweed pollen vaccines, and related mite vaccines; or (3) mixtures of other allergen vaccines provided that stability data and data on clinical efficacy are available. Where mixtures are marketed, the relative amounts of each component of the mixture should be indicated.

3. Mechanisms of immunotherapy

3.1. Introduction

Hallmarks of human allergic inflammation are the IgE-dependent activation of mast cells and eosinophils and tissue eosinophilia in which cytokines play a major role. Initial studies in mice revealed two distinct CD4⁺ T lymphocyte subsets based on their profile of cytokines (78). Following activation, T helper-1 cells (Th1) produce interferon-gamma (IFN- γ) and interleukin 2 (IL-2), but no IL-4 or IL-5 whereas T helper-2 cells (Th2) cells produce mainly IL-4, IL-13, and IFN- γ , but no IL-2 or IFN- γ . Both subsets produce IL-3 and granulocyte-macrophage colony-stimulating factor (GM-CSF). This functional dichotomy of CD4⁺ T cells was subsequently demonstrated by analysis of T-cell clones obtained from atopic donors, healthy subjects, and patients with infectious diseases (79). IL-4 (80, 81) and the similar recently described IL-13 (82) are important for IgE heavy-chain isotype switching by cells. This process is inhibited by the Th1 cytokine IFN- γ which, in turn, may be induced by IL-12 (83). IL-5 is a major selective growth factor for the terminal differentiation, activation, and persistence of eosinophils (84) in tissues (possibly by inhibiting apoptosis of eosinophils).

Studies have provided insight into the mechanisms of allergen-specific immunotherapy. Earlier work focused on circulating antibody and effector cells. Recent studies suggest that these changes may be secondary to the influence of immunotherapy on T-cell response to allergen. Most work has examined the effect of subcutaneous immunotherapy rather than immunotherapy administered by local routes. Mechanisms are probably hetero-

geneous, depending on the nature of the allergen, the site of the allergic disease, the route, dose, and duration of immunotherapy, the use of different adjuvants, and, last but not least, the genetic status of the host.

3.2. Serum antibody concentrations

3.2.1. Specific IgE

During conventional immunotherapy, serum allergen-specific IgE concentrations initially rise and then gradually fall to baseline levels over months (85). Pollen immunotherapy may result in blunting of the usual seasonal increases in specific IgB (86). During immunotherapy, some studies have found that serum IgE levels increase while basophil histamine release (87) or target organ sensitivity decreased at the same time. These effects may be related to the differences in molecular characteristics of IgB-dependent histamine-releasing factor (88) or in different IgB isoforms (89), which may have different physiological properties.

3.2.2. Specific IgG

Two opposed modes of action have been attributed to IgG in immediate-type allergy (90). A small fraction of IgG may have anaphylactic properties, although this property cannot be attributed to IgG4. Furthermore, allergen-specific IgG1 and IgG3, but not IgG4, induce eosinophil degranulation via the Fc γ R1 receptor (91).

IgG antibodies induced by immunotherapy may act as allergen-blocking antibodies (92, 93). These observations suggest the so-called "blocking antibody" theory (94, 95) which postulates that IgG competes with IgB for allergen binding, thereby blocking IgB-dependent activation of mast cells. Recently, human monoclonal IgG antibodies from an immunized birch pollen allergic patient were shown to block IgE binding to the major birch-pollen allergen, Bet v 1, and block Bet v 1-induced histamine release (96). However, changes in antibody concentrations are unrelated to the clinical response to immunotherapy with inhalant allergen vaccine (97, 98). Immunotherapy using "rush" protocols is effective long before any changes in antibody synthesis can occur. With venom immunotherapy, an early increase in IgG antibody levels is associated with protection against insect sting in a population of patients but has no predictive value in individual patients (95, 99, 100). With long-term venom immunotherapy, there appears to be a late onset, non-IgG-mediated mechanism which suppresses allergic sensitivity (101).

IgG subclasses may have differential effects on the allergic response. Many studies have shown that immunotherapy induces marked rises in allergen-specific IgG, particularly IgG1 and IgG4 subclasses (102). Resting IgG1 antibody levels, but not IgG4 antibody levels, were predictive of the development of the late response after allergen provocation (103). A high IgG4 antibody level is associated with failure of immunotherapy with inhalant allergens (102).

The role of IgG, in particular tissue or mucosal secretion of antibodies, needs further study.

3.3. Effector cells

Immunotherapy may act by reducing inflammatory cell recruitment, activation, or mediator release. Immunotherapy in mite sensitive children results in a decrease in mast cells in nasal brushings (104,105). Grass pollen immunotherapy in adults is associated with a decrease in cutaneous mast cell numbers, including both "connective tissue" (tryptase and chymase containing) and "mucosal" (tryptase only) mast cells (106) as well as a reduction in histamine and POD, levels in nasal secretions after allergen challenge (107). In ragweed sensitive patients, in a dose and time-dependent fashion, conventional immunotherapy inhibits immediate release of mast cell mediators (11) (8) and eosinophil numbers in nasal lavage in response to allergen provocation (109, 110). Birch pollen immunotherapy inhibits the seasonal increase in bronchial responsiveness to histamine produced mainly IL-4, IL-13, and IL-5, but not IL-2 or IFN- γ . Both subsets produce IL-3 and granulocyte-macrophage/colony-stimulating factor (GM-CSF). This functional dichotomy of CD4⁺ T_H cells was subsequently demonstrated by analysis of I-cell clones obtained from atopic donors, healthy subjects, and patients with infectious diseases (79). IL-4 (80, 81) and the similar recently described IL-13 (82) are important for IgB heavy-chain isotype switching by cells. This process is inhibited by the Th1 cytokine IFN- γ which, in turn, may be induced by IL-12 (83). IL-3 is a major selective growth factor for the terminal differentiation, activation, and persistence of eosinophils (84) in tissues (possibly by inhibiting apoptosis of eosinophils).

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3.4. Lymphocyte response

Immunotherapy may act by modifying the T cell response to a subsequent natural allergen trigger. It is logical that successful immunotherapy might be associated with a shift in IL-4/IFN- γ production either as a consequence of downregulation of Th2 responses or increased Th1 responses (115). There is now good evidence that this may indeed occur. Studies of allergen-induced late responses in the skin (116) and nose (117) indicate that immunotherapy results in a decrease in CD4⁺ cell recruitment and a reduction in local eosinophilia. These changes are accompanied by increases in a subpopulation of CD4⁺ cells expressing IFN- γ transcripts after allergen provocation, whereas the number of cells expressing mRNA for IL-4 and IL-5 remained unchanged. In the target organ, these late increases in IFN- γ cells out of season correlate closely with the clinical response to immunotherapy, measured by seasonal symptoms and medication requirements, suggesting that these upregulated Th1 responses may be "protective" rather than simple bystander events (117). Studies of late cutaneous biopsies suggest that these responses may be amplified and sustained by local production of IL-12, a potent inducer of Th1 responses. The cell source of IL-12 is the tissue macrophage (CD68⁺ cells). There is a reciprocal association between IL-12 cells and IL-4 cells and a positive association with IFN- γ cells which supports that the IFN- γ responses may be IL-12 driven (118). An alternative explanation of these observed increases in IFN- γ cells may be the generation of allergen specific CD8⁺ T cells (119). Increases in CD8⁺ T cells have been observed in tissue (116) after conventional immunotherapy. Studies of T cell lines (allergen specific polyclonal Th1 cells) and clones provide further support for the

idea of a shift in T cell responses. A decrease in IL-4 and increase in IFN- γ after immunotherapy occurs in bee venom sensitive patients in a time-dependent fashion up to 8 weeks after a rush protocol (120). A decrease in IL-4 production by Th1 cell lines (but no change in proliferative response or IFN- γ production) was found in grass and mite sensitive patients after immunotherapy (121). IFN- γ increase and IL-4 decrease were found in supernatants of peripheral blood mononuclear cells after venom immunotherapy (122).

The mechanism of this "switch" is a matter of current debate. Factors determining Th1 and/or Th2 responses include the nature of the antigen (allergen), the allergen dose (123) and nature of antigen (allergen)-presenting cell. Low-dose allergen presentation by B cells or dendritic cells favours Th2 responses, whereas high-dose allergen processing and presentation by macrophages favours Th1 responses. Use of different adjuvants and allergen modification may be important. A current controversy is whether this shift occurs as a consequence of allergen-specific immune unresponsiveness of Th2/Th0 cells "anergy" (124, 125) or is due to upregulation of a distinct subset of Th0/Th1 cells ("immune deviation") (120).

As mentioned above, studies in tissues suggest that immune deviation may be more relevant. The T cell surface marker CD28 is downregulated after anergy induction. Studies of CD28 expression by peripheral blood mononuclear cells after venom immunotherapy have not identified a downregulation of this marker, again consistent with immune deviation (126). However, a study demonstrated a decrease in allergen (PLA2)-specific proliferation and reduced production of both IFN- γ and IL-4 *in vitro* by T cell lines after venom immunotherapy. These responses are allergen-specific and reversible by addition of either IL-2 or IL-15, providing the first evidence of "anergy" after immunotherapy in humans (127). However, the mechanisms of venom immunotherapy in nonatopic subjects may be different from the mechanisms of immunotherapy with inhalant allergens in atopic patients.

4. Efficacy of subcutaneous immunotherapy

4.1. Introduction

Studies to assess the efficacy of immunotherapy fulfilled the following criteria:

- 1) *double-blind, placebo-controlled randomized study*
- 2) *study published in English as a full paper in a peer-reviewed journal*

Table 2. Dose of major allergen required to achieve clinical efficacy

Allergen source Dose p.g/l	Major allergen	Reference	
Cat 16	Fel d1	Taylor et al.	140
		Obmar et al.	141
		Sundir et al.	142
8—16		Alvarez-Cuesta et al.	144
15			
13			
<i>Datmatophagoides pteronyssinus</i> 0.5—11.5	Der p 1	Wahn of al,	138
		Haugaard et al.	137
		Bousquet et al.	50
5			
Short-ragweed pollen 2—19	Amb a 1	van MEtre et al.	131
		van Metre et al.	185
		Creticns et al.	138
4—47			
12—24		Creficos etal.	145
10			
Grass (timothyj pollen 25—41	PHI p 5	Dsferballe	128
	PHI pG	flsterballe	128
13—20			
Vespu/a venom 5	yes g 5	Hunt et al.	48
		MOlleretal.	147
5			
	Api ml	Hurtetal.	48
12			
		Muller of ci.	147
12			
	flolmS	HuntetaL	48
3			
	DoleS	Huntetal.	48
3			

- 3) *patients selected* according to well-defined clinical criteria and a specific allergy diagnosis
- 4) *allergen vaccines defined*. If possible, the vaccine was standardized and the dose(s) or major allergen defined. This latter requirement is not applicable for most studies carried out before 1990. One study found that for effective grass pollen immunotherapy, the vaccine should contain all the active proteins, and not only major allergens (128). However, for ragweed-pollen immunotherapy, a single major allergen, Amh a 1, was found to be equally effective, as was the whole ragweed-pollen vaccine in one study (129), but not in another one (130).
- 5) *an optimal maintenance dose*. Low dose immuno therapy is usually ineffective(131 —133), and high doses of allergen vaccines may induce a high and unacceptable rate of systemic reactions. Thus, optimal doses using vaccines labeled either in biologic units (134, 135) or in mass of major allergens (40) have been proposed. 'The optimal dose is defined as the dose of allergen vaccine inducing a clinically relevant effect in most patients without causing unacceptable side-effects (136, 137). The optimal dose should be the target for maintenance dose for all patients (134). Quantization of major allergens may be used to define allergen doses for effective immunotherapy (Table 2). There is a good evidence from immunotherapy studies with ragweed,

grass, mite, cat, and venom allergens that a maintenance dose of 5—20 gg of major allergen per injection is associated with significant improvement in patient symptom scores (25, 108,136—147). Systemic side-effects in some patients will require adjustment of this goal.

6) *A sufficient duration of treatment.* The efficacy of immunotherapy is related to the duration of the treatment in some studies (148,149), although efficacy is often demonstrated during the first year of treatment (51, 150).

7) *Reported data of the clinical efficacy.*

Immunotherapy is specific for the antigen administered (75, 151) and requires a complete allergy evaluation before it is initiated. Since allergens interact with nasal, bronchial, and ocular mucosa, it seems appropriate to consider the efficacy of immunotherapy by allergen species rather than by a specific allergic disease.

4.2. Objectives

4.2.1. Immunotherapy as a curativetreatment

The treatment of allergic diseases combines immunologic and pharmacologic therapy. In many patients, medications can relieve allergic symptoms without causing side-effects. The differences between pharmacologic and immunologic treatments of allergic diseases are not restricted to safety and efficacy. Drugs provide symptomatic relief whereas allergen avoidance and immunotherapy are the therapeutic modalities which have the potential to modify the natural course of the disease.

Perennial rhinitis and asthma are multifactorial and complex diseases in which allergic factors and nonallergic triggers interact and result in chronic inflammation. The role of inhalant allergens in exacerbations of rhinitis and asthma has been demonstrated. Inhalation of allergens leads to nasal and bronchial inflammation. Two different situations may exist (152). Exposure to pollens is usually self-limited because various species pollen-ate only for a defined period. Pollen induced allergic reactions may lead to a transient nonspecific bronchial hyperreactivity and nasal hyperreactivity which persist for days or weeks after a specific pollinating season. However, domestic mites and other perennial allergens to which there is a continuous exposure, may induce persistent inflammation and nonspecific hyperreactivity of the nose and the bronchi. Patients with chronic asthma develop airways remodeling, which in some patients results in irreversible airflow obstruction (153).

These considerations suggest that immunotherapy may be more rapidly effective for patients who are allergic to seasonal allergens than in those who are allergic to perennial allergens and have persistent disease. These patients may have permanent airways abnormalities which cannot be reversed by immunotherapy.

The major objectives of immunologic treatment are, in the short term, to reduce responses to allergic triggers which precipitate symptoms, and eventually, to decrease the inflammatory response and to prevent the development of persistent disease.

4.2.2. Immunotherapy as a preventivetreatment

At present, allergen avoidance and immunotherapy are the only treatments that modify the course of an allergic disease either by preventing the development of new sensitivities (154) or by altering the natural history of disease or disease progression (see chapter 7).

4.3. Immunotherapy with Hymenoptera venoms

4.3.1. Efficacy

Venom immunotherapy with purified venoms is effective treatment for the vast majority of venom allergic patients (48, 155—158). Patients sensitized to honeybees may be less well protected than those allergic to *Vespula* (158, 159).

Many treatment schedules have been proposed aiming for satisfactory protection, minimal side-effects, and

optimal convenience (5, 147, 160, 161). The maintenance dose may be reached within 2–3 h in ultra rush protocols to four to six weeks with more conventional schedules. Ultra rush protocols are usually well tolerated (161,162). More systemic side-effects have been reported with honeybee venom immunotherapy than with *Vespula* venom immunotherapy (5, 49, 163). When the maintenance dose is reached, injections are given every one to 2 months (164).

4.3.2. Duration

Loss of venom skin test sensitivity, although unusual even during prolonged venom immunotherapy, is generally considered a safe criterion for stopping venom immunotherapy (6, 101, 165). Most patients remain protected when restung after stopping venom immunotherapy of 3–5 years duration despite the persistence of positive venom skin tests (159, 166). Some patients, especially those with a history of a severe reaction before immunotherapy and who have systemic allergic side-effects from immunotherapy may be more prone to develop systemic field-sting reactions after discontinuation of venom immunotherapy (6, 101).

4.4. Immunotherapy with inhalant allergens

4.4.1. Immunotherapy for pollen allergy

The efficacy of pollen immunotherapy is suggested by the decrease of target organ sensitivity when comparing pre- and post-treatment of nasal, bronchial, and/or conjunctival allergen challenge (for review, see Ref 167).

Efficacy of immunotherapy has been scientifically documented in the majority of optimally designed double-blind, placebo-controlled trials in the treatment of *rhinitis* due to grass (51, 59, 98, 107,168–179) (Table 3), ragweed (68,129,131, 133, 150, 180–188) (Table 4), *Parietaria* (189, 190), mountain cedar (191), and coconut tree pollen (192). Controlled studies in children have confirmed the efficacy of immunotherapy in pollen induced allergic rhinitis (105, 193). In one study, perennial immunotherapy appeared to be more effective than preseasonal treatment (194).

Immunotherapy with grass and ragweed pollen vaccines is effective to treat allergic *conjunctivitis* (51, 59, 63, 168).

Controlled studies have investigated the efficacy of immunotherapy in *pollen asthma* (Table 5). Some studies demonstrate that improvement of the PD₂₀FEV₁ after allergen bronchial challenge decreases in patients receiving immunotherapy (172, 175, 195). Double-blind, placebo-controlled studies using aqueous, standardized vaccines or formaldehyde-allergoids have shown that immunotherapy has a beneficial effect on bronchial symptoms and/or decreases the needs for asthma medications (59, 107, 145, 148, 169, 173, 174, 176, 178, 189, 191, 196–198). Two studies failed to demonstrate efficacy (199, 200), one of them possibly because most of the subjects were also allergic to molds (199).

Double-blind, placebo-controlled trials of immunotherapy with vaccines of other pollen species have not been published. Although it is postulated that immunotherapy is effective with such pollen vaccines (3), proper trials remain to be done.

Studies comparing the efficacy of immunotherapy and pharmacotherapy are needed. One such study (70) compared the efficacy of a topical steroid with Pollinex~ vaccine in the treatment of ragweed hay fever and concluded that the pharmacologic treatment was superior in efficacy and safety however, this study is flawed because it did not use the full schedule of immunotherapy, and Pollnex vaccine is not very effective for treatment of ragweed pollen allergy (68).

Polysensitized patients The IgE antibody response to environmental allergens is highly heterogeneous. For example, patients allergic only to grass pollens differ clinically and immunologically (201, 202) from those allergic to multiple pollen species. A double-blind, placebo-controlled study which compared the efficacy of immunotherapy in patients allergic to grass or multiple pollen species indicated that grass-pollen-allergic patients but not the polysensitized patients were significantly improved with immunotherapy (107). In clinical practice, the majority of patients receive multiple allergen immunotherapy. This issue needs further study

Patients sensitized to certain pollen species often have the oral allergy syndrome due to cross-reactive epitopes in raw fruits and vegetables (203–207). In theory immunotherapy with birch pollen or other pollen species may decrease these food-allergy symptoms. Double-blind, placebo-controlled studies have not been carried out. Two studies have been reported, but the number of patients was small and effectiveness usually not demonstrated (208) except in a single case report (209).

4.4.2. Immunotherapy for domestic-mite allergy

Immunotherapy with mite vaccines is more effective than crude house-dust vaccines (210). Crude house-dust vaccines should not be utilized.

In most, but not all studies of bronchial challenge with domestic mite (*D. pteronyssinus* and/or *D. farinae*) vaccines, after immunotherapy the threshold dose eliciting an immediate bronchial obstruction was increased and the late-phase reaction was inhibited (50, 54, 138, 149, 211—215). These studies suggest that immunotherapy is effective and may decrease inflammation since the late-phase reaction was decreased.

Immunotherapy was shown to reduce symptoms and/or the need for asthma medications in some studies, especially in children, (54, 212, 216—221), but in other studies the results were inconclusive (222—224) (Table 6). A single study has examined the effect of immunotherapy with storage-mite

Table 4. Results of double-blind, placebo-controlled studies in ragweed-pollen rhinitis

Reference scores	Patient Number			Extract	Schedule	Protocol	Symptom-medication		
	A	P					Dose	Duration	
Arbesman & Reisman 180	19	18		Repository	12 injections	Preseason	IV: 4000 PNU	2 years	NS vs
placebo									
Cockcroft et al.	68	22	21	Aqueous Pollinex®	12 injections	Preseason	IV: 10000 PNU	2 years	Improved
Improved: A: 67%,					4 injections	Preseason	M: 4000 Noon U	1 year	
Grammer et al.	181	21	19	Polymer glutaraldehyde	Classical	15 injections	M: 6250 PNU; C: 50000 PNU	15 weeks	P: 38% P~<0.02
Hirsch et al.	132	81	74	Aqueous	Rinkel	Pre- and	M: 27—41 PNU;	2 years	NS for
ragweed						coseason	0.1—0.15 gg AgE		
Uchtenstein et al.	182	24	24	AgE	Classical	Pre-season	C: 17—800 .g	1 year	P<0.01
Lichtenstein at al.	150	18	30	AgE	Classical	Preseason	1.0mg	1 year	P<0.01
				AgE plus K	Classical	Preseason	1.4mg	1 year	P<0.01
				Crude ragweed, aqueous	Classical	Preseason	C: 8800 PNU	1 year	P<0.01
Lowell & Franklin	183	12	12	Aqueous	Classical	Preseason	Not stated	1 year	P'<fl.OI
Meriney et al.	184	10	10	Formald. allergoid	Classical	Preseason	C: 10710 PNU	20 weeks	P<0.01
van Metre et al.	131	12	12	Aqueous	Rinkel	Pre- and	C: 94 ng AgE	3 months	NS
van Metre et al.	185	15	17	Aqueous	Classical	Pre- and	C: 70kg AgE	1 year	P<D.01
						c o s e a s o n			
						c o s a s o n			
						c o s e a s o n			
Norman et al.	129	21	21	Aqueous whole ragweed	Classical	Preseason	C: 8483 PNU per year	4 years	years and
4: PcO.02									
Years Sand 4: P<0.04				AgE	Classical	Preseason	C: 185530 PNU per year	4 years	
Norman & Lichtenstein 187	20	21		Alum-precipitate	Classical	Preseason	C: 13746 (PNU (year 1 3 years		P.<0.006
Norman et al.	188	22		Formald. allergoid	Cluster	Preseason	C: 63600 PNU (year 1) 2years		P-cOOI
				Aqueous	Classical	Preseason	C: 2000 PNU (year 1) 2 years		P.<0.01

M: maximal dose; C: cumulated dose; P: placebo treatment; A: active treatment.

(*Lepidoglyphus destructor*) vaccine and found that it was clinically effective (225).

A large controlled study addressed the issue of the most appropriate group of candidates for mite immunotherapy (226). Two hundred and fifteen patients were enrolled and were followed up for 1 year with symptom-medication scores and assessment of pulmonary function. Patients who had other perennial allergies, or aspirin intolerance and/or chronic sinusitis did not improve. Among the patients allergic only to *D. pteronyssinus*, children had a significantly greater improvement than adults. Patients with irreversible airflow limitation (FEV₁ under 70% of predicted values after an adequate pharmacologic treatment) did not benefit from immunotherapy.

Double-blind, placebo-controlled studies with domestic mite vaccines showed that immunotherapy was effective in alleviating symptoms of perennial allergic rhinitis (55, 61, 216—218, 227—230) (Table 7).

4.4.3. Immunotherapy for animal protein allergy

A number of studies have demonstrated significant improvement in bronchial sensitivity in patients with cat-allergic asthma after cat vaccine immunotherapy (52, 140—142, 231—238) (Table 8). Three studies have confirmed the clinical efficacy of cat immunotherapy, showing improvement in symptoms (141, 143, 144) and reduction of medication needs (144) in patients who kept their animal at home.

4.4.4. Immunotherapy for mold allergy

Mold allergens often cause rhinitis and asthma. Multiple mold allergy is often present. The quality of mold vaccines available in the past was often poor (239). However, immunotherapy with standardized *Cladosporium* and *Alternaria* vaccines was found to be effective in rhinitis and/or asthma in three studies (240—242).

4.4.5. Immunotherapy with other vaccines

Efficacy of house-dust vaccine immunotherapy is doubtful, and the characterization of these vaccines is poor. Double-blind, placebo-controlled studies of immunotherapy with bacterial vaccines for treatment of rhinitis and/or asthma did not show efficacy (for review, see Ref 243). There are no studies of immunotherapy with *Candida albicans* and *Trichophyton* and the characterization of these vaccines is usually poor.

Table 5. Double-blind, placebo-controlled studies in pollen asthma*

Reference	Species	Patient number		Extract	Duration	Symptom and medication scores
		A	P			
Armentia-Medina et al. 198	Bermuda grass	19	11	Standardized	1 year	P-c0001
Bousquet et al. 69	Grass	18		Standardized	1 year	P-c001
	Grass	15	14	Formald. allergoid	1 year	P~z001
Bousquet et al. 168	Grass	13		HMW-allergoid	1 year	P~<001
	Grass	39	18	HMW-allergoid	1 year	PO.01
Creticos et al. 145	Ragweed	40	37	Standardized	1—2 years	0: NS to PO01
Dolz et al. 148	Grass	14	14	Standardized-alum	3 years	P.<0001
Frankland & Augustin 169	Grass	50	50	Pollaccine	1 year	P-cDO01
		50	50	Purified allergen		P-c0001
Hill et al. 200	Grass	11	9	Aqueous	2 years	NS
McAllen 172	Grass	47	23	Allpyra®	1 year	NS
		40		Depot allergen	1 year	P=005
Macbiels et al. 173	Grass	18	18	Der p-immune complexes	1 year	P,c00C1**
Macbiels et al. 174	Grass	12	37	Der p-immune complexes	1 year	P=00~2
Ortolani et al. 175	Grass	8	7	Standardized	1 year	P-c001
Pastorello et al. 176	Grass	10	9	Allergoid	1 year	Pc005
Varney et al. 178	Grass	20	20	Standardized-alum	1 year	Improvement

HWM: high molecular weight; 0: drug score; A: active treatment; P: placebo treatment; Der p: *Dermatophagoides pteron-ssinus*.

* All studies were not specifically designed for pollen asthma, but bronchial symptoms have been reported.

**First part of pollen season.

4.5. Meta-analysis of efficacy of immunotherapy in asthma

A meta-analysis of clinical trials of allergen (2.2—4.9). The odds for reduction in medication after immunotherapy was undertaken to assess the efficacy of mite immunotherapy was 4.2 (95% CI 2.2—7.9). The efficacy of this form of therapy in asthma (244). A combined odds for reduction in nonspecific bronchial hyperresponsiveness in a computerized bibliographic search revealed 20 randomized, placebo-controlled, double-blind trials of The mean effect size for any allergen immunotherapy was 6.8 (95% CI 3.8—12.0).

immunotherapy for treatment of asthma. therapy on all continuous outcomes was 0.71 (95% Criteria analyzed included asthmatic symptoms, CI 0.43—1.00), which would correspond to a mean medication requirements, and measurement of lung 7.1% predicted improvement in FEy₁ from function and nonspecific bronchial hyperreactivity. immunotherapy Although the benefits of allergen Categorical outcomes were expressed as odds ratios immunotherapy could be overestimated because of

Table 6. Double-blind, placebo-controlled studies in mite asthma

Reference	Patient number		Allergen	Extract	Duration	Symptoms	
	A	P					
BPT							
Bousquet et al.	50	20	10	Der p	Aqueous standardized, rush	7 weeks	P001, epr
and lpr							
DSouza et al.	216	46	45	Oar p	Aqueous	1 year	P=O02 Improved
Franco et al.	219	24	25	Oer p	Alum-standardized	15 months	NS?
Gaddie et al.	222	20	25	Oep p	Tyrosine-adsorbed	1 year	NS
Machick et al.	212	24	11	Oerp	Oer p-immune complexes	1 year	P-cOCCi PcOO5
Marques & Amara-Avila	218	16	12	Oer p	Tyrosine-adsorbed	1 year	Improved
Newton et al.	224	7	7	Oer f	Alum-precipitated	15 months	NS PcOCO5
Olsen et al.	220	17	6	Oar p or Oar f	Alum-standardized	1 year	POO1 P-cOOS
							CS: &005
Pauli et al.	223			Oar p	Tyrosine-adsorbed	1 year	NS
Picbler et al.	221	16	14	Oer p plus Der t	Alum-standardized	1 year	P001
Metbact,oline: P-cOCOS							
van Bayer & Stevens	213	9	9	Oer p	Aqueous standardized, semirusb	1 year	epr: P>04;
pr: PcO.O2							
Warner et al.	54	27	24	Oer p	Tyrosine-adsorbed	1 year	Improved,
pr							

Ocr p: *Berm atopbagoides pteron-sinus*; Oer f: *B. faunae*; epr: early-phase reaction; pr: late-phase reaction; BPT: bronchial provocation test with allergen; CS:

inhaled corticosteroids; A: active treatment P: placebo treatment. Intragroup P value.

Table 7. Results of double-blind, placebo-controlled studies in house-dust-mite rhinitis

Reference challenge	Patient number		Extract	Duration	Symptom-medication scores	Nasal
	A	P				
Blainey et al.	55	17	18	Tyrosine-adsorbad	14 months	7
Improved						
Corrado et al.	61	33	33	Conjuvac ⁵	2 years	PcOOO1
DSouza et al.	216	48	48	Aqueous	12 injections	P-cOOI
Improved for A and N* P-cO025						
Ewan et al. 228		16	19	Alum-standardized	3 months	P-cool P<00s
Gabriel et al. 217		33	33	Aqueous	1 year	Improved NS
McHugh et al. 227		30	30	Alum-standardized	1 year	NS (3monthsj P<O,O5~(3 montbsjPcOOI (12months NSjT2 months
			20	Alum-pyridine jAPj	NS (Sand 12 months	P-cO05*~(12 months
Pichler et al. 221		16	14	Alum-standardized	1 year	PcOOO6\$, P':004\$\$

A: active treatment P: placebo treatment IV: maximal dose.

* A: asthma; N: nasal symptoms,

-- Statistioal difference from baseline and placebo.

~ Statistical difference from baseline only.

Pvalue: 5: intragroup, \$5: intergroup.

unpublished negative studies, an additional 33 such studies would be necessary to overturn these results.

vaccines in unselected allergic asthmatic children

A study reported a double-blind, placebo-controlled trial of immunotherapy for treatment of mild to severe asthma in a nonselected population of allergic children (245). The children were closely supervised and given optimal medical therapy. The results showed no significant difference between the placebo and the active-treatment groups. However, there are several factors which may have led to the negative results. Moderately severe asthmatics who receive optimal treatment may not show any additional benefit from immunotherapy.

4.7 *Long-term efficacy of immunotherapy*

The initial studies of treatment of allergic rhinitis did not show that after cessation immunotherapy efficacy was prolonged (129, 183). The results of more recent studies show that the effect of immunotherapy for grass (114, 246), tree (247), or ragweed pollen allergy (248, 249) lasted several years after it was discontinued. These differences may be due to greater doses of vaccine administered in later studies. When there are relapses, the immunologic memory persists and these patients may be good responders to a new immunotherapy regime (250).

In a double-blind, placebo-controlled study of mite-sensitive children, most children who received placebo treatment after one year of active treatment relapsed within months, whereas those in the active treatment group had a persistent effect of immunotherapy (251). With immunotherapy with a standardized house dust mite vaccine administered for one to up to 6 years, it was found that immunotherapy was more effective after it was discontinued if it had been administered for at least 3 years (113). In this study, the effect of immunotherapy on the reduction of the skin test end points at the end of

Table 8. Double-blind, placebo-controlled studies in animal-dander asthma

Reference	Patient number						Symptoms	challenge
	A	P						
				Bronchial				
NS reactivity								
Taylor et al.	140	5	5	Cat	Standardized?	3—4 months		>10
Alvarez-Cuesta et al.	144	14	14	Cat	Aqueous standardized	12 months	P<0.001	>c3.4±2.5
NS								
Haugaard & Dahl	143	15	9	Cat, dog	Standardized, alum	12 months	Cat: improved	x4.5
Improved								
Ohman et al.	141	9	8	Cat	Aqueous standardized	4 months	NS	NS
hIS								x1.4
Sundin et al.	142	15	17	Cat dog	Standardized, alum	12 months	Cat improved	Cat xli
Improved								
		7					Dog improved	Dog xS
hIS								
Valovirta et al.	236	15	12	Dog	Alum-adsorbed	12 months		NS
van Metre et al.	238	9	13	Cat	Standardized	12 months		x2.8
hIS								

A: active treatment P: placebo treatment NS: not significant.

Table 9. Grading of systemic reactions (from Mailing & Waeke [4])

I Nonspecific reactions	Reactions probably not IgE-mediated; i.e. discomfort, headache, arthralgia, etc.
21 Mild systemic reactions	Mild rhinitis and/or asthma (PEFR over 60% of predicted or of personal best values) responding adequately to antihistamines or inhaled β ₂ -agonists

3) Non-life-threatening systemic reactions
urticaria, angioedema, or severe asthma (PEER under 60% of predicted or of

4j Anaphylactic shock
Rapidly evoked reaction of itching, flushing, erythema, bronchial obstruction. etc. requiring intensive treatment

the treatment was correlated with the duration of efficacy after immunotherapy cessation.

Efficacy of a 3-year course of animal dander immunotherapy was assessed 5 years after it was discontinued. One third of the subjects continued to have increased tolerance to cat exposure (252).

4.8. *Compliance to immunotherapy*

Compliance to any treatment administered for asthma or rhinitis is frequently low (253—260) and can preclude the efficacy of the treatment. Every effort should be made to educate the patient in order to improve compliance.

5. Safety of immunotherapy

5.1. *Introduction*

There are several types of reactions that occur with allergen immunotherapy, local and systemic.

Local reactions occur at the injection site. They can be divided into reactions that occur within 20—30 mm and those that occur later than 30 mm after an injection. Local reactions can cause patient discomfort. Therefore, adjustments in vaccine dosage may be necessary when such reactions occur.

Subcutaneous nodules, which occur at the site of injection, are more common with aluminium-adsorbed vaccines (261). They may persist but normally disappear and do not necessitate an adjustment in the immunotherapy dose. Aluminium-free preparations should be used in patients in whom these nodules develop (262) and persist.

Systemic reactions are reactions characterized with generalized signs and/or symptoms occurring distant from the injection site. Such reactions usually begin within a few minutes after the injection and more rarely after 30 mm. The grading of the severity of systemic reactions has been proposed in the EAACI position paper on immunotherapy (Table 9) (4). When systemic reactions occur, reevaluation of the patient's immunotherapy program is indicated.

Although there are occasional case reports of immunologic diseases associated with immunotherapy (263, 264), several prospective studies did not demonstrate such sequelae.

5.2. *Risk factors based on nonfatal systemic reactions*

The time of onset of nonfatal systemic reactions was reviewed from 38 published papers (265), and most reactions were found to begin within 15—20 mm, regardless of the schedule used. Most systemic reactions were mild and were successfully managed with conventional measures. In two of the 38 studies reviewed, the time of onset of the systemic reaction could be correlated with the severity of the reaction. However, some systemic reactions begin 30—60 mm after the vaccine injection (266).

Asthma appears to be a significant risk factor for systemic reactions (267, 268). Uncontrolled asthma and FEV₁ under 70% of predicted values are risk factors for developing a bronchial reaction (145, 269). Moreover, patients with asthma tend to have more severe bronchial reactions than nonasthmatic patients.

Large local reactions do not predict the onset of a subsequent systemic reaction (270). In one study of 2989 systemic reactions, most of them occurred in the absence of previous large local reactions (265).

Drugs may either prevent or potentiate systemic reactions. β -Blockers are known to potentiate systemic reactions and interfere with treatment. On the other hand, two separate reports described a decreased incidence of systemic reactions when patients received premedication with a combination of methylprednisolone, ketotifen (not available in the USA and some other countries), and a long-acting theophylline (268, 271). Other studies suggested the importance of premedication to reduce systemic reactions (272). H₁-blockers were also shown to reduce the rate of systemic reactions (273—275).

The use of high-dose, potent, standardized vaccines with inhalant allergens (5, 228, 268, 271, 276, 277) or

rush immunotherapy with venoms (5, 49, 160, 278—282) or inhalant allergens (283) may be associated with a higher risk for a systemic reaction. In the AAAAI prospective study on the side-effects of venom immunotherapy, 1410 patients were followed up (278). Ninety-two percent of the treated subjects achieved maintenance dose. Twelve percent of subjects experienced systemic reactions. The incidence of pruritus and angioedema/urticaria was similar with mild, moderate, or severe systemic reactions. The systemic reaction severity did not correlate with the severity of the most recent sting systemic reaction, the most severe historical sting systemic reaction, the most severe systemic reaction during venom skin tests, the total dose of venom, the degree of skin test reactivity, or the lowest concentration yielding a positive skin test. Most systemic reactions occurred between 1 and 50 µg and at maintenance. Honeybee or *I. olivipes* venoms were most likely to produce systemic reaction.

Other factors identified which may increase the likelihood of a systemic reaction are an incorrect injection technique and erroneous dose.

5.3. Risk factors based on fatal reactions

il patients, within 20 to 30 minutes in one, and after more than 30 minutes in one. In both reports, the cause of death was associated with respiratory compromise in most patients, reinforcing the need for special precautions in treating high-risk patients with asthma. Additional cases were reported (287—289) and data were similar to previous reports.

5.4. Risk factors for immunotherapy

Accordingly, it is essential that strict attention be paid to risk factors and that techniques of management be initiated after immunotherapy to minimize these risks. Guidelines (4, 265, 285, 290) have been suggested, emphasizing thorough training of all personnel involved as well as the treatment of systemic reactions. The development and use of standardized vaccines are encouraged. Certain risk factors identified for immunotherapy include:

- 1) errors in dosage
- 2) presence of symptomatic asthma
- 3) high degree of hypersensitivity (by specific IgE measurements)
- 4) use of 13-blockers
- 5) injections from new vials
- 6) injections made during periods of symptomatic skin tests or exacerbation

In 1986, the British Committee on Safety of Medicines reported 26 immunotherapy-associated deaths since 1957, five of which had occurred during the preceding 18 months (284). The importance of deaths due to an acute asthma episode was highlighted in this report.

The following year, the completed analysis of the AAAAI study was reported, comprising 46 fatalities from 1945 to 1984, 30 of which had sufficient data for analysis (285). Six of these 30 deaths were associated with skin tests and 24 with immunotherapy. The mean age for all subjects was 33 years (range 7—70 years). Risk factors for a fatality include subjects with asthma who are suffering from a flare or seasonal exacerbation, patients with higher degrees of sensitivity, and those on 13-blockers. Fifteen of these 24 fatalities associated with immunotherapy had the onset of symptoms within 20 minutes after their injections. Three had onset of symptoms within thirty minutes and two after more than thirty minutes. A subsequent report (286) cited 17 fatalities associated with immunotherapy for the years 1985 to 1989. Sixty-five percent of these patients were undergoing build-up therapy. Other factors associated with fatalities in both reports include changing to a new vial of unstandardized vaccine, dosing error or inappropriate dose adjustment, not waiting after injection and home injection. Onset of anaphylaxis occurred within 20 minutes in

5.5. Precautions For Immunotherapy

The percentage of subjects who experience a systemic reaction from immunotherapy is small but appears to increase as the immunotherapy schedule is accelerated and high-dose regimens are used in highly sensitive subjects.

Maintenance immunotherapy appears to be associated with fewer systemic reactions than the build-up period of rush and accelerated schedules of immunotherapy.

Although premedication with antihistamines has been shown convincingly to reduce the prevalence of systemic side-effects, their use should not reduce the need for the waiting period after injection, and, some

investigators are concerned that systemic reactions may be masked.

The twenty minute waiting period, as recommended by the AAAAI, is adequate for conventional immunotherapy (291). The EAACI recommends a 30-minute waiting period (4). However, a longer waiting period is necessary for high-risk subjects or in the following situations:

- 1) rush immunotherapy
- 2) unstable asthma, control of asthma with drugs is required before any injection
- 3) high degree of hypersensitivity
- 4) β -blockers.

5.6. Equipment recommended for settings where allergen immunotherapy is administered

The following equipment and reagents should be available (292):

- 1) stethoscope and sphygmomanometer
- 2) tourniquets, syringes, hypodermic needles, and large-bore (14-gauge) needles
- 3) aqueous epinephrine HCl 1:1000 (4, 292,293)
- 4) equipment for administering oxygen
- 5) equipment for administering intravenous fluids
- 6) oral airway
- 7) antihistamine for injection
- 8) corticosteroids for intravenous injection
- 9) vasopressor.

The proper use of these reagents and equipment by appropriately trained personnel should provide effective initial treatment for most, if not all, systemic reactions to allergenic vaccines. 'the prompt recognition of systemic reactions and the immediate use of epinephrine are the mainstays of therapy

There are several invasive procedures which are only rarely needed for treatment of systemic reactions. These include:

- 1) direct laryngoscopy
- 2) DC cardioversion (electrical countershock)
- 3) tracheotomy
- 4) intracardiac injection of drugs.

The rare situation in which these procedures might be essential does not justify the risk of their being made available for use under less than ideal circumstances; therefore, it is neither necessary nor practical to insist that these procedures be immediately available to personnel using allergenic vaccines (294).

5.7. Conclusion

Several million immunotherapy injections are administered annually the risk of a fatal systemic reaction from immunotherapy is extremely small. However, any systemic reaction is unacceptable, and all physicians prescribing and/or administering such therapy must be aware of these risks and institute appropriate office procedures to minimize them. Reassessment of the "optimal dose" of standardized vaccines may be necessary to adjust for the increased potency of standardized vaccines. This may improve the overall safety of immunotherapy by limiting the incidence of systemic reactions while providing more predictable therapeutic results.

6. Other routes of immunotherapy

6.1. Introduction

Parenteral allergen injection has been the principal approach for the application of immunotherapy in the treatment of allergic respiratory airway diseases. However, the inconvenience of frequent injection visits, the discomfort associated with injections, and the possibility of adverse reactions have led to the investigation of

alternate routes of delivery of effective doses of allergen vaccines. Local administration of allergens can also have the advantage of stimulating the local immune system where the allergic reaction occurs. Moreover, it has been shown that the local application of allergens has a systemic effect. This section will review the studies employing noninjective routes of specific immunotherapy. These include:

- 1) oral, in which the vaccine (prepared as drops, capsules, or tablets) is immediately swallowed
- 2) sublingual-swallow, in which the vaccine has to be held sublingually for 1—2 mm and then swallowed
- 3) sublingual-spit, in which the vaccine is held in the mouth sublingually for 1—2 mm and then spit
- 4) nasal, in which the vaccine (aqueous or powdered) is delivered in the nose by proper devices
- 5) bronchial, in which the vaccine (aqueous or powdered) is delivered into the bronchi by proper devices.

The first attempts at local immunotherapy were made early in the century, and local immunotherapy has been used in clinical practice without rigorous controls for decades. A renewed interest in local immunotherapy has arisen since 1980 after the reports of fatalities due to immunotherapy. However, this form of therapy is still a matter of debate in many countries, and more studies are needed.

6.2. Efficacy and safety

In order to eliminate any doubt of the validity of the papers included in this review, strict inclusion criteria have been used:

- 1) placebo-controlled, double-blind studies
- 2) studies published in English in peer-reviewed journals (abstracts not accepted)
- 3) allergen vaccines and doses defined
- 4) symptom/medication scores provided
- 5) appropriate treatment protocol.

6.2.1. Oral route

Four of the seven studies fulfilling the above criteria did not show efficacy (295—298) (Table 10). A reduction of symptom-medication scores was

Table 10. Results of double-blind, placebo-controlled studies with oral immunotherapy

Reference	Patient number			Allergen species	Extract	Duration	Symptom-medication scores	
	A	P	B				N	C
Giovane et al.	300	10	B	Mites	Aqueous, standardized	3 years	N, B:	<i>P</i> < 0.05
None								
Mdller et al.	299	22	22	Birch	Enteric-coated	10 months	N:	NS
severe								8
rhinitis								1
asthma								
Mosbech et al.	297	24	27	Grass	Enteric-coated	1 year	N:	NS
urticaria								5
Oppenheimer et al.	29B	23	25	Cat	Aqueous, standardized	3 months	N:	NS
severe?								2
Taudorf et al.	296	25	27	Grass	Enteric-coated	6 months	N:	NS
severe								None
Taudorf et al.	301	18	21	Birch	Enteric-coated	18 months	N:	NS, C:
	P-cOOS, C:		NS					1 urticaria

N: nasal; B: bronchial; C: conjunctival; 0: drug score; 61: gastrointestinal tract; A: active treatment P: placebo treatment; N 5: not significant.

reported in two other studies (299, 300). In an additional study, clinical efficacy was claimed, but the only significant difference between placebo and the active group was for eye symptoms (301). A reduction in reactivity during allergen-specific nasal or conjunctival challenge was also described in some trials (299–301). Finally, the duration of treatment appeared to be crucial, since a significant clinical improvement appeared only after 12 months of treatment (300). Therefore, clinical efficacy is not demonstrated, and further studies are needed before this form of therapy can be recommended for clinical practice.

Significant adverse reactions (urticaria) related to the treatment which utilized very high doses of vaccine were observed only in two studies (297, 301). In the other studies, no severe side-effects were observed, and no significant differences in the side-effect profile were noted between the active treatment and placebo groups. Some mild gastrointestinal side-effects were observed in several studies, but discontinuation of the treatment was not necessary.

6.2.2. Sublingual route

Sublingual immunotherapy can be delivered by either the sublingual-spit method or a sublingual-swallow. However, only the sublingual-swallow method has demonstrated evidence of clinical efficacy

Five sublingual-swallow studies did not meet the inclusion criteria and were excluded. Two used exceptionally low-dose regimens and did not provide symptom/medication scores (302, 303); one did not report allergen doses administered (304); one was not double-blind, placebo-controlled (305) and one had methodological problems (306). Four studies (307–310) demonstrated the clinical effectiveness of sublingual-swallow immunotherapy with grass, *Parietaria*, and mite vaccines (Table 11). A reduction in reactivity during allergen-specific nasal or bronchial challenge was also described. Doses of allergen used in these studies are usually over 5–20-fold greater than those required to achieve efficacy with subcutaneous immunotherapy

The only double-blind, placebo-controlled study on sublingual-spit was carried out in eat-allergic patients (311). It cannot be analyzed due to methodological problems. In the cat room exposure levels were highly variable and data on medications were not provided.

No systemic side-effects have been reported in adults, and in most of these studies, there was no difference between the placebo and the active treatment in side-effects. However, in one study performed in children, systemic side-effects (urticaria and/or asthma) were reported (309).

Sublingual administration of very low-dose allergen solutions including both food and inhalant allergen vaccines have been prescribed by some physicians, particularly those identified as “clinical ecologists” who treat patients with allergic symptoms and various other undefined symptoms. ‘there are no data which show that this form of therapy is effective.

6.2.3. Nasal route

Four studies (312–315) could not be used because of methodological limitations (Table 12). Thirteen out of 14 studies found a significant improvement of nasal symptoms both in perennial (1 study) and seasonal allergic rhinitis (12 studies) (316–329). In general, the effectiveness of nasal immunotherapy seems to be dose-related, and clinical improvement appears to have been greater with aqueous and powdered vaccines than with modified ones. Since only one study was done in pediatric patients, there is a need for more studies in this age group. A reduction in reactivity during allergen-specific nasal challenge was observed in several studies.

Table 11 Results of double-blind, placebo-controlled studies with sublingual-swallow immunotherapy

Reference	Patient number		Symptom-medication Allergen species	Systemic Extract	Duration	scores	side-effects	
	A	P						
Feliziani et al.	307	18	16	Grass	Aqueous, standardized	3.5 months	N: PcOOI	None
Sabbab et al.	308	29	29	Grass	Aqueous, standardized	17 weeks	N: P.cO.0S	10

mild

								Medications reduced
Tan et al.	309	30	28	9. <i>pteronysinus</i>	Aqueous, standardized	18 months	N, B: P<0.001	
Troise et al.	310	15	16	<i>Parietaria</i>	Aqueous, standardized	10 months	N: P<0.05	32 episodes 3 severe asthma 1 mild

N: nasal symptoms; B: bronchial symptoms; A: active treatment P: placebo treatment NS: not significant.

The earliest studies administering aqueous vaccines found a high rate of local side-effects (rhinitis) making the usefulness of nasal immunotherapy somewhat questionable (330). Dry-powder vaccines show an efficacy comparable to aqueous vaccines and demonstrate efficacy with decreased side-effects. Pretreatment with nasal cromoglycate may further reduce nasal side-effects. One paper (322), in which a dried powder preparation was administered, reported asthma after nasal immunotherapy.

6.2.4. Bronchial route

There are two controlled clinical trials of bronchial immunotherapy with mite vaccines (331, 332) which provide controversial evidence of efficacy. Bronchospasm was induced in the majority of patients in both studies.

6.3. Practical aspects

6.3.1. Prescription

Since local immunotherapy is self-administered, it is recommended that the prescription and formulation for such therapy only be made by a physician trained in allergy. Patients must be instructed to carefully follow the physician's administration schedule and to have regular physician visits.

6.3.2. Administration technique

The administration schedule varies, but involves a build-up phase, where the vaccine is administered at increasing doses, and a maintenance phase, where the maximal dose is administered two or three times a week. Furthermore, local immunotherapy can be administered either preseasonally or perennially or by rush schedules.

6.4. Conclusions

Properly controlled, well-designed studies employing sublingual and intranasal immunotherapy provide evidence that this form of therapy may be a viable alternative to parenteral injection therapy to treat allergic airway diseases. Further studies are needed to define better the most appropriate patients for this form of therapy, the optimal therapeutic target dose, and its effectiveness as compared to conventional injection immunotherapy.

7. Pediatric issues

7. Introduction

The use of allergen immunotherapy in children requires a consultation with a specialist, because of the special problems which exist in this age group. The diagnosis of allergic rhinoconjunctivitis in children under the age of 4 to 5 may be difficult and the differential diagnosis between allergic rhinitis and recurrent acute viral infections of the respiratory tract may pose problems.

Most specialists prescribe immunotherapy after the age of 5 years. Some recommend immunotherapy at 1–2 years of age (4). Controlled studies on the benefits versus risks of immunotherapy in patients below 5 years of age are needed (4, 333). However, when immunotherapy is prescribed for an infant, the physician who administers the injections should be able to appropriately treat a systemic reaction in this age group (4).

Motivation for immunotherapy is needed from both the child and the parents to avoid discontinuation of immunotherapy. Such compliance can be achieved with adequate allergy diagnosis and with adequate information of immunotherapy including the information regarding the adverse reactions.

72. Advantages of immunotherapy in children

Immunotherapy when introduced at the onset or during the early phase of the disease (4) may modify the natural course of the allergic disease (113, 154). Immunotherapy is thought to be more effective in children than in adults.

When immunotherapy is administered to children who only have allergic rhinoconjunctivitis, it may prevent the development of asthma. The Preventive Allergy Treatment (PAT) study has been initiated in children aged from 7 to 13 to answer the question “does specific allergen immunotherapy prevent the development of asthma?” Preliminary data suggest that immunotherapy impedes progression from allergic rhinoconjunctivitis to asthma (334, 335).

Immunotherapy did not prevent the development of sensitization to new allergens in polysensitized patients (336). However, a prospective case-control study in 44 asthmatic children aged from 2 to 6 years, who were monosensitized to domestic mite was done to assess whether immunotherapy prevents new sensitization during a three year follow-up (154). All 22 children in the control group (no immunotherapy) as compared to 12/22 children in the immunotherapy group developed new sensitivities as determined by skin prick testing and allergen specific IgE in serum. This study suggests that immunotherapy may alter the natural course of allergy progression by preventing sensitization to new allergens.

Severe allergic reactions to Hymenoptera venoms are rare but occur during infancy and early childhood (337) and immunotherapy may be initiated by an allergist trained to treat children.

73. Problems of immunotherapy in children

- 1) More studies are needed to determine how immunotherapy may modify the allergic disease or impair progression to asthma.
- 2) Rush immunotherapy was associated with a higher incidence of systemic reactions in children less than 5 years of age than in older subjects (271). When a bronchial reaction induced by immunotherapy occurs it may be more difficult to control than reactions which occur later in life (4).
- 3) Small children do not understand that injections cause pain. Therefore, fear of injections or the psychological trauma induced by an anaphylactic reaction should be considered before initiating such therapy (338, 339). This is a major reason why other routes of immunotherapy should be carefully and critically evaluated in children for efficacy and safety
- 4) The optimal dose for maintenance therapy in small children is unknown.
- 5) It is not known whether repeated doses of aluminium hydroxide may induce side-effects in small children.

8. Indications

Hymenoptera-venom immunotherapy is the only effective treatment of insect sting-induced anaphylaxis. Immunotherapy with inhalant allergens reduces symptoms and/or medication needs for patients with allergic asthma and rhino-conjunctivitis.

Immunotherapy is currently not used to treat food allergy, however, it has been used experimentally (340, 341), and when evaluated, injections should be administered in hospitals in clinical trials. Studies have not confirmed the efficacy of immunotherapy for atopic dermatitis (342, 343) and if given for this disease, immunotherapy should only be used in clinical trials.

The mechanisms and treatment of drug-induced anaphylaxis are different than allergen immunotherapy. Therefore, it is not discussed in this document.

Specific immunotherapy should be prescribed by specialists and administered by physicians who are trained to treat anaphylaxis.

8.1. Relative contraindications

Relative contraindications for immunotherapy include (4):

- 1) serious immunopathologic and immunodeficiency diseases
- 2) malignancy
- 3) severe psychological disorders

- 4) treatment with α -blockers (344), even when administered topically
- 5) poor compliance
- 6) severe asthma uncontrolled by pharmacotherapy and/or irreversible airways obstruction (FEy₁ consistently under 70% of predicted after adequate pharmacologic treatment) (8), except for Hymenoptera venom hypersensitivity
- 7) significant cardiovascular diseases which increase the risk of side-effects from epinephrine, except for Hymenoptera venom hypersensitivity
- 8) children under 5 years of age, except for Hymenoptera venom hypersensitivity.

Pregnancy is not considered a contraindication for continuation of immunotherapy, but, in general, treatment should not be started during pregnancy (345).

8.2. Immunotherapy for Hymenoptera venom sensitivity

In patients who have had a systemic reaction, standard preventive care should include:

- 1) advice concerning avoidance of insect stings
- 2) prescription of an emergency kit including epinephrine (unless medically contraindicated)
- 3) consideration of venom immunotherapy

8.2.1. General indications

The indications for venom immunotherapy are provided in the FAACI Position Paper (6). An absolute indication in any age group is a history of severe systemic reactions associated with respiratory and/or cardiovascular symptoms and positive diagnostic tests (skin tests and/or serum specific IgE). Such therapy should not be prescribed without documented IgE-mediated allergy. Children who have had mild systemic reactions characterized only by symptoms of mild angioedema and urticaria usually have a favourable prognosis. The re-sting reaction rate is low (10–20%), and the reactions are almost always of the same mild degree. Venom immunotherapy is not recommended for these children. Adults who have similar mild systemic symptoms appear to have a similar prognosis. Venom immunotherapy is recommended for such patients in the USA, but not usually in Europe. Large local and other unusual reactions are not an indication for venom immunotherapy.

The generally recommended maintenance dose is 100 μ g of venom protein which corresponds to about one or two bee stings (346) and probably more than one or two *Vespula* stings (6). It was found that 100 μ g of venom significantly protected more patients than 50 μ g (347) which also has been recommended (348). Maintenance doses up to 200 μ g may be indicated in honeybee sensitive bee-keepers (349) and in treatment failures (350). Moreover, it is possible that this higher maintenance dose may lead to a more rapid loss of sensitization to venom (165).

In the absence of venom immunotherapy, 25–65% of patients with a systemic reaction will have another reaction when subsequently stung. Therefore, it has been proposed to test every patient with a history of venom allergy with a provocation test with the sting of a living insect in an intensive care unit and to treat only those who react (351–353). However, besides risking a life-threatening reaction during the challenge which may be extremely difficult to be reversed (48), the provocation test is not completely predictive of a subsequent sting reaction, since 20% of patients who had a negative initial sting challenge react to another challenge within the next 6 months. A single sting challenge is therefore insufficient to select patients for venom immunotherapy (354, 355).

8.2.2. Venom immunotherapy in special situations

Beekeepers are at high risk of honeybee venom allergy (349). Venom immunotherapy should be offered to all beekeepers with a history of an anaphylactic reaction, even when they discontinue their profession (6). Wearing protective clothing and the immediate availability of an emergency kit are essential precautions (292).

In *elderly patients*, allergic reactions to Hymenoptera stings may be more severe (337). The fatality rate is higher than in children and young adults, because of associated pre-existing cardiovascular and/or respiratory disease, even though the risk of re-exposure is usually lower. Therefore, venom immunotherapy is indicated in all

elderly patients with a history of severe systemic reactions and positive diagnostic tests.

There are several species of *stinging ants* *Solenopsis* (imported fire ants) and *formicomyrmex* (harvester ants) in the order Hymenoptera. Patients of all ages who have had systemic reactions to ants, regardless of the severity, and who have had positive *in vivo* or *in vitro* IgB tests are at risk for anaphylaxis following a subsequent sting. Although venoms have replaced whole-body vaccines for most Hymenoptera, purified ant venoms are not available for imported fire-ant and harvester-ant immunotherapy. However, there is clinical evidence that imported fire-ant whole-body vaccines contain the venom antigens and are effective (356). These vaccines are not standardized, and double-blind, placebo-controlled studies with sting challenges before and after treatment have not been done.

8.3. Subcutaneous immunotherapy for allergic rhinoconjunctivitis and asthma

The indications of immunotherapy in allergic asthma and rhinitis have been separated in some guidelines (7—9). This artificial separation has led to unresolved issues (357, 358), possibly because the allergen-induced IgE-mediated reaction has not been considered to be a multi-organ disease. It is therefore important to consider immunotherapy based on the allergen sensitization rather than on the disease itself.

8.3.1. General considerations

Double-blind, placebo-controlled studies have confirmed the efficacy of immunotherapy. Clinical efficacy does not necessarily mean clinical indication, especially since controlled trials of immunotherapy are optimally designed and may not always be applicable to daily medical practice. Safe and effective pharmacologic treatment is also available for the treatment of allergic diseases. Thus, before starting immunotherapy, it is essential to appreciate the respective value of allergen avoidance, pharmacotherapy, and immunotherapy (4,7,359, 360). *in vitro* or *in vivo* tests alone without an adequate history and physical examination will lead to inappropriate or less optimal care. Certain factors must be weighted before beginning immunotherapy:

- 1) demonstration that the disease is due to IgB-mediated allergy (Table 13)
- 2) determine all symptoms caused by allergens
- 3) assess the allergen exposure (361) and before initiating immunotherapy, avoidance of exposure to the allergen(s) causing the symptoms of the IgB-mediated reaction should always be attempted. However, most common aeroallergens cannot be completely avoided, and this is particularly true for patients allergic to domestic mites or to multiple allergens.
- 4) potential severity of the disease to be treated
- 5) efficacy of available treatment modalities
- 6) patient's attitude to available treatment modalities
- 7) quality of allergen vaccines used for treatment. When possible standardized allergens should be utilized (362)
- 8) cost and duration of each form of treatment
- 9) risk incurred from the allergic diseases and the various forms of treatment.

Additional pharmacotherapy may be necessary to control symptoms in patients receiving immunotherapy

Although rare, deaths caused by immunotherapy usually occur in patients with asthma and are due to acute bronchial obstruction (284, 286). To minimize risk for severe reactions and improve efficacy, therefore, it is essential to follow precise recommendations (4, 7) (table 14).

In allergic rhinitis, immunotherapy is indicated for subjects:

- 1) in whom antihistamines and topical medications insufficiently control symptoms
- 2) who do not wish to be on pharmacotherapy
- 3) in whom pharmacotherapy produces undesirable side-effects
- 4) who do not desire to receive long-term pharmacologic treatment.

In allergic asthma, immunotherapy is indicated for subjects:

- 1) who do not present a severe form of the disease. FEy₁ levels should be over 70% of predicted values after adequate pharmacologic treatment
- 2) in whom symptoms are not adequately controlled by allergen avoidance and pharmacologic treatment
- 3) who have both nasal and bronchial symptoms
- 4) who do not wish to be on long-term pharmacotherapy
- 5) in whom pharmacotherapy produces undesirable side-effects.

8.3.2. Pollen allergy

Immunotherapy is indicated in pollen-induced allergic diseases based on its severity and duration. Moreover, it is commonly accepted that immuno

Table 13. Considerations for initiating immunotherapy

- 1) Presence of demonstrated gE-mediated disease
Positive skin tests and/or serum specific gE
 - 2) Documentation that specific sensitivity is involved in symptoms
Exposure to allergen(s) determined by allergy testing related to appearance of symptoms
If required, allergen challenge with relevant allergen(s)
 - 3) Characterization of other triggers that may be involved in symptoms
 - 4) Severity and duration of symptoms
Subjective symptoms
Objective parameters; e.g., work loss, school absenteeism
Pulmonary function (*essential*): exclude patients with severe asthma
Monitoring of pulmonary function by peak flow
 - 5) Response of symptoms to nonimmunologic treatment
Response to allergen avoidance
Response to pharmacotherapy
 - 6) Availability of standardized or high-quality vaccines
 - 7) Contraindications
Treatment with p-blocker
Other immunologic disease
Inability of patients to comply
 - B) Sociologic factors
Cost
Occupation of candidate
Impaired quality-of-life despite adequate pharmacologic treatment
 - 9) Objective evidence of efficacy of immunotherapy for selected patient (availability of controlled clinical studies)
-

Table 14. Recommendations to minimize risk and improve efficacy of immunotherapy (from International Consensus Report on Oligosis and Management of Asthma [7])

- 1) Specific immunotherapy must be prescribed by specialists and administered by physicians trained to manage systemic reactions if anaphylaxis occurs
 - 2) Patients with multiple sensitivities may not benefit as much as do patients with single sensitivity from specific immunotherapy; more data are necessary
 - 3) Patients with nonallergic triggers will not benefit from specific immunotherapy
 - 4) Specific immunotherapy is more effective in children and young adults than later in life
 - 5) It is essential for safety reasons that patients should be asymptomatic at time of injections because lethal adverse reactions are more often found in asthma patients with severe airways obstruction
 - 6) FEy₁ with pharmacologic treatment should reach at least 7fl% of predicted values, for both efficacy and safety reasons
-

therapy is indicated if the season is prolonged or in polysensitized patients exposed to several subsequent pollen seasons (e.g. tree and grass pollen sensitivity) (2, 4).

Since rhinoconjunctivitis occurs in most, if not all patients, who have pollen allergy, and asthma occurs in some of the most severe patients, it is impossible to propose indications for immunotherapy without considering all symptoms (59, 178). Immunotherapy is indicated when asthma during the pollen season complicates rhinoconjunctivitis. British guidelines on immunotherapy state that patients with asthma should not receive immunotherapy (363), but, this is the only country with such a recommendation.

8.3.3. *Immunotherapy with domestic mite allergens*

House dust extract is not recommended for allergen immunotherapy (4). Patients are candidates for mite vaccine immunotherapy if mite avoidance measures are not effective. Patients in whom symptoms are only caused to a minor degree by exposure to mites are not candidates for immunotherapy. Indications for immunotherapy are based on the severity and the duration of allergic rhinitis and asthma.

8.3.4. *Immunotherapy with animal dander allergens*

Avoidance is the treatment of choice for animal dander-induced allergic diseases. However, complete avoidance is often impossible even in environments in which animals are not present (364). Immunotherapy to animal dander allergens may be prescribed for patients in whom avoidance of animal allergens is not effective, or when animal exposure persists in the home or work environment.

8.3.5. *Immunotherapy with molds*

Avoidance, where possible, of indoor mold allergens is the treatment of choice. Certain studies have demonstrated clinical improvement when well-characterized vaccines of *Cladosporium* or *Alternaria* have been used to treat mold-induced allergy. Patients with positive diagnostic tests who have symptoms from other relevant mold allergens may be considered for immunotherapy.

8.3.6. *Immunotherapy with other allergens*

Immunotherapy with vaccines of undefined allergens, such as bacteria, *Candida albicans*, or *Trypophyton*, is not recommended (4).

8.3.7. *Monitoring of immunotherapy*

Monitoring the effectiveness of immunotherapy with inhalant allergens is based on the clinical response and the reduction of pharmacotherapy. *In vitro* or *in vivo* markers to assess efficacy are not available.

8.4. *Other routes for allergen administration*

The subcutaneous route is the usual route for administration of immunotherapy, but oral, sublingual, nasal, and bronchial routes have been proposed. A workshop of the EAACI and ESPACI has been convened in Portofino (Italy), September 28, 1996 to assess the efficacy and safety of other routes of immunotherapy.

Nasal immunotherapy may be indicated in carefully selected adult patients with rhinitis caused by pollen and possibly for mite. The potential candidates for nasal immunotherapy are patients who cannot be properly controlled by conventional pharmacotherapy, who have presented with previous systemic reactions induced by injection immunotherapy, or refuse injections. Patients should be carefully informed of the potential risks of a systemic reaction, since the allergen vaccines are self-administered at home in the absence of a physician, and how to treat an allergic reaction should it occur.

Sublingual (swallow) immunotherapy may be indicated in pollen and mite induced rhinitis. It is safe in adults. The treatment is currently not recommended for children except as part of a controlled study. Further studies with standardized vaccines are needed to define further the indication of sublingual immunotherapy.

Bronchial and oral immunotherapy, because of lack of efficacy and the risk of severe side-effects (bronchial

immunotherapy), are not recommended except in controlled studies.

8.5. *Indications for immunotherapy in children*

- 1) IgE-mediated allergic rhinoconjunctivitis and asthma
- 2) severe anaphylactic reactions to Hymenoptera stings
- 3) the same considerations for diagnosis and treatment as indicated for adults apply to children.

Immunotherapy for children is more complicated than for adults because of their age and dependent nature. Educating the family about immunotherapy is of great importance and is crucial for its success.

Children (under 15 years in Europe and 18 years in the USA) should be accompanied by an adult when they receive their injection unless parental permission for therapy without their presence has been obtained. Precautions to be taken following an injection involve family, friends, and teachers. Adolescents are particularly noncompliant, and physicians must emphasize that compliance is essential for effective immunotherapy.

9. Costs

Asthma and allergic diseases account for a significant proportion of overall health care and expenditure in industrialized countries. For example, the costs for asthma represent 1—2% of the total direct and indirect health costs in any country (8, 365). Over the past 10 years, costs for asthma and allergic diseases have increased more than for most other diseases. The greatest proportion of expenses for asthma is currently due to hospital care.

Allergic diseases usually begin early in life and persist. Moreover, many patients who have allergic rhinitis also eventually develop asthma. The short-term costs are known, however, the long-term costs from the morbidity and the complications from these diseases are not established and probably account for even higher costs.

Inhaled corticosteroids, which are costly, significantly reduce hospitalizations and costs for asthma (366—368). Expert care results in more effective asthma treatment and reduces morbidity and, by inference, costs incurred for these diseases (369—375).

For rhinoconjunctivitis, however, such studies are lacking. The disease significantly reduces quality-of-life (376) and may increase the incidence of viral respiratory diseases and sinusitis, thus adversely impacting direct and indirect costs for patients with these diseases.

10. Future strategies for immunotherapy

10.1. *Therapeutic vaccines of the future*

The term “immunotherapy” has traditionally referred to treatment of allergic diseases and asthma by repeated injections of extracts containing relevant allergens. Advances in basic immunology should lead to new, safer, and substantially more effective, methods to manipulate the human immune response, introducing the development of “therapeutic vaccines”. These new approaches will be of potential therapeutic benefit for diseases, such as asthma, allergic diseases, as well as autoimmune diseases, such as type I diabetes and multiple sclerosis. Therapeutic manipulation of the human immune response requires knowledge about

- 1) the relevant antigens implicated in the immunopathology of the disease
- 2) the changes in the immune response that are most effective to prevent and treat the disease
- 3) the “vaccination” strategy that will alter the immuneresponse.

The therapeutic targets in autoimmune diseases remain unknown, but a number of relevant approaches have been identified using experimental non-obese diabetes mouse models of type I diabetes (insulin-dependent diabetes mellitus) and of multiple sclerosis (experimental allergic encephalomyelitis). Notably, glutamic acid decarboxylase and insulin are key antigens in diabetes models, whereas myelin basic protein and proteolipid protein can induce experimental allergic encephalomyelitis in experimental multiple sclerosis models (377). Generally, Th1 cytokines contribute to the pathology of these autoimmune diseases; in contrast, abrogation of the production of IFN- γ or induction of IL-4 production ameliorates the diseases in mice (377, 378).

In view of the important role that T helper cells play in many diseases, by their production of cytokines, many approaches attempt to alter the balance of Th1 and Th2 cells in affected tissues or organs. For example,

disruption of the pathway mediated by costimulatory molecular interactions (CD28 interacting with its ligands CD80 or CD86) results in improvement or worsening of insulin-dependent diabetes mellitus or experimental allergic encephalomyelitis. Improvement was found to be associated with a reduction in the ratios of Th1/Th2 cytokines and aggravation of the diseases with an enhancement in the ratios of Th1/Th2 cytokines (379, 380). At present, methods to manipulate the immune response in mouse models are not uniformly protective against disease; in part, because current knowledge about the underlying mechanisms is incomplete. For example, an antigen may not only induce tolerance (or immune deviation) but may also prime T cells for a subsequent immune response that may worsen the disease (381—383). Several therapeutic trials, targeted to induce tolerance of insulin or myelin, have resulted in some preliminary evidence of efficacy (384—386). Concomitant changes in *in vitro* cytokine production profiles of the antigen-specific T cells have been observed. Thus, on the basis of such novel approaches, therapeutic vaccination may be used to treat autoimmune diseases, perhaps by decreasing the ratios of Th1/Th2 cytokines, or to treat allergic diseases and asthma by increasing them.

Many of the important allergens have now been cloned (35), key epitopes have been identified, and modified allergens are available (387). Recombinant DNA technology allows for the large-scale production of highly purified and defined allergens for diagnostic and therapeutic purposes. Specific approaches to new forms of immunotherapy to treat allergic diseases, which are based on recombinant DNA technology and the use of synthetic allergen-derived peptides will be discussed below.

10.2. Novel delivery systems

Proteins are subject to absorption, biodistribution, metabolism, and degradation at sites and rates which may not permit effective interactions with components of the immune system. Drug carrier technology may overcome some of these obstacles. Because of the lipid and particulate nature of liposomes, increased delivery of proteins to lymphatics, lymph nodes, and macrophages may be possible when a protein is associated with a liposome (388). Such an approach is currently under investigation in humans, but definitive results are still lacking (56, 57, 389).

10.3. Nonanaphylactic allergen- or allergen fragments or peptides for active immunotherapy

It is suggested that a key mechanism for successful conventional immunotherapy is to alter T-cell function, either by diminishing production of both Th1 and Th2-like cytokines (121, 127), and/or by inducing “immune deviation” from a Th2-like cytokine pattern to a Th1-like pattern (120, 122). One promising strategy to increase the dose of allergens which can be used with increased safety is through the use of allergen derivatives which do not cause anaphylaxis. Isoforms of major allergens have been identified which show no or very low IgE-binding capacity but contain T cell stimulating sequences. Such hypoallergenic isoforms have been identified for major tree-pollen allergens (390, 391) and have been produced *in vitro* for the mite allergen Der p 2 by site-directed mutagenesis (392, 393). The expression of two recombinant allergen moieties of the major birch pollen allergen Bet v 1 has resulted in the generation of fragments containing T cell activation-inducing epitopes which did not cause anaphylaxis activity due to disruption of the three-dimensional structure of Bet v 1 (394). A similar approach using allergen-derived peptides demonstrated that human T-cells become nonresponsive following stimulation with suboptimal concentrations of short ragweed-allergen-derived peptides (395). Furthermore, when mice are treated with antigen-derived peptides that represent major T cell epitopes of the cat allergen Fel d 1, the mice became tolerant to the complete native allergen (396, 397). Clinical studies, using T cell-derived peptides, are in progress to determine whether peptide immunotherapy may represent an effective low-risk alternative to traditional immunotherapy (398). However, whether every T-cell epitope will be needed for a good result has yet to be determined.

10.4. IgE-binding haptens of major allergens for passive saturation of effector cells and induction of blocking antibodies

The cross-linking of mast cell and basophil bound IgE antibodies with specificity for at least two different allergen epitopes is required to trigger release of inflammatory mediators. Localization of IgE-binding sites can be achieved by mapping of immunodominant epitopes as was demonstrated for the timothy pollen allergen, Phl p 1 (399). Epitope mapping must be supplemented by structural analysis, so that antibody binding sites can be superimposed on the three dimensional allergen structure (400), as demonstrated for birch profilin (401). Nonanaphylactic IgE-binding haptens might be used for local saturation of effector cells to prevent subsequent

activation by exposure to the complete allergen for active immunotherapy to induce blocking IgU-antibodies directed against the same IgE-epitopes.

10.5. Plasmid DNA immunization

Injection of DNA encoding the antigen (DNA vaccination) is an alternative approach to raise protective immunity compared to injection of the antigen itself. Intramuscular injection of plasmid DNA containing bacterial genes with a suitable appropriate promoter is followed by transfection of the host cells which will produce bacterial proteins, and elicit humoral and cytotoxic lymphocyte-mediated responses. Mucosal vaccines induce local immune response, both by type 1 and type 2 dependent pathways (402, 403). Intradermal immunization with pDNA, encoding *E. coli* beta galactosidase, induces a Th1 response, whereas immunization with the entire protein induces a Th2 response (404). Based on these results, as well as the observation that plasmid cDNA immunization results in the downregulation of IgE production, it can be speculated that immunization with pDNA encoding for allergens will modulate the cytokine production profile of allergen specific T cells and thereby provide a novel type of immunotherapy for allergic diseases (404, 405).

10.6. Allergen-specific antibodies and antibody fragments for passive therapy in the allergic effector organs

“Non regime” allergen-specific antibodies were discovered over five decades ago, and certain allergen-specific IgG antibodies are able to inhibit IgE mediated anaphylactic reactions (406), particularly those belonging to the IgG4 subclass, might be induced by specific immunotherapy (407). Several studies failed, however, to correlate the presence and/or increase of allergen-specific IgG (98) and IgG subclass antibody serum levels (408) with clinical improvement during immunotherapy. Previous studies comparing the role of allergen-specific IgG antibody in the inhibition of IgE production and subsequent downregulation of allergic responses following immunotherapy were hampered by the lack of well-defined allergen vaccines. Studies using recombinant birch pollen allergen, Bet v 1 (409, 410), recombinant timothy pollen allergens, PhI p 1, PhI p 2, PhI p 5 (411), and recombinant Der f 2 (412, 413) have shown that sera from allergic and nonallergic individuals contain allergen-specific IgG-antibodies belonging to different subclasses and that levels of allergen-specific IgE in serum do not correlate with IgG-subclass responses.

Using epitopes from major allergens prepared by recombinant DNA technology, it has been reported that IgE and/or IgG antibodies of allergic patients bind to the same but also to different epitopes (413). These results indicate that IgE and IgG responses in allergic patients may also evolve in a non sequential fashion, and that IgE and IgG antibodies may possess different affinities or fine specificities for certain epitopes. Taken together, it appears that allergen-specific IgE and IgG responses in allergic individuals are poorly synchronized with respect to epitope recognition and affinity. Differences in epitope recognition by IgE and IgG antibodies have been demonstrated for monoclonal human IgG antibodies specific for Bet v 1 (96). However, the demonstration that certain Bet v 1 specific IgG antibodies block IgE binding to Bet v 1 and inhibit Bet v 1 induced histamine release in allergic patients suggests that blocking IgG antibodies may be utilized to reduce IgE mediated allergic reactions (96, 414). Therefore, recombinant allergen-specific F(ab')₂ fragments may be useful for passive (blocking) therapy of IgE mediated allergic reactions.

10.7. Immunotherapy with humanized anti-IgE monoclonal antibodies or IgE-mimotopes

IgE has a pivotal role in the pathogenesis of allergic diseases, and inhibiting the IgE response by preventing IgE synthesis or blocking the effector phase, through the use of neutralizing anti-IgE antibodies, should have potential therapeutic value. Support for this concept comes from the observation that high levels of anti-IgE antibodies at birth seem to be associated with a reduced predisposition to develop atopic disorders (415). It should be noted, however, that human anti-IgE antibodies have been found to both inhibit and enhance binding of IgE to its low affinity receptor CD23 (416), underscoring the importance of such antibodies in regulating IgE synthesis. Similarly, anti-IgE antibodies have been described that either enhance or downregulate the IgE mediated release of mediators from mast cells and basophils, suggesting that a subpopulation of such antibodies have the potential to interfere with the functional activity of IgE antibodies (417).

A humanized anti-human IgE mouse monoclonal antibody has been reported which binds to free IgE, but not to IgG or to mast cell-bound IgE and blocks binding of IgE to its high-affinity receptor, Fc ϵ -RI (418). In preclinical studies, this antibody inhibited allergen-induced IgE synthesis by cultured human lymphocytes, and although it did not suppress skin test reactivity, it was shown to reduce allergen-induced bronchial provocation

(419, 420).

Another potentially useful strategy is to induce autoantibodies against the Fc γ RI binding site on human IgE by using recombinant human IgE fragments or mimotopes comprising the receptor binding site (421, 422).

11. Clinical research needs

Future studies are needed to evaluate current immunotherapy in the following areas:

- 1) cost-effectiveness
- 2) quality-of-life and patient's satisfaction
- 3) comparison with pharmacologic treatment
- 4) "add-on" studies, e.g. effect of immunotherapy added to treatment with topical therapy
- 5) optimal duration
- 6) reduction of the severity of asthma in terms of hospital admissions and emergency room visits
- 7) decrease of the incidence of asthma in patients suffering from allergic rhinitis, and no asthma
- 8) altering the natural course of the allergic rhinoconjunctivitis and/or asthma
- 9) decreasing the long-term sequelae associated with allergic rhinitis and/or asthma
- 10) long-term effects after cessation
- 11) efficacy and safety for animal allergy (dogs, horses)
- 12) efficacy and safety for occupational allergy (e.g. laboratory animal allergy)
- 13) efficacy and safety in children under 5 years of age.

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