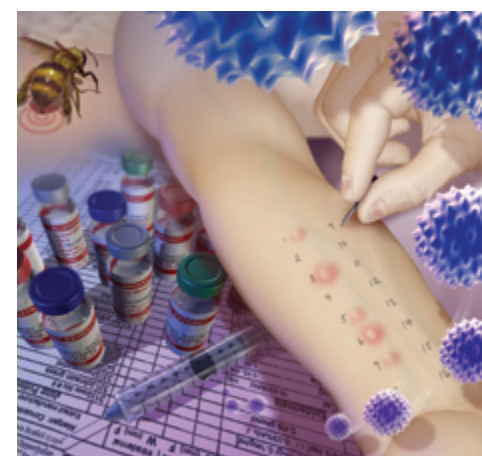
Allergen Immunotherapy

JENNIFER L. HUGGINS, M.D., and R. JOHN LOONEY, M.D.
University of Rochester School of Medicine and Dentistry, Rochester, New York

Allergen immunotherapy (also called allergy vaccine therapy) involves the administration of gradually increasing quantities of specific allergens to patients with IgE-mediated conditions until a dose is reached that is effective in reducing disease severity from natural exposure. The major objectives of allergen immunotherapy are to reduce responses to allergic triggers that precipitate symptoms in the short term and to decrease inflammatory response and prevent development of persistent disease in the long term. Allergen immunotherapy is safe and has been shown to be effective in the treatment of stinging-insect hypersensitivity, allergic rhinitis or conjunctivitis, and allergic asthma. Allergen immunotherapy is not effective in the treatment of atopic dermatitis, urticaria, or headaches and is potentially dangerous if used for food or antibiotic allergies. Safe administration of allergen immunotherapy requires the immediate availability of a health care professional capable of recognizing and treating anaphylaxis. An observation period of 20 to 30 minutes after injection is mandatory. Patients should not be taking beta-adrenergic blocking agents when receiving immunotherapy because these drugs may mask early signs and symptoms of anaphylaxis and make the treatment of anaphylaxis more difficult. Unlike antiallergic medication, allergen immunotherapy has the potential of altering the allergic disease course after three to five years of therapy. (Am Fam Physician 2004;70:689-96,703-4.



✉ **Patient information:** A handout on allergy shots, written by the authors of this article, is provided on page 703.

See page 633 for definitions of strength-of-recommendation labels.

Allergen immunotherapy involves subcutaneous injections of gradually increasing quantities of specific allergens to an allergic patient until a dose is reached that will raise the patient's tolerance to the allergen over time, thereby minimizing symptomatic expression of the disease. Because the proteins and glycoproteins used in allergen immunotherapy are extracted from materials such as pollens, molds, pelt, and insect venoms, they were originally called allergen extracts. In 1998, the World Health Organization (WHO) proposed the term "allergen vaccine" to replace "allergen extract," because allergen immunotherapy is an immune modifier just as vaccines are.[1]

The efficacy of allergen immunotherapy has been known since 1911, when Noon injected an extract of grass pollen into a person in England whose allergic symptoms coincided with the pollination of grass.[2] Since then, controlled studies have shown that allergen immunotherapy is effective in patients with allergic rhinitis, allergic conjunctivitis, allergic asthma, and allergic reactions to Hymenoptera venom.[3-6] Patients with one or more of these diagnoses are considered for immunotherapy if they have well-defined, clinically relevant allergic triggers that markedly affect their quality of life or daily function, and if they do not attain adequate symptom relief with avoidance measures and pharmacotherapy. Despite proven efficacy, the exact mechanism of allergen immunotherapy remains unknown.

Selection of Patients

To make a definitive diagnosis of allergy, IgE-mediated, type I, immediate-hypersensitivity

skin testing typically is performed by scratching diluted allergen into the skin surface or by injecting it intradermally. A positive skin test reaction reflects the presence of specific IgE antibodies to the tested allergen, and a correlation of the specific IgE antibodies with the patient's symptoms, suspected triggers, and allergen exposure is definitive. In vitro, allergen-specific immunoassays to detect serum IgE antibodies are less sensitive than skin testing but may be used in patients with skin diseases that would obscure skin testing results or in those who cannot stop taking medications that suppress the skin test response. The circumstances in which allergen immunotherapy is particularly useful are summarized in *Table 1*. The allergens for which immunotherapy is known to be effective are Hymenoptera venom,[5] pollens,[5,6] cat dander,[7] dust mites,[8] cockroaches,[9] and fungi.[10] Allergy immunotherapy is not efficacious for atopic dermatitis, urticaria, or headaches, and cannot be used for food allergies because the risk of anaphylaxis is too great.

Type 1 immediate-hypersensitivity skin testing with clinical correlation is used to diagnose a specific IgE-mediated allergy.

TABLE 1
Best Indications for Immunotherapy

Allergic rhinitis, conjunctivitis, or allergic asthma
History of a systemic reaction to Hymenoptera and specific IgE antibodies to Hymenoptera venom
Patient wishes to avoid the long-term use or potential adverse effects of medications
Symptoms are not adequately controlled by avoidance measures or medications
Cost of immunotherapy will be less than cost of long-term medications

Benefits

Durham and colleagues[11] conducted a randomized, double-blind, placebo-controlled trial to look at effects in patients who had received three to four years of immunotherapy. They were able to demonstrate a marked reduction in allergy symptom scores and antiallergic medication usage, as well as an alteration in the natural course of allergic disease. Preliminary reports suggest that immunotherapy for allergic rhinitis may reduce the risk for later development of asthma in children.[12,13] In addition, early treatment with allergen immunotherapy in children who were sensitive only to house dust mites reduced development of sensitivity to other allergens.[14] In contrast to the use of antiallergic medication, allergen immunotherapy has the potential to alter the natural course of allergic disease and prevent progression or development of multiple allergies. Consequently, many allergists have suggested its use earlier in the course of allergic disease.

In 2000, the Immunotherapy Committee of the American Academy of Allergy, Asthma, and Immunology (AAAAI) provided a five-year cost comparison of medication usage and single-injection allergen immunotherapy for allergic rhinitis. The cost of medications is much greater than that of single-injection immunotherapy. Long-term costs deriving from the morbidity and complications of allergic diseases are not established, but allergies usually begin early in life and persist if not treated with allergen immunotherapy. A reasonable assumption is that allergen immunotherapy dramatically lowers the cost of treating allergic diseases.

The Authors

JENNIFER L. HUGGINS, M.D., is a fellow in allergy/immunology and rheumatology at the University of Rochester School of Medicine and Dentistry, Rochester, N.Y. She received her medical degree from the University of Kansas School of Medicine, Kansas City, Kan. Dr. Huggins completed postdoctoral training in pediatrics at Baylor University in Houston, and in internal medicine at the University of Rochester School of Medicine and Dentistry. She is board-certified in internal medicine and pediatrics.

R. JOHN LOONEY, M.D., is associate professor of medicine and director of the Allergy and Immunology training program at the University of Rochester School of Medicine and Dentistry, where he also received his medical degree and completed postdoctoral training in internal medicine, infectious diseases, and immunology. He is board certified in infectious diseases, allergy and immunology, and rheumatology.

Address correspondence to Jennifer L. Huggins, M.D., University of Rochester, Allergy/Immunology/Rheumatology Fellow, 601 Elmwood Ave., Box #695, Rochester, NY 14642 (e-mail: Jennifer_Huggins@urmc.rochester.edu) Reprints are not available from the authors.

Standardization, Storage, and Mixing of Allergen Vaccines

Ideally, vaccines should be standardized with a defined potency and labeled with a common unit.[15] Such standardization would eliminate the variability in vaccines and allow for safer and more effective dosing. The Bioequivalent Allergy Unit (BAU), which is assigned by the U.S. Food and Drug Administration based on quantitative skin testing performed on a reference population of allergic patients known to be highly skin-test–reactive to that allergen, reflects clinical potency and is currently used for standardization of vaccines.

Standardized allergens available in the United States include cat dander, grass pollens, dust mites, and short ragweed pollen. Unstandardized vaccines may vary widely in biologic activity based on manufacturer and by lot, depending on the allergen content of the raw material and the conditions of extraction. Furthermore, the labeling conventions of Protein Nitrogen Units (PNUs) or weight by volume (wt/V) reflect protein content but not allergenic potency. Research is underway on new technologies for DNA and protein analysis that would allow an allergen vaccine to be characterized by the content of the major allergen and the consistency of each lot to be monitored accurately.

Vaccine strength is maintained by a number of procedures, including lyophilization and reconstitution with a stabilizer that contains an antimicrobial agent. A volume effect can occur as a result of adherence of the allergen to the vial surface; the larger the surface area of the vial, the more allergen is lost. Glycerol and human serum albumin (0.03 percent) are used to mitigate the volume effect. Glycerol has the added advantage of being an antimicrobial agent. At a concentration of 50 percent, glycerol inhibits enzymatic degradation of the allergens, but it may be irritating at this concentration. The combination of human serum albumin as a stabilizer and phenol as an antimicrobial additive often is used. However, human serum albumin typically is refused by patients who are Jehovah's Witnesses.

The maintenance concentrate is the dose of vaccine considered to be therapeutically effective for each of its constituent components.

Vaccines must be stored properly to preserve biologic activity *(Table 2)*.[16,17] Vaccines should be refrigerated at 4°C (39.2°F) because storage at ambient room temperature results in loss of potency within weeks, with degradation occurring within days at higher temperatures. Critical to vaccine potency is the dilution effect: highly concentrated vaccines are more stable than dilute vaccines.[18] The vaccine label should always be checked for the expiration date.

For immunotherapy to be effective, an optimal dose of each allergen must be determined. When a patient has multiple sensitivities caused by related and unrelated allergens, vaccines containing mixtures of these allergens may be prescribed. As multiple vaccines are mixed, not only will the concentration of each allergen be decreased, but certain allergens will interact. For example, fungi, dust mites, insect venoms, and cockroach have high proteolytic enzyme activity and may be combined with each other but should not be mixed with other allergens. Insect venoms usually are given alone.

TABLE 2
Factors Affecting Allergen Vaccine Potency

Time (always check the expiration date)
Storage temperature
Concentration
Volume of the vial
Type and number of allergens in the vial
Diluent used
Preservatives added

Information from references 16 and 17.

Vaccine Administration

The maintenance concentrate is the dose of vaccine that is considered to be therapeutically effective for each of its constituent components. The maintenance concentrate

TABLE 3
Required Information on Vaccine Vials

Patient's name, date of birth, and patient number
Generalized content of the vaccine*
Expiration date
Dilution from maintenance concentrate in volume per volume (v/v)
Number identifier†
Appropriate colored caps‡

**—Specific contents of each vaccine should be written on a standardized form similar to the "Immunotherapy Mix Components" form found online at: http://www.aaaai.org.*
†—In the numbering system, the maintenance concentrate should always be number 1; subsequent dilutions should be numbered from the maintenance concentrate.
‡—The color-coding system should always start with red for the maintenance concentrate followed by yellow, blue, green, and silver, in that order.

Information from reference 18.

should be determined by a prescribing allergist and clearly written on a standardized Maintenance Concentrate Prescription Form (available online at http://www.aaaai.org). An optimal maintenance dose in the range of 5 to 20 mcg of major allergen per injection correlates with efficacy. Maintenance concentration is usually achieved by administering between 18 and 27 serial dose increments at weekly intervals (a build-up schedule written by the allergist) until the maintenance concentrate is achieved. In a typical build-up schedule, the patient will reach the maintenance concentrate in six months, but patients with a higher degree of allergen sensitivity may require a longer build-up phase. The maintenance dose usually is administered every three to four weeks, and maximum benefit typically is achieved in four to five years. Some patients will note early improvement in their symptoms, but long-term benefit seems to be related to the cumulative dose of vaccine given over time.

To reduce administration errors, the AAAAI recommends a universal, consistent, and redundant labeling system for every vial *(Table 3)*.[18]

Some circumstances warrant adjustments in the dosage schedule. In these situations, communication between the family physician and the prescribing allergist is encouraged to increase safety and avoid unexpected reactions. If the interval between injections is prolonged *(Table 4)*, the dose of vaccine must be reduced; when a new maintenance vial is obtained from the manufacturer, a dose reduction of 50 percent is recommended. For example, 0.5 mL of 1:500 V/V dilution should be reduced to 0.25 mL of 1:500 V/V dilution. The dose is increased every seven to 14 days until the maintenance dose is reached again. No evidence-based guidelines for dose adjustments following local, systemic, or delayed reactions are available, and the allergist should provide treatment suggestions for each of these reactions *(Table 5)*.

Listed in *Table 6* are items that should be reviewed before injecting the patient. The desired injection site is the outer aspect of the upper arm, midway between the shoulder and the elbow in the groove between the deltoid and triceps muscles. The injection is given subcutaneously, preferably with a 26- or 27-gauge needle; if blood is aspirated initially, the vaccine should not be injected. The plunger on the syringe should be depressed at a rate that does not result in wheal formation or excessive pain. Mild pressure should be applied to the injection site for about one minute, and a bandage may be placed if needed. Rubbing the injected area causes rapid absorption and should be avoided.

TABLE 4
Sample Adjustment to Immunotherapy Following Interruption of Dosage Schedule

Weeks from last injection	*Dosage adjustment**
6	Repeat previous dose.
7	Drop back two increments.
8	Drop back three increments.
9	Check with allergist.

NOTE: *Patients on maintenance therapy (injections every three to four weeks). Increments are provided by the allergist.*

**—Patients must return weekly until they reach the maintenance concentrate again.*

Safety Issues

Allergen immunotherapy is safe, but the potential for an adverse reaction is always

present. Although these reactions are rare, they can be life-threatening. In 1924, Lamson reported the first case of death following immunotherapy.[19] A statistical review of the literature about systemic reactions following allergen immunotherapy by Lockey and colleagues[20] found that severe systemic reactions occurred in less than 1 percent of the patients receiving conventional immunotherapy in the United States. From 1985 to 1993 in the United States, 52.3 million administrations of immunotherapy resulted in 35 deaths. These numbers equate to a mortality incidence of less than one per 1 million.[21]

Patients with medical conditions that reduce their ability to survive systemic allergic reactions are not candidates for allergen immunotherapy. Examples of such conditions include chronic lung disease with a forced expiratory volume in one second (FEV_1) of less than 50 percent, beta-blocker or angiotensin-converting enzyme (ACE) inhibitor therapy, unstable angina or myocardial infarction, uncontrolled hypertension, and major organ failure. Allergen immunotherapy also cannot be used in patients who would have difficulty reporting signs and symptoms of a systemic reaction, such as children younger than three or four years. In addition, beta-blocker or ACE-inhibitor therapy may mask early signs of anaphylaxis. Patients who have not been compliant with other forms of therapy are not likely to be compliant with immunotherapy, thus necessitating frequent alteration in dosage schedules and increasing the chance for errors. Patients should be assessed with each injection for newly acquired risks that may not have been present at the beginning of allergen immunotherapy.

Patients with severe, poorly controlled asthma are at higher risk for systemic reactions to immunotherapy injections than patients with stable, well-controlled asthma.[20] Some physicians measure peak expiratory flow readings in all patients with asthma before administering allergen immunotherapy and withhold injections if the reading is less than 70 percent of predicted. Other measures that should be performed to minimize the risk of adverse reactions to allergen immunotherapy are listed in *Table 6*.

Because a systemic reaction occurring during pregnancy may produce severe fetal hypoxia or precipitate premature uterine contractions, immunotherapy should not be initiated during pregnancy.[18] However, immunotherapy can be maintained during pregnancy provided the patient is tolerating and benefiting from the injections.

TABLE 5
Potential Adverse Reactions to Allergy Vaccines and Suggested Treatment

Adverse reaction	*Suggested treatments*
Local reaction	
Common, occurs at the injection site, IgE-mediated, manifested primarily by wheal and flare with pruritus, usually begins 20 to 30 minutes after injection	Local cold pack; oral antihistamine; topical steroid; if reaction recurs, consider premedication with an antihistamine; rinse the syringe with diphenhydramine (Benadryl) or epinephrine before vaccine; consult allergist for dose adjustment
Large local induration	
Occurs at injection site, IgG complex (Arthrus) reaction, manifested by pain, tenderness, and hard swelling	Oral steroids, nonsteroidal anti-inflammatory drug, oral antihistamine
Systemic reactions	
Low incidence (< 0.05 to 3.5 percent), manifestations can include: urticaria, angioedema, increased respiratory symptoms (nasal or pulmonary), increased ocular symptoms, and hypotension.	Tourniquet above injection site; aqueous epinephrine 1:1,000 IM: (adults, 0.3 mL; children, 0.01 mL per kg; readminister every 10 minutes if systemic symptoms persist, up to three times); diphenhydramine, IM or IV (adults, 25 to 50 mg; children, 1 to 2 mg per kg); histamine H_2 receptor blockers IV or orally for epinephrine-resistant hypotension; IV fluids or vasopressors, as needed; consider glucagon if patient is taking a beta blocker; consult allergist before any additional doses.
Delayed reaction	
May be local or systemic	Oral antihistamine (liquid is preferred); prednisone, 5 to 20 mg orally every 12 hours for two doses (depending on the patient's weight); epinephrine is not helpful; consult allergist for dose adjustment.

IM = intramuscular; IV = intravenous.

TABLE 6
Checklist for Safe Vaccine Administration

Identify the patient.
Analyze the health status of the patient before every injection. The risk of anaphylaxis is increased if the patient:
Has a fever, is acutely ill, or has a newly reported illness.
Is having an exacerbation of asthma or respiratory difficulties.
Is having an exacerbation of allergy symptoms.
Is taking new medications, namely beta blockers and angiotensin-converting enzyme inhibitors.
Inquire about any reactions occurring with the previous injection and consult with the allergist about appropriate adjustments to therapy.
Institute a checklist to reduce clerical and nursing errors:
Identify the patient's record by name and, preferably, photograph.
Check the identity, expiration date, concentration, and cap color of the vaccine vial.
Record the proper dose of vaccine on the immunotherapy record and the arm used to administer the vaccine. Alternate arms.
Draw the proper dose.
Administer the vaccine only after the patient's identity has been rechecked by comparing the patient's name with the name on the vial from which the vaccine is taken.
Remind the patient to remain in the office for 30 minutes following the injection. Check the injection site before the patient leaves.
Strenuous exercise one hour before and two hours after the injection increases the chance of anaphylaxis and should be avoided.
Document any adverse reactions.

TABLE 7
Equipment and Medications Needed to Treat Anaphylaxis

Stethoscope
Tourniquet
Equipment for monitoring blood pressure
Large-bore (14-gauge) IV catheter
Epinephrine, 1:1,000 for IM injection
Oxygen
Oral airway
Equipment for administering IV fluids
H_1 and H_2 antihistamines for injection
Corticosteroid for IV injection
Vasopressor
Glucagon for use in patients receiving beta-adrenergic blocking agents

IV = intravenous; IM = intramuscular.

The immunotherapy dose should not be increased in a pregnant patient until after delivery.

Anaphylaxis is the most serious risk related to allergen immunotherapy. The vaccines must be administered in a setting with trained professionals who are equipped to recognize and treat anaphylaxis[22] *(Table 7)*. A retrospective study found that most systemic reactions occurred within 30 minutes of injection.[23] Hence, the current recommendation is to allow at least 20 to 30 minutes of observation following an injection. Patients who have had a systemic reaction after more than 30 minutes following an injection require longer observation; in addition, they should be given injectable epinephrine to carry and instructions about how to use it. Nonetheless, reactions may occur without warning signs or symptoms, and documentation of informed consent must be obtained from the patient *(Figure 1)*.

Assessment of Immunotherapy Efficacy

After one year on a maintenance dose, clinical improvement should be apparent.[24] The therapy often may be discontinued after three to five years because by then the disease course has been altered.[11] Evaluation by an allergist, at least annually, should include monitoring of adverse reactions, assessment of efficacy, reinforcement of compliance and safe administration of immunotherapy, and determination of whether adjustments in the dosing schedule or allergen content are necessary.

The authors indicate that they do not have any conflicts of interest. Sources of funding: none reported.

The authors thank Barbara J. Kuyper, Ph.D., and David M. Siegel, M.D., for assistance with the manuscript.

REFERENCES

1. Allergen immunotherapy: therapeutic vaccines for allergic diseases. Geneva: January 27-29, 1997. Allergy 1998;53(44 suppl):1-42.
2. Noon L, Cantab BC. Prophylactic inoculation against hay fever. Lancet 1911;1:1572-4.
3. Pichler CE, Helbling A, Pichler WJ. Three years of specific immunotherapy with house-dust-mite extracts in patients with rhinitis and asthma: significant improvement of allergen-specific parameters and of nonspecific bronchial hyperreactivity. Allergy 2001;56:301-6.

Informed Consent

Date:

Patient name:

Date of birth:

I have been made aware by _____ of the following:

During the build-up phase of my allergen immunotherapy (allergy injections), I agree to come every seven to 10 days to safely increase my vaccines at every visit. If more than 10 days have passed since my last visit, my dose will be adjusted as necessary.

Local reactions are not uncommon. I will monitor the size of the reactions and the length of time they last and inform the medical staff.

Generalized reactions occur less commonly and may include symptoms of itching of the skin; sudden itching of the nose, mouth, ears, and throat; hives, wheezing, coughing, tightness of the chest, plugging of the nose, or sneezing. Although rare, serious reactions may result in significant respiratory reactions or anaphylactic shock, which may be life-threatening. A serious reaction usually occurs within 30 minutes after an injection.

I agree to remain in the medical facility for 30 minutes after my injections and to immediately report any symptoms to the medical staff.

I have had the opportunity to have all of my questions about allergen immunotherapy answered to my satisfaction. I have been informed of the potential risks and benefits of allergen immunotherapy and available alternative therapies.

Signature of patient or guardian: _____

Date: _____

Signature of witness: _____

Date: _____

Figure 1. Example of informed consent form for allergen immunotherapy.

Strength of Recommendations

Key clinical recommendation	SOR labels	References
Allergen immunotherapy is effective in patients with allergic rhinitis, allergic conjunctivitis, allergic asthma, and allergic reactions to Hymenoptera venom.	A	3, 4, 5, 6
The allergens for which immunotherapy is known to be effective are Hymenoptera venom, pollens, cat dander, dust mites, cockroach, and fungi.	A B	5, 6, 8, 10 7, 9
In patients who had received three to four years of immunotherapy, a marked reduction in allergy symptom scores and antiallergic medication usage, as well as an alteration in the natural course of allergic disease, was demonstrated.	A	11
Immunotherapy for allergic rhinitis may reduce the risk for later development of asthma in children.	B	12, 13
Early treatment with allergen immunotherapy in children who were sensitive only to house dust mites reduced development of sensitivity to other allergens.	C	14
Patients with severe, poorly controlled asthma are at higher risk for systemic reactions to immunotherapy injections than patients with stable, well-controlled asthma.	B	20

4. Adkinson NF Jr, Eggleston PA, Eney D, Goldstein EO, Schuberth KC, Bacon JR, et al. A controlled trial of immunotherapy for asthma in allergic children. N Engl J Med 1997;336:324-31.
5. Ross RN, Nelson HS, Finegold I. Effectiveness of specific immunotherapy in the treatment of asthma: a meta-analysis of prospective, randomized, single or double-blind, placebo-controlled studies. Clin Ther 2000;22:329-41.
6. Abramson MJ, Puy RM, Weiner JM. Is allergen immunotherapy effective in asthma? A meta-analysis of randomized controlled trials. Am J Respir Crit Care Med 1995;151:969-74.
7. Varney VA, Edwards J, Tabbah K, Brewster H, Mavroleon G, Frew AJ. Clinical efficacy of specific immunotherapy to cat dander: a double-blind, placebo-controlled trial. Clin Exp Allergy 1997;27:860-7.
8. Olsen OT, Larsen KR, Jacobsan L, Svendsen UG. A one-year, placebo-controlled, double-blind house-dust-mite immunotherapy study in asthmatic adults. Allergy 1997;52:853-9.
9. Kang BC, Johnson J, Morgan C, Chang JL. The role of immunotherapy in cockroach asthma. J Asthma 1988;25:205-18.
10. Dreborg S, Agrell B, Foucard T, Kjellman NI, Koivikko A, Nilsson S. A double-blind, multicenter immunotherapy trial in children, using a purified and standardized *Cladosporium herbarum* preparation. I. Clinical results. Allergy 1986;41:131-40.
11. Durham SR, Walker SM, Varga EM, Jacobson MR, O'Brien F, Noble W, et al. Long-term clinical efficacy of grass-pollen immunotherapy. N Engl J Med 1999;341:468-75.
12. Moller C, Dreborg S, Ferdousi HA, Halken S, Host A, Jacobsen L, et al. Pollen immunotherapy reduces the development of asthma in children with seasonal rhinoconjunctivitis (the PAT-study). J Allergy Clin Immunol 2002;109:251-6.
13. Grembiale RD, Camporota L, Naty S, Tranfa CM, Djukanovic R, Marsico SA. Effects of specific immunotherapy in allergic rhinitic individuals with bronchial hyperresponsiveness. Am J Respir Crit Care Med 2000;162:2048-52.
14. Des Roches A, Paradis L, Menardo JL, Bouges S, Daures JP, Bousquet J. Immunotherapy with a standardized *Dermatophagoides pteronyssinus* extract. VI. Specific immunotherapy prevents the onset of new sensitizations in children. J Allergy Clin Immunol 1997;99:450-3.
15. Nelson HS. The use of standardized extracts in allergen immunotherapy. J Allergy Clin Immunol 2000;106(1 pt 1):41-5.
16. Nelson HS. Effects of preservatives and conditions of storage on the potency of allergy extracts. J Allergy Clin Immunol 1981;67:64-9.
17. Nelson HS, Ikle D, Buchmeier A. Studies of allergen extract stability: the effects of dilution and mixing. J Allergy Clin Immunol 1996;98:382-8.
18. Joint Task Force on Practice Parameters. Allergen immunotherapy: a practice parameter. Ann Allergy Asthma Immunol 2003;90(1 suppl 1):1-40.
19. Lamson RW. Sudden death associated with injection of foreign substances. JAMA 1924;82:1090.
20. Lockey RF, Nicoara-Kasti GL, Theodoropoulos DS, Bukantz SC. Systemic reactions and fatalities associated with allergen immunotherapy. Ann Allergy Asthma Immunol 2001;87(1 suppl 1):47-55.
21. Turkeltaub P. Deaths associated with allergenic extracts. FDA Medical Bulletin 24, May 1994.
22. Pumphrey RS. Lessons for management of anaphylaxis from a study of fatal reactions. Clin Exp Allergy 2000;30:1144-50.
23. Greenberg MA, Kaufman CR, Gonzalez GE, Rosenblatt CD, Smith LJ, Summers RJ. Late and immediate systemic-allergic reactions to inhalant allergen immunotherapy. J Allergy Clin Immunol 1986;77:865-70.
24. McHugh SM, Lavelle B, Kemeny DM, Patel S, Ewan PW. A placebo-controlled trial of immunotherapy with two extracts of *Dermatophagoides pteronyssin*us in allergic rhinitis, comparing clinical outcome with changes in antigen-specific IgE, IgG, and IgG subclasses. J Allergy Clin Immunol 1990;86(4 pt 1):521-31.