

Wheat Allergy: Clinical and Laboratory Findings

Zahra Pourpak^a Mahboubeh Mansouri^a Mehrnaz Mesdaghi^a
Anoushiravan Kazemnejad^b Abolhasan Farhoudi^a

^aImmunology, Asthma and Allergy Research Institute, Tehran University of Medical Sciences, and

^bDepartment of Biostatistics, School of Medicine, Tarbiat Modares University, Tehran, Iran

Key Words

Food hypersensitivity · Immunoglobulin E · Skin tests · Wheat

Abstract

Background: Food allergy affects 6–8% of infants and wheat allergy is one of the common food allergies among children. The clinical and laboratory manifestations of wheat allergy were evaluated in this study.

Methods: Thirty-two children (≤ 12 years old) with suspected wheat allergy were evaluated for wheat allergy. The patients underwent wheat skin prick test (SPT), measurement of wheat-specific IgE and wheat challenge test. The patients with a convincing history of anaphylaxis following ingestion of wheat or with a positive challenge test, and those with a history of immediate hypersensitivity reaction following ingestion of wheat in addition to a positive wheat SPT and/or positive wheat-specific IgE were considered wheat allergic. Then, the laboratory and clinical manifestations of their disease were studied.

Results: Among patients with suspected wheat allergy, 24 patients with definite wheat allergy were identified. Anaphylaxis was a dominant clinical feature, accounting for 54.1% of acute symptoms. Chronic allergy symptoms like asthma and eczema were noted in 50% of the patients. Wheat-specific IgE was higher in patients with

anaphylaxis ($p < 0.02$) and the risk of anaphylaxis was 14.4 times more in patients with wheat-specific IgE equal to or more than 3+. **Conclusions:** Anaphylaxis had occurred in a remarkable number of patients repeatedly, which demonstrates the severity of the reactions, poor knowledge of the disease and probable existence of more patients with mild reactions. Regarding the higher level of wheat-specific IgE in patients with anaphylaxis, wheat-specific IgE could be used to predict the severity of symptoms.

Copyright © 2004 S. Karger AG, Basel

Introduction

Food allergy denotes an adverse reaction to foods or food additives in which an underlying immunologic mechanism can be shown [1, 2]. Its incidence in young children is about 1.3% and among adults about 0.3% [1]. It is more prevalent in the first few years of life and affects about 6–8% of infants [3]. Food allergy plays a pathogenic role in up to 40% of infants and small children with moderate to severe atopic dermatitis [1, 2]. Hypersensitivity reactions type I–IV can cause food allergy, but IgE-mediated reactions (type I) are the dominant mechanism [4, 5]. Food allergy may manifest itself by local symptoms or systemic reactions (as anaphylactic shock). In many

KARGER

Fax + 41 61 306 12 34
E-Mail karger@karger.ch
www.karger.com

© 2004 S. Karger AG, Basel
1018–2438/04/1332–0168\$21.00/0

Accessible online at:
www.karger.com/iaa

Correspondence to: Dr. Zahra Pourpak
Immunology, Asthma and Allergy Research Institute
Children Medical Center, No. 62, Gharib St, Keshavarz Blvd
PO Box 14185-863, Tehran 14194 (Iran)
Tel. +98 21 693 5855, Fax +98 21 642 8995, E-Mail zpourpak@hbi.ir

emergency wards, food allergies have been reported as the main and most common cause of anaphylaxis, which begins during the first 30 min after ingestion of the allergenic food and demands emergency medical care [5]. The main target organs involved in food allergies are cutaneous, respiratory and gastrointestinal systems [5–7].

Cereal families, including wheat, barley, corn, rice, rye and oat, are food materials consumed in large quantities all over the world [8]. Cereal grains account for about 70% of the world protein consumption. The main sources of protein and carbohydrate intake especially in Asian countries are cereals [5, 8]. Wheat is among the major foods that induce food hypersensitivity reactions in children [9–12]. Different data about prevalence of allergy to cereals are available. In a study of pediatric patients, 70 of 225 children were found to have at least one positive skin prick test (SPT) to one of the cereals (including wheat, barley, rice, corn, oat and rye), but only 15 patients had at least one positive double-blind placebo-controlled food challenge to cereal grain [13]. Another study in 196 children and adolescents with food allergy has shown that wheat allergy accounts for 22% of food allergies [10].

Similar to other IgE-mediated food allergies, symptoms of wheat allergy become apparent within 30 min after ingestion of the culprit food with cutaneous, respiratory and gastrointestinal manifestations, which may lead to anaphylaxis [14]. These reactions are clinically distinct from other adverse reactions to wheat proteins, including baker's asthma, celiac disease and wheat-dependent, exercise-induced anaphylaxis [15–17]. Although there are some similarities, differences are present in wheat-specific allergens, patterns of exposure, immunologic mechanism and pathophysiology [11].

Regarding the considerable prevalence of wheat allergy among food allergies [9–12] and as the patients referred to our clinic had severe symptoms due to wheat allergy, the present study was done to find out common clinical and laboratory manifestations of wheat allergy, for a better recognition of the disease and accordingly to increase our knowledge of this illness.

Material and Methods

In a 1-year period, a group of children younger than 12 years old (mean 2.4 years) who were referred to the Allergy and Immunology Clinic of Children Medical Center with cutaneous, respiratory or gastrointestinal manifestations of allergy were examined. Among them, 32 children with suspected wheat allergy were selected for this study. They had a history of allergic reaction following the ingestion of wheat products or they had a positive SPT or specific IgE for wheat

allergens in the first step of allergy evaluation. After having explained the laboratory and clinical evaluations to their parents and with their consent, a questionnaire was filled out for all patients, including information such as the patients' personal history, family history of allergy, breast-feeding state, the time of the introduction of solid foods, the onset of the disease, repeated symptoms after ingestion of wheat, emergency care, and hospitalization. After a careful physical examination, the tests were performed, including the following discussed below.

Eosinophil Counting

An eosinophil count of more than 450 cells/ μ l was considered as eosinophilia [18].

Skin Prick Test

All of the patients underwent SPT with wheat flour extract, except those with acute skin lesions, urticaria, asthma exacerbation or a history of anaphylaxis during recent months, in whom this test was delayed until they had recovered from the disease. A positive and a negative control were also included in the test. Normal saline was used as a negative control and histamine dihydrochloride 1 mg/ml as a positive control. The SPT responses were read for histamine at 10 and for allergens at 20 min. It was considered positive if the diameter of the wheal was at least 5 mm for histamine and at least 3 mm for allergens, without reaction to the negative control. According to the patient's history, SPTs were also done for some other food allergens in children (such as milk, egg, soy, nuts, seafoods, potato and orange). The extracts were obtained from Hollister-Stier, Miles, Canada.

Total Serum IgE

Total serum IgE was measured, using the ELISA method [19] (Clonesystems, Hamburg, Germany) and was compared to the mean normal value for age [20].

Wheat-Specific IgE

After sampling 1 mm of peripheral blood, sterile serum was separated and immediately frozen in -80°C . Wheat flour-specific IgE was measured using the ELISA method [19] (Clonesystems). According to the kit manual, the results were categorized into seven groups (0–6+). The results equal to 1+ or more were considered positive.

Elimination Diet

After these evaluations, cereal products were eliminated from the patients' diet for 1–2 weeks [5].

Open Food Challenge

Although double-blind placebo-controlled food challenge is the gold standard for diagnosing food allergies in infants and young children (especially less than 3 years old) whose allergy symptoms are predominantly objective (rash, hives, vomiting, diarrhea, and decreased pulmonary function) and the effect of suggestion is minimal, open food challenge seems suitable to prove food allergy [21, 22]. After the elimination diet, if the patient became symptom free, an open food challenge test with wheat flour was done, except for those (1) who had an obvious history of life-threatening reactions (systemic allergic reactions affecting at least 2 organs simultaneously and making an emergency medical intervention necessary [23]) following the ingestion of wheat, at least once during the 2 previous years [12] and (2) with a clear history of immediate hypersensitivity reaction follow-

ing the ingestion of wheat, a good response to wheat elimination and a positive wheat-specific IgE and/or SPT.

The diagnosis of wheat allergy in these groups is definite and the food challenge test is superfluous [24, 25].

Those children taking an antihistamine were advised not to take it for a certain period of time before provocation, based on the type of the antihistamine [26]. Each patient came in a fasting state to our clinic early in the morning, where all emergency equipment was at hand. Before the challenge test, a complete physical examination was done for every patient. If there were any signs or symptoms of allergy, which could alter the results of the study such as skin lesions, wheezing and rhinitis, the food challenge test would be canceled. The amount of bread for the challenge test was determined to be equal to the mean daily intake. The bread for the challenge test was a single type, obtained from a single bakery. The bread was divided into 5 pieces. The time interval between having the pieces of bread was 30 min. The patients were under close observation. The results of the food challenge were considered positive by a pediatric allergy specialist, if one or more of the following objective clinical reaction(s) were noted: urticaria, angioedema, wheezing, vomiting, diarrhea, abdominal pain, exacerbation of eczema or anaphylactic shock. At the end of the challenge test, the patient was observed for 4–8 h. If the patient had any clinical reaction during the test, it was stopped and medical intervention was given; then the patient was observed for 6–12 h or as an inpatient. The patients were advised to record and report any symptoms they experienced after discharge during the first day, or return to hospital if they had severe symptoms.

Statistical Analysis

t test was used to compare the means and Fisher's exact test was employed to compare the groups. Odds ratio was calculated to find out the risk of anaphylaxis according to the wheat-specific IgE level.

Results

Of 32 patients with suspected wheat allergy according to their history and primary evaluation, 27 (84.3%) had positive wheat SPT and 29 (90.6%) had positive wheat-specific IgE. History of life-threatening reactions was found in 13 patients. The performance of the challenge test is not allowed in this group. The challenge test was done in 11 patients. They were clearly positive in 3 patients (27.2%), and 8 patients with negative or suspicious challenge tests were excluded from the study. Systemic responses (nausea, vomiting, hypotension, dyspnea and generalized urticaria) occurred in 2 patients and the other patient had a response in the form of facial morbilliform rashes. [Of the excluded patients, 4 had positive wheat SPT and 7 had positive wheat-specific IgE (with low concentrations). Regarding the higher diagnostic value of the challenge test [27] and as these patients did not have relevant symptoms, we did not consider them as wheat allergic.] The challenge test was performed neither in patients with anaphylaxis nor in those with a clear his-

tory of immediate allergic reaction following an ingestion of wheat in addition to positive wheat SPT and/or specific IgE. The patients with a history of anaphylaxis following ingestion of wheat (13 patients), those with a positive wheat challenge test (3 patients) and the patients with a definite history of type I hypersensitivity reaction following ingestion of wheat accompanied by positive wheat SPT and/or specific IgE (8 patients) were considered wheat allergic.

Finally, 24 patients with a definite diagnosis of wheat allergy were identified. The range of age of wheat-allergic patients was 8 months to 8 years (mean 2.4 years). Of the patients, 17 (71%) were male and 7 (29%) female. The onset of the disease was before 6 months of age in 10 (41.6%) and 6–12 months in 14 (58.3%) patients (mean 5.7 months). All of the patients had experienced the disease within the first year of life and had a clear history of immediate allergic reactions following the ingestion of wheat products. Fourteen patients (58.3%) had respiratory symptoms (containing cough, wheezing, dyspnea and rhinoconjunctivitis), 20 (83.3%) had cutaneous manifestations (containing urticaria, angioedema and erythema) and 6 (25%) had gastrointestinal symptoms (containing nausea, vomiting, diarrhea and abdominal pain). Thirteen patients (54.1%) had at least 1 episode of anaphylaxis, while 5 had at least 3 episodes. The age at first anaphylaxis was 4–9 months in all of these patients (table 1). Anaphylaxis episodes started in all patients with cutaneous manifestations (generalized urticaria with or without flushing, angioedema and itching), rhinoconjunctivitis and then wheezing or cough, which were accompanied by nausea, vomiting, abdominal pain, palpitation and hypotension in some patients and had resulted in emergency ward admission in all of the patients. Beside acute hypersensitivity reactions, 12 patients (50%) had a chronic manifestation of allergy. Of those, 8 (75%) had asthma, 6 (50%) had eczema and there was no report of chronic gastrointestinal manifestations. Solid foods were introduced at 3–9 months of age in all patients. Of the patients, 19 (79.1%) had a positive history of allergy (rhinitis, eczema, asthma) in at least one of their parents and 4 (16.6%) in both parents. In 68.4% (13/19) of them the patient's mother suffered from allergy.

Wheat SPTs were positive in 23 (95.8%) and wheat-specific IgE was positive in 22 (91.6%) patients. Mean wheat-specific IgE was 73.8 Ucs/ml (Ucs represents Clonesystems units) in patients with anaphylaxis and 24.8 Ucs/ml in patients without anaphylaxis, which shows a statistically significant difference ($p < 0.02$). Anaphylaxis was more common in patients with wheat-spe-

Table 1. Immediate allergic reactions following ingestion of wheat, total serum and wheat-specific IgE of wheat-allergic patients

No.	Sex/age ¹	Respiratory symptoms	Cutaneous symptoms	Gastro-intestinal symptoms	History of anaphylaxis	Age of first anaphylaxis (months)	Anaphylaxis episodes	Total serum IgE (IU/ml)	Wheat-specific IgE
1	M/8	+	+	–	+	6	1	230	6+
2	M/1.5	–	+	–	+	7	2	280	6+
3	M/2	+	+	–	–	–	–	198	6+
4	F/2	+	+	+	+	4	2	110	3+
5	M/1	–	+	–	–	–	–	1,000	4+
6	M/2.6	+	+	+	–	–	–	830	6+
7	M/3	+	+	–	+	6	3	120	6+
8	F/2.7	–	+	–	+	4.5	3	88	6+
9	M/3.2	+	+	–	+	4	2	190	6+
10	M/2.2	–	+	+	+	6	2	1,400	6+
11	F/1.1	+	+	–	+	6	2	1,180	4+
12	M/1.5	–	–	–	+	9	4	60	4+
13	M/3.5	+	+	+	+	6	1	66	4+
14	M/1.2	–	+	+	–	–	–	320	3+
15	F/1	–	+	–	–	–	–	250	–
16	F/4	+	–	–	+	7	5	110	4+
17	M/2.1	+	+	–	–	–	–	1,200	1+
18	M/4	–	–	+	+	6	4	550	6+
19	M/1	–	+	–	–	–	–	1,000	2+
20	F/4	+	+	–	–	–	–	3,600	–
21	F/1.6	+	+	–	–	–	–	160	4+
22	M/1.5	+	+	–	–	–	–	700	2+
23	M/5	+	+	–	–	–	–	420	2+
24	M/0.7	–	–	–	+	7	2	37	2+

Age is given in years.

Table 2. Common food allergens in patients with wheat allergy (according to SPT)

Food allergens	Positive SPT
Soy	13 (54%)
Egg	13 (54%)
Milk	7 (29%)
Fish	3 (12%)
Almond	2 (8%)
Peanut	2 (8%)
Potato	2 (8%)
Orange	2 (8%)

cific IgE equal to or more than 3+ (≥ 3.6 Ucs/ml; $p < 0.02$) and the chance of anaphylaxis was 14.4 times more in these patients than in those with wheat-specific IgE less than 3+ (< 3.6 Ucs/ml; odds ratio = 14.4, 95% confidence interval = 1.36–152.5).

All of the patients had high total serum IgE for their age. Total serum IgE was equal to or less than 100 IU/ml in 4 (16.6%), 101–300 IU/ml in 9 (37.5%), 301–1,000 IU/ml in 7 (29.1%) and more than 1,000 IU/ml in 4 patients (16.6%). Eosinophilia (eosinophils > 450 cells/ μ l) was detected in 11 patients (45.8%), and eosinophil count was more than 700 cells/ μ l in 8 patients (33.3%).

Among the patients with wheat allergy, 20 also had a positive SPT for other food allergens, which are shown in table 2. The most common food allergens according to SPT (after wheat) were soy, egg, milk and seafood (without any history of related systemic reactions), respectively.

Discussion

Anaphylaxis was a common manifestation of wheat allergy in this study, although it is commonly believed that allergic reactions to wheat are rarely severe [28] and

wheat allergy occasionally causes anaphylactic reactions [29]. The rate of severe reactions seems to be higher in our country. Because our clinic is a referral center, to which mostly patients with severe diseases are referred, the real number of patients with wheat allergy who have mild symptoms and are never diagnosed might be higher and the rate of anaphylaxis in all wheat-allergic patients might be lower.

An open food challenge test is recommended for the diagnosis of food allergy in young children especially those less than 3 years old [22] and as the mean age of our patients was less than 3 years (2.4 years), the open food challenge test seems suitable to confirm the diagnosis. The challenge test was positive in 3 patients with a life-threatening response in 2 of them. Therefore, taking enough safety measures in wheat challenge tests is recommended, as Reibel et al. [30] have emphasized, too. They showed that in wheat, egg, cow's milk and soy challenge tests, the patient's history of wheat allergy is an indicator of the probability of a positive challenge test and the need for medical intervention. Regarding the high prevalence of acute anaphylactic reactions following the ingestion of wheat, the establishment of an intravenous access before the wheat challenge test is recommended. Repeated episodes of anaphylaxis (2 times or more) in patients with wheat allergy show that there is not enough awareness of the danger of wheat allergy among patients and physicians; therefore, patients with a wheat allergy should be educated enough to prevent a recurrence of symptoms and to use adrenalin in emergency situations, and the physician should focus on such an important illness.

According to the results of this study, atopy and family history of allergy were reported by 79.1% of the patients, which is compatible with other studies [31, 32]. Also, 68.4% of these patients' mothers were atopic. There may be an X-linked inheritance, as 71% of our patients were boys. Is there really any relationship between wheat allergy and sex? Further investigations are needed to evaluate this relationship.

The introduction of solid foods was at 3–9 months of age in all patients, which is almost concurrent with the mean age at disease onset (5.7 months). Regarding the simultaneous introduction of solid foods and onset of the disease, and the health of the children during the exclusive breast-feeding period, it seems that wheat allergens do not pass into the breast milk and in contrary to cow's milk allergy, consumption of wheat by the mothers usually does not precipitate allergy symptoms in their children [33]; therefore, the mother's abstinence cannot help in the treatment. The onset of the clinical manifestation of

wheat allergy was within the first year of life in all patients. Therefore, it is recommended that introduction of solid foods should be delayed until the child is 6 months old and wheat should not be used in the first year of life in atopic children. More research is recommended to clarify whether such a delay can prevent this sensitization or just delay it.

As shown in table 2, the most common allergenic foods among patients with wheat allergy, after wheat, were eggs, soy and cow's milk. Although cow's milk is one of the most common food allergens among children [5], according to the results of this study, cow's milk allergy is less prevalent than egg and soy among children with wheat allergy, which should be noticed by physicians in the determination of the diet of wheat-allergic children.

All of the patients had a total serum IgE level higher than the mean normal value for their age [20], and 16.6% of patients had an IgE level higher than 1,000 IU/ml. High levels of serum IgE are found in 60% of allergic patients [5], but all patients with wheat allergy in this study had high serum IgE, and 91.6% had positive wheat-specific IgE and 70.8% had wheat-specific IgE equal to 3+ or more. This can show the importance of type 1 (IgE-mediated) hypersensitivity reactions in these patients. Considering the results of this study, there is a relationship between the wheat-specific IgE level and anaphylaxis. High specific IgE concentration can predict a severe reaction. It is compatible with the studies of Sampson [27, 34] who showed that the concentration of hen's egg, cow's milk, peanut and fish-specific IgE can predict the severity of food allergy symptoms.

Finally, we recommend (1) exclusive breast-feeding until 6 months of age and avoiding wheat especially in atopic children in the first year of life, (2) education of the patient's family to avoid wheat products and how to apply emergency interventions in emergency situations, (3) increasing the awareness in physicians and medical staff as regards prevalence and danger of wheat allergy, and (4) taking enough safety measures in wheat challenge tests.

Acknowledgment

We thank Miss Ghiasi and Mr. Toosi for their kind cooperation in this study and Dr. Ashrafi for editing the manuscript.

References

- 1 Sicherer SH, Sampson HA: Food hypersensitivity and atopic dermatitis: Pathophysiology, epidemiology, diagnosis, and management. *J Allergy Clin Immunol* 1999;104:S114–122.
- 2 Eigenmann PA, Sicherer SH, Borkowski TA, Cohen BA, Sampson HA: Prevalence of IgE-mediated food allergy among children with atopic dermatitis. *Pediatrics* 1998;101:E8.
- 3 Bock SA: Prospective appraisal of complaints of adverse reactions to foods in children during the first 3 years of life. *Pediatrics* 1987;79:683–688.
- 4 Sicherer SH: Manifestations of food allergy: Evaluation and management. *Am Fam Physician* 1999;59:415–424, 429–430.
- 5 Sampson HA: Adverse reactions to foods; in Middleton E, Reed CE, Ellis EF, Adkinson NF, Yunginger JW, Busse WW (eds): *Allergy Principles and Practice*. St Louis, Mosby, 1998, vol 2, pp 1162–1182.
- 6 Burks AW, James JM, Hiegel A, Wilson G, Wheeler JG, Jones SM, Zuerlein N: Atopic dermatitis and food hypersensitivity reactions. *J Pediatr* 1998;132:132–136.
- 7 Kulig M, Bergmann R, Niggemann B, Burow G, Wahn U: Prediction of sensitization to inhalant allergens in childhood: Evaluating family history, atopic dermatitis and sensitization to food allergens. The MAS Study Group. Multicentre Allergy Study. *Clin Exp Allergy* 1998;28:1397–1403.
- 8 Nakase M, Usui Y, Alvarez-Nakase AM, Adachi T, Urisu A, Nakamura R, Aoki N, Kitajima K, Matsuda T: Cereal allergens: Rice-seed allergens with structural similarity to wheat and barley allergens. *Allergy* 1998;53(46 suppl):55–57.
- 9 Sampson HA, McCaskill CC: Food hypersensitivity and atopic dermatitis: Evaluation of 113 patients. *J Pediatr* 1985;107:669–675.
- 10 Sampson HA, Ho DG: Relationship between food-specific IgE concentrations and the risk of positive food challenges in children and adolescents. *J Allergy Clin Immunol* 1997;100:444–451.
- 11 James JM, Sixbey JP, Helm RM, Bannon GA, Burks AW: Wheat alpha-amylase inhibitor: A second route of allergic sensitization. *J Allergy Clin Immunol* 1997;99:239–244.
- 12 James JM, Sampson HA: An overview of food hypersensitivity. *Pediatr Allergy Immunol* 1992;3:67–78.
- 13 Jones SM, Sampson HA: The role of allergens in atopic dermatitis; in Leung DYM (ed): *Atopic Dermatitis: From Pathogenesis to Treatment*. Texas, Landes, 1996.
- 14 Varjonen E, Vainio E, Kalimo K: Life-threatening, recurrent anaphylaxis caused by allergy to gliadin and exercise. *Clin Exp Allergy* 1997;27:162–166.
- 15 Blands J, Diamant B, Kallos P, Kallos-Deffner L, Lowenstein H: Flour allergy in bakers. Identification of allergenic fractions in flour and comparison of diagnostic methods. *Int Arch Allergy Appl Immunol* 1976;52:392–406.
- 16 Sturges RP, Ellis HJ, Ciclitira PJ: Cereal chemistry, molecular biology, and toxicity in coeliac disease. *Gut* 1991;32:1055–1060.
- 17 Armentia A, Martin-Santos JM, Blanco M, Carretero L, Puyo M, Barber D: Exercise-induced anaphylactic reaction to grain flours. *Ann Allergy* 1990;65:149–151.
- 18 Lim KG, Weller PF: Eosinophilia and eosinophil-related disorders; in Middleton E, Reed CE, Ellis EF, Adkinson NF, Yunginger JW, Busse WW (eds): *Allergy Principles and Practice*. St Louis, Mosby, 1998, vol 2, pp 783–798.
- 19 Ownby DR: In vitro assays for the evaluation of immunologic reactions to foods. *Immunol Allergy Clin North Am* 1991;11:851–857.
- 20 Sly M: Allergic disorders; in Behrman RE, Kliegman RM, Jenson HB (eds): *Nelson Textbook of Pediatrics*. Philadelphia, Saunders, 2000, p 651.
- 21 Bock SA, Sampson HA, Atkins FM, Zeiger RS, Lehrer S, Sachs M, Bush RK, Metcalfe DD: Double-blind, placebo-controlled food challenge (DBPCFC) as an office procedure: A manual. *J Allergy Clin Immunol* 1988;82:986–997.
- 22 Bahna SL: Practical considerations in food challenge testing. *Immunol Allergy Clin North Am* 1991;11:843–851.
- 23 Pearlman DS, Bierman CW: Immunologic aspects of pediatric illnesses; in Stiehm ER (ed): *Immunologic Disorders in Infants and Children*. Philadelphia, Saunders, 1996, pp 603–641.
- 24 Durham SR, Church MK: Principles of allergy diagnosis; in Holgate ST, Church MK, Lichtenstein LM (eds): *Allergy*. St Louis, Mosby, 2001, pp 1–16.
- 25 Metcalfe DD, Sampson HA: Workshop on experimental methodology for clinical studies of adverse reactions to foods and food additives. *J Allergy Clin Immunol* 1990;86:421–442.
- 26 Demoly P, Michel FB, Bousquet J: In vivo methods for study of allergy, techniques and interpretation; in Middleton E, Reed CE, Ellis EF, Adkinson NF, Yunginger JW, Busse WW (eds): *Allergy Principles and Practice*. St Louis, Mosby, 1998, vol 1, pp 430–439.
- 27 Sampson HA: Food allergy. 2. Diagnosis and management. *J Allergy Clin Immunol* 1999;103:981–989.
- 28 Taylor SL, Hefle SL: Foods as allergens; in Brostoff J, Challacombe SJ (eds): *Food Allergy and Intolerance*. Philadelphia, Saunders, 2002, pp 403–412.
- 29 Takizawa T, Arakawa H, Tokuyama K, Morikawa A: Identification of allergen fractions of wheat flour responsible for anaphylactic reactions to wheat products in infants and young children. *Int Arch Allergy Immunol* 2001;125:51–56.
- 30 Reibel S, Rohr C, Ziegert M, Sommerfeld C, Wahn U, Niggemann B: What safety measures need to be taken in oral food challenges in children? *Allergy* 2000;55:940–944.
- 31 Green R, Luyt D: Clinical characteristics of childhood asthmatics in Johannesburg. *S Afr Med J* 1997;87:878–882.
- 32 Eslamnour A, Pourpak Z, Farhoudi A, Movahedi M, Gharagozlou M, Farzaneh P, Omid Z: The relationship between serum total IgE level and eosinophil count with the severity of atopic dermatitis (abstract). *Allergy Clin Immunol Int* 2000(suppl 2):191.
- 33 Gerrard JW: Cow's milk and breast milk; in Brostoff J, Challacombe SJ (eds): *Food Allergy and Intolerance*. Philadelphia, Saunders, 2002, pp 435–444.
- 34 Sampson HA: Utility of food-specific IgE concentrations in predicting symptomatic food allergy. *J Allergy Clin Immunol* 2001;107:891–896.