Clinical Features of Food Allergy during the 1st Year of Life: The ADAPAR Birth Cohort Study

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Abstract
Background: Although food allergies (FAs) occur most commonly during the 1st year of life, there is limited information available regarding the epidemiology of FAs. In this study, we investigated the incidence of FA and the associated risk factors during the 1st year of life in southeast Turkey. Methods: This study is a prospective evaluation of 1,377 infants born at the Balcalı Hospital (Çukurova University) and includes four routine follow-up assessments until the age of 1 year. At birth, a physical examination was performed, cord blood samples were taken, and parents completed a baseline questionnaire. Follow-up visits were scheduled at 3, 6 and 12 months and included the infants’ physical examination and a follow-up questionnaire. A skin prick test (SPT) was performed and food-specific IgE levels were measured at 6 and 12 months. Telephone interviews were conducted when the infants were 9 months of age, and the questionnaire was administered. The diagnosis of FA was based on food-specific IgE levels, positive SPT results, associated clinical findings and an oral food challenge (OFC) test. Results: Patient histories, physical examinations and laboratory results indicated a possible FA in 90 infants (6.5%) during the 1st year of life. All of them underwent OFC testing with the suspected foods, and FA was confirmed in 33 cases (2.4%). Cow’s milk allergy was the major cause of FA. Skin reactions were major clinical findings in FA. A family history of atopy was identified as the major risk factor for FA. Conclusions: The prevalence and risk factors of FA in our region are consistent with those reported in the literature.

Introduction

Food allergy (FA) can occur at any age, although the age group from 0 to 3 years is most commonly affected. Overperception of FA in the community, absence of a screening test, failure to diagnose the condition with a single test, and the cumbersome and time-consuming nature of provocation tests contribute to the difficulty in conducting population-based trials [1]. Similar to other parts of the world, a limited number of studies has been conducted on the epidemiology of allergic diseases in Turkey. It has been reported that the frequency of FA among infants up to 3 years of age is 6–8% in western
countries [2]. In the first birth cohort study by Altıntaş et al. [3] in our region, the incidence of cow’s milk allergy was found to be 1.55%.

Cohort studies are of fundamental importance and value in defining the mechanism responsible for the development of allergic diseases [4]. Particularly birth cohort studies help to elucidate the development of the disease through a longitudinal approach. Although there are no general recommendations for the prevention of FA, some previous studies have defined several risk factors for FA development. These factors include a family history of atopic disease [5], environmental exposure to allergens (including microbes, cigarette smoke and other pollutants) [6], age at solid food introduction and duration of breastfeeding [7, 8]. A determination of these risk factors might lead to recommendations for the prevention of FA.

Several studies have been performed to shed light on possible associations between elevated immunoglobulin (IgE) levels in the cord blood and the development of allergic diseases in later stages of life. To date, contradictory results have been reported for the association between a high cord blood IgE level and atopy. Small sample sizes and brief observation periods used in most of the studies and failure to use objective findings for allergen sensitization in some studies may have contributed to these contradictory results [9–11].

The ADAPAR (Adana Pediatric Allergy and Risk Factor) birth cohort study aimed to establish the incidence of FA and associated risk factors in infants followed up from birth until 1 year of age in Adana, a large city in Southeast Turkey on the Mediterranean Sea.

**Materials and Methods**

**Study Design**

The ADAPAR birth cohort study was a population-based study conducted in a single center on unselected subjects. It included 1,475 infants delivered at the Balcıli Hospital (Çukurova University) in Adana between February 2010 and February 2011. The local Human Research Ethics Committee approved the study. All parents gave written informed consent for the participation in the study.

A number of tests and examinations were carried out at the time of delivery and at follow-ups (fig. 1). A physician performed the clinical examination and cord blood sampling of each infant at birth. The study questionnaires about the infants were of two cue), including the first examination examined the infants, the mother of each infant filled in the follow-up questionnaire form. This way, information regarding breastfeeding and infant diet was obtained, and questions about nutritional supplements, infectious diseases, medications, vaccinations, smoking in the household, and signs and symptoms of allergic disorders were asked. At the 9-month follow-up, mothers were called via telephone and asked to complete an extended follow-up questionnaire a second time. Physical examination, skin prick tests (SPT) and specific (s)IgE level measurement were carried out when there was any suspicion of an allergic disease at that time. Additionally, SPTs were performed and sIgE was measured at 6 and 12 months of age. When follow-up examinations were missed and infants could not be examined, the mothers were contacted via phone and the same extended questionnaire was completed by them.

The 12th-month interview was completed in 1,377 (93.3%) infants, who were enrolled in the study. In addition, SPT and sIgE level measurements were carried out when there was any suspicion of allergic disease raised by history and/or physical examination.

**Cord Blood Collection and Analysis**

In all participants, the umbilical cord was sterilized and cord blood at an amount of 5 ml was drawn from the umbilical vein immediately after delivery. The sera were then centrifuged at 3,000 rpm for 15 min, and their sera were preserved at −30°C until further biochemical analyses. The ImmunoCAP® specific IgE test (UniCAP; Phadia, Upsala, Sweden) was used to measure cord blood IgE levels (expressed in kUA/l). To prevent the influence of contamination with maternal blood, in addition to IgE testing, all cord samples were also tested for IgA at the Biochemistry Laboratory of the Çukurova University on the same day. The quantifications of IgA levels were measured by the turbidimetric method (Beckman Coulter Unicel DXC 800). IgA results ≥11 mg/dl led to the exclusion of the cord blood IgE result.

**Food-Specific IgE Measurements**

At the start of the study, a screening test was carried out using an ImmunoCAP® kit in infant sera for the six most common food allergens. A positive screening test prompted the analysis of IgE levels specific to cow’s milk, hen’s eggs, soy, wheat, fish and peanuts, and levels >0.35 kU/l were considered positive.

**Skin Prick Tests**

Commercially available extracts of major inhalant allergens (Allergopharma, Germany) were used for SPTs: tree (alder, hazel, poplar, elm and sallow), mold (Alternaria alternata, Cladosporium herbarum and Fusarium moniliforme), pollen (grass, barley, oat, rye, wheat, velvet, orchard, timothy, blue grass and meadow fescue), Dermatophagoides pteronyssinus and D. farinae, and food allergens (milk, egg, wheat, peanut and banana). The SPTs included standard methods [12]; when the mean wheal size was more than 3 mm larger than the negative control, the result was deemed positive.
Diagnostic Procedure

FA was suspected in infants who had a history of any reaction related either to the skin or the respiratory/gastrointestinal system after intake of a specific food; infants with a serum sIgE >0.35 kUA/l, or infants with a positive SPT. These infants underwent elimination diet and were then offered an oral food challenge (OFC) [13]. Prior to testing, infants followed an elimination diet for 15 days. All suspected foods were eliminated from the infant’s diet or the mother’s diet when the infant was breastfed.

Standardized OFCs were performed in children with suspected FA [14]. The suspected food was administered starting at a minimum amount, and then incremental doses were given at 20-min intervals until the total challenge dose was tolerated or an adverse reaction occurred. The starting dose of the food was determined according to the history of reaction intensity, or the results of sIgE or SPT. The administered total dose was the normal daily intake of the food in question adjusted for age in months. The infants were observed for at least 2 h after the last dose before going home. The
parents of all infants were contacted via telephone 72 h after test completion in order to determine any findings suggestive of FA. FAs were defined as positive when urticaria, angioedema, vomiting, diarrhea, flare-ups of eczema, or respiratory and cardiovascular symptoms developed during the challenge procedure.

Statistical Analysis

Data were analyzed using SPSS for Windows v. 15.0. Findings in the groups with and without FA were statistically compared. Relationships between FA in infancy and various maternal, environmental and perinatal risk factors were assessed using Pearson’s χ² test. The Mann-Whitney U test was used to compare the groups’ cord blood IgE concentrations. Post hoc tests were used in all groups with significant statistical difference in ANOVA to identify the groups with a higher mean. Multiple logistic regression analysis was used to analyze the effects of factors that were identified as statistically significant or considered clinically important for FA. Results were presented as odds ratio with 95% confidence intervals. A p value <0.05 was considered statistically significant for all tests.

Results

Characteristics of the Participants

Among 1,475 potential participants, 1,377 infants (93.3%; 732 boys and 645 girls) were finally enrolled in the study. Moving of a family to another residence, severe disease or poor parental cooperation were the main causes of failure to participate. The majority (920 infants) underwent at least one SPT and/or food-specific IgE determination by the end of 1 year (table 1).

There were no significant differences with respect to race/ethnicity between participants and those who declined. The characteristics of the infants enrolled have been described elsewhere [15].

Cord Blood IgE and IgA Levels

Cumulative IgA levels for all cord blood samples were <11 mg/dl. The mean cord blood IgA value was 8.5 ± 2.2 mg/dl. No statistically significant relationship was observed regarding cord blood IgA and IgE levels (p > 0.05). The mean cord blood IgE level of the 1,377 infants was 28.9 ± 19.2 kU/l (median: 25.0 kU/l, range, 13–49 kU/l). We did not find any relationship between cord blood IgA level and FA outcome (p > 0.05).

Clinical Characteristics and Laboratory Findings of FA Infants

During the study, SPT was applied at least once and/or sIgE was determined in 920 infants, and 115 tests were positive (115/920 = 12.5%). Based on the positive sIgE and/or SPT, 115 foods were tested in total, and in 90 infants, reactions were considered suspicious (90/920 = 9.8%).

OFC was not performed in 1 infant because of a history of anaphylaxis following cow’s milk consumption. In
total, 114 OFC tests were performed on 89 infants with positive SPTs and/or positive food-specific IgE. Among all OFCs, 38 tests (38/114 = 33.3%) were found to be positive. A positive OFC was observed in 32 (32/89 = 35.9%) infants (fig. 2).

In 11 of 13 infants with a positive history as well as a positive SPT and sIgE levels, the OFC test yielded a positive result (84.6%). However, OFC was found to be positive in 4 of 8 infants with a positive clinical history and at least one positive laboratory test (sIgE and/or SPT). Interestingly, positive OFCs were found in 18 of 93 patients (19.3%) whose parents were not aware of food-related symptoms despite the positive sIgE and/or SPT. OFC was also performed in 3 infants with a negative SPT and sIgE levels because their families reported allergic symptoms; consequently, FA was diagnosed in 1 of them. In fact, this patient was regarded as having non-IgE FA (fig. 2). The frequencies of sensitization (SPT/sIgE) and OFC results at each follow-up are shown in table 1.

FA was diagnosed in 33 of 1,377 infants (2.4%). Of the infants diagnosed with FA, 66.6% were male (n = 22) and 33.4% (n = 11) were female. The age at diagnosis was 6.9 ± 2.8 months. Frequencies of FA are presented for each follow-up visit in table 1. Cutaneous symptoms (urticaria, maculopapular rash, angioedema and eczema) were the most common symptoms occurring during food challenge (n = 28; 74%). As the second most common symptom, gastrointestinal symptoms (vomiting, nausea and food rejection) were observed in 7 patients (18%); 3 patients showed respiratory symptoms (bronchospasm, nasal discharge and nasal stuffiness; table 2). Twenty-two reactions (57.9%) developed during the first 2 h (early-onset reactions) while 42.1% (n = 16) occurred within 72 h (late-onset reactions). Early-onset reactions included cutaneous (56.5%, n = 13), gastrointestinal (30.4%, n = 7) and respiratory tract (8.7%, n = 2) reactions, whereas late reactions included cutaneous (93.8%, n = 15) and respiratory tract (6.2%, n = 1) reactions. While all of the early-onset skin reactions were in the form of pruritus and urticaria, all of the late-onset skin reactions were exacerbations of eczema.

Culprit allergens established by OFC test included cow’s milk (n = 20, 51.3%), eggs (n = 17, 43.7%), chicken meat (n = 1, 2.5%) and bananas (n = 1, 2.5%; table 2). SPT, sIgE and OFC findings in patients diagnosed with FA are presented in table 3.
Table 4 summarizes comparisons of demographic and questionnaire findings for the groups with and without FA. Male gender was predominant among infants with FA compared to infants without FA (p = 0.027). Presence of AD, a history of wheezing and a history of atopic disease in a family member (mother, father or sibling) were found to be significantly higher in the infants with than without FA (p < 0.05).

The mean cord blood IgE value was 39.17 ± 26.3 kU/l in infants with FA and 28.56 ± 1.54 kU/l in non-FA infants; there was no statistically significant association between elevated cord blood IgE level and the development of FA (p > 0.148).
The results regarding the effects of potential risk factors for FA are presented in Table 5. Results of multiple regression analysis showed that the presence of FA in the other sibling (odds ratio 18.90, 95% confidence interval 1.59–224.05) substantially increased the risk for FA development.

**Other Allergic Diseases**

At the end of the 1st year, eczema was diagnosed in 59 (4.3%) infants. Of these infants, 12 (20.3%) showed sensitivity against 14 different foods proven by OFC. At least one wheezing attack occurred in 220 infants during the 1st year of life (16%). FA was more common in wheezy infants (4%) compared to the others (2%), and the difference was statistically significant (p = 0.036).

During the study, no inhalant allergen sensitivity was detected with SPT among the infants.

**Discussion**

Allergic diseases cause significant morbidity during childhood. Globally, and particularly in developed countries, the frequency of allergic diseases among children is on the increase and FA prevalence differs from one country to another and even between different regions of the same country [16]. Differences in the lifestyle and environmental conditions in addition to the genetic background may play important roles in the development of allergic diseases [17]. In developed countries, preventive and protective measures against allergic diseases are being developed based on risk factors established in cohort studies, which were conducted particularly during infancy and childhood. However, very few birth cohort studies have been conducted in developing countries.

Although the true incidence of FA is difficult to establish due to the differences in methodology, among children FAs are reportedly on the rise [16]. This is the first publication of a cohort study on FA in the 1st year of life in Turkey. We found that the prevalence of FA is 2.4% in children younger than 12 months of age in Adana, Turkey. The prevalence of FA in the present study is consistent with other reports in the literature [17–19]. Some other birth cohort studies have reported higher FA prevalence rates [20–22]. However, the higher prevalence observed in those studies may have been caused by the lack of food challenge testing. The only birth cohort study ever conducted in our country by Altintas et al. [3] found a prevalence of 1.55% for cow’s milk allergy in 1995 in

![Table 4. Comparisons of demographic and questionnaire findings in the groups with and without FA](image-url)

**A Birth Cohort Study on Food Allergy**

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Adana, which is comparable to the 1.45% found in our birth cohort study conducted at the same center 20 years later.

For the diagnosis of FA, a detailed analysis of the patient’s history should be followed by an evaluation based on laboratory tests, and the diagnosis should be confirmed with a food challenge test, since self-reported food hypersensitivity tends to be overestimated [23]. In only 30% of all cases, the FA diagnosis is confirmed with a FA test [2, 18]. In our study, only 18.2% of the infants with positive SPT and/or sIgE test, but without a history of FA, had a positive OFC. Consequently, parents are sometimes not aware of symptoms related to FA. This condition should be taken into consideration and parents should be informed about the findings of FA in developing countries. However, this statistics reached 50% in the presence of at least one positive laboratory test result along with an allergic history. In addition, as our findings reveal, an OFC test in infants with positive SPT and/or sIgE and a history of allergy might yield positive OFC results that exceed expectations. The main limitation of our study is the small size of infant subgroups included in the diagnostic procedure, which increased the difficulty in identifying factors predicting an FA diagnosis.

Symptoms of FA observed in several studies share some common characteristics. Skin reactions are most commonly observed among FA cases, and many studies worldwide support this observation [18, 19]. Consistent with the literature, skin reactions were the major finding (74%) during OFC in our infants.

In birth cohort studies, cow’s milk and eggs were the most common allergens responsible for FA in children younger than 12 months [21]. Similarly, foods that were most commonly associated with FA in the current study were cow’s milk (51.3%) followed by eggs (43.6%). Products containing cow’s milk (e.g. baby formula or yogurt) are major contributors because they continue to be initiated as the first primary supplemental foods in our country and region, and it may be suggested that the increased prevalence of cow’s milk allergy may be associated with current dietary habits.

Many meta-analyses have shown that relying on breastfeeding as the sole source of nutrition may contribute to the development of oral tolerance and could prevent some FAs and atopic disorders [7]. The protective effect of breastfeeding may be due to several factors: decreased exposure to allergenic food, the secretory IgA content of breast milk provides passive protection against foreign proteins and pathogens, and soluble factors in breast milk which may induce earlier maturation of the gut barrier and the infant’s immune response [24]. Moreover, different cohort studies have provided various results regarding the relationship between the risk of FA and breastfeeding. Some studies have reported an increased risk with breastfeeding whereas others reported a decreased one, and some studies even found no relationship [25]. In the present study, we could not find any relationship between breastfeeding and FA development. This result suggests that hereditary and/or environmental risk factors are more important for FA.

### Table 5. Risk factors for FA development based on results of multivariate logistic regression analysis

<table>
<thead>
<tr>
<th>Risk factors</th>
<th>Odds ratio (95% CI)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (male)</td>
<td>1.37 (0.46–4.05)</td>
<td>0.568</td>
</tr>
<tr>
<td>Birth season</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spring</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Summer</td>
<td>1.94 (0.29–12.94)</td>
<td>0.493</td>
</tr>
<tr>
<td>Autumn</td>
<td>0.76 (0.09–5.95)</td>
<td>0.796</td>
</tr>
<tr>
<td>Winter</td>
<td>1.61 (0.28–9.32)</td>
<td>0.592</td>
</tr>
<tr>
<td>Socioeconomic level</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Medium</td>
<td>0.50 (0.10–2.37)</td>
<td>0.385</td>
</tr>
<tr>
<td>High</td>
<td>1.34 (0.11–15.34)</td>
<td>0.813</td>
</tr>
<tr>
<td>Maternal vitamin E intake during pregnancy</td>
<td>0.97 (0.02–48.02)</td>
<td>0.990</td>
</tr>
<tr>
<td>Maternal folic acid intake during pregnancy</td>
<td>3.56 (0.14–88.78)</td>
<td>0.439</td>
</tr>
<tr>
<td>Maternal allergic disease</td>
<td>3.83 (0.54–26.76)</td>
<td>0.176</td>
</tr>
<tr>
<td>Paternal allergic disease</td>
<td>1.54 (0.06–38.50)</td>
<td>0.790</td>
</tr>
<tr>
<td>Allergic disease in a sibling</td>
<td>18.90 (1.59–224.05)</td>
<td>0.020</td>
</tr>
<tr>
<td>Exclusive breastfeeding time (≥4 months)</td>
<td>5.77 (0.40–81.82)</td>
<td>0.195</td>
</tr>
<tr>
<td>Cord blood IgE level</td>
<td>1.01 (0.99–1.03)</td>
<td>0.149</td>
</tr>
</tbody>
</table>

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In recent studies, in agreement with our results, male gender [26], infantile eczema [27] and a family history of atopy [28, 29] have been determined as important risk factors for the development of FA. Moreover, many studies have shown an association with AD. The relationship between AD and FA is well known. Infants with AD are reported to be sensitized mainly to food allergens [30–33]. However, others have shown that infants sensitized to food allergens may also have an increased risk for the development of wheezing and AD [34]. In this study, the frequency of wheezing and AD was found to be significantly higher among infants with than without FA.

Several studies have evaluated the association between FA and family history. The risk for allergy is reported to be increased among cases with a family history of allergy [28, 29]. Schnabel et al. [21] found that early-sensitized children with a history of parental atopy were associated with persistent FA. Interestingly, according to the results of our study, a history of allergy in a sibling was a risk factor for the development of FA.

Use of cord blood IgE as a predictor of allergic diseases in infancy has already been investigated [35]. Among these studies, a birth cohort study by Pesonen et al. [36] found a significantly higher incidence of FA and AD in subjects with elevated cord blood IgE levels, and the presence of higher IgE levels in the cord blood was a major risk factor for FA. In our study, we did not observe a statistically significant association between the development of FA and cord blood IgE levels.

Recently, a possible association between FA and food-specific IgA levels has been studied. Konstantinou et al. [37] aimed to clarify the relationship between food sIgA levels and their role in tolerance induction. They demonstrated an effect of food sIgA on tolerance induction, but did not find any correlation between peripheral blood IgA levels and tolerance. In our study, cord blood IgA levels were not associated with FA outcome.

In conclusion, in this long-term, prospective study, FA prevalence rates and risk factors were similar to those reported in the literature. Further studies are needed to elucidate the relationship between cord blood IgE levels and allergic diseases. Nowadays, there is no proven method of preventing the development of FA. We believe that our results provide further insight into FA development and trigger future studies aiming to improve the management of FA by health care services and to develop programs dedicated to the prevention of allergic diseases.

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References


8 Milner JD, Stein DM, McCarter R, Moon RY: Early infant multivitamin supplementation is associated with increased risk for food allergy and asthma. Pediatrics 2004;114:27–32.


