

DR FLORE AMAT (Orcid ID : 0000-0001-7843-7999)

PROFESSOR JOCELYNE JUST (Orcid ID : 0000-0002-5646-2429)

Article type : Invited Review

Food allergy phenotypes: the key to personalized therapy

Antoine Deschildre (1)*, Stéphanie Lejeune (1)*, Mélanie Cap (1), Sophie Flammarion (1),
Line Jouannic (3), Flore Amat (2), Jocelyne Just (2).

1 CHU Lille, Pediatric Pulmonology and Allergy Department, Hôpital Jeanne de Flandre, and
Université Nord de France, F 59000 Lille, France

2. Asthma and Allergy Center, Hôpital d'Enfants Armand-Trousseau - 26, Avenue du Dr.
Arnold Netter, 75571 Paris Cedex 12 – INSERM, UMR_S 1136, Sorbonne Universités,
UPMC Univ Paris 06 – Institut Pierre Louis d'Epidémiologie et de Santé Publique, Equipe
EPAR, 75013, Paris, France

3 CHU Lille, Nutrition Department, Hôpital Jeanne de Flandre, F 59000 Lille, France

*: Antoine Deschildre and Stephanie Lejeune contributed equally to the redaction of the
manuscript.

Corresponding author:

Antoine Deschildre,

Pneumologie et allergologie pédiatriques

Pôle enfant

Hôpital Jeanne de Flandre

This article has been accepted for publication and undergone full peer review but has not
been through the copyediting, typesetting, pagination and proofreading process, which may
lead to differences between this version and the Version of Record. Please cite this article as
doi: 10.1111/cea.12984

This article is protected by copyright. All rights reserved.

CHRU de Lille

59037 Lille cedex

France

E-mail: antoine.deschildre@chru-lille.fr

Tel: +33 3 20 44 50 72

Fax: +33 3 20 44 40 79

Contributors

Antoine Deschildre, Stéphanie Lejeune: main involvement in the writing of the manuscript.

Mélanie Cap, Flore Amat, Line Jouannic, Jocelyne Just: substantial involvement in writing and revising the manuscript prior to submission.

All the authors have approved the final version of the article

Abstract:

Food allergies (FAs) are of increasing public health concern and are characterized by a large spectrum of diseases. Their diversity is well known for immunologic pathways (IgE, non-IgE mediated FAs), and natural history. Many other factors and patient characteristics are involved including type of food, exposure route, allergic comorbidities, gender, racial and ethnic backgrounds, co-factors and health conditions. Food allergen components and sensitization profiles are also involved in FA phenotypes. A new approach to chronic disorders based on the identification of phenotypes through extensive knowledge of all the complex components is also applicable to FAs and could lead towards integrative care management. Diagnostic biomarkers for FAs are emerging which also contribute to better care modalities. The aim of this article is to highlight current knowledge regarding the phenotypic diversity of FA. This review will focus on IgE-mediated FAs and how identifying

phenotypes may help to better understand the pathophysiological complexity, improve diagnosis and lead to personalized treatment strategies.

Many non-communicable chronic diseases and cancers involve complex gene-environment interactions and are influenced by diverse features including socio-economic elements and psychological factors as well as age and gender. A new phenotypic approach has been developed taking into account extensive knowledge of these complex components to promote integrative care management, i.e., predictive, preventive, personalized and participatory (P4) medicine [1]. This concept is also applicable to allergic diseases. Mathematical modeling applied to cohorts of asthmatic patients have confirmed that asthma is not one disease but characterized by different phenotypes [2]. A similar phenotypic approach based on cohort studies is now emerging for food allergies (FAs). Clinicians are convinced that FAs are characterized by different disorders and consequently different treatment challenges. The diversity of the immunologic pathways (both IgE and non-IgE mediated) and natural history of FAs is well known. Many factors and patient characteristics are also involved: type of food, severity of the reaction, exposure route, allergic comorbidities, gender, racial and ethnic backgrounds, health condition ... (figure 1). Food allergen components and sensitization profiles are also involved in FA phenotypes. Finally, biomarkers have been identified and these biomarkers also contribute to understanding FA diversity. This review will focus on phenotypes of IgE-mediated FAs. The aim is to highlight current knowledge of these different aspects of FA and how their better understanding and a phenotypic approach may help not only to improve the diagnosis but also personalize treatment strategies.

I. Food allergy: different immunologic pathways and natural histories

As mentioned above, the diversity of the immunological mechanisms involved in FA is well known. This is illustrated by the gastro-intestinal expression of food induced allergy disorders caused by IgE-mediated mechanisms (ranging from oral allergic syndrome to digestive anaphylaxis) and non-IgE mediated or cellular mechanisms (eosinophilic disorders, food protein induced enterocolitis syndrome). Some FAs are associated with a congenital immunodeficiency, such as, for example, autosomal-dominant hyper-IgE syndrome associated with DOCK8 mutations [3] or a variant of immune dysregulation, polyendocrinopathy, enteropathy, X-linked (IPEX) syndrome, caused by mutations in the forkhead box P3 (FOXP3) gene; Although a clinical triad of intractable diarrhea, insulin-dependent diabetes, and eczematous dermatitis characterize most IPEX patients, some of them present with severe FA [4].

Another important characteristic of FAs is their natural history. Children with cow's milk and hen's egg IgE-mediated allergies usually recover in the first years of life, but also later in life as observed in cohort studies (Tables I and II) [5- 11]. Some factors are associated with the risk of persistence, such as increasing levels of specific IgEs (sIgE), or the association with a respiratory allergic comorbidity [12-13]. Tolerance of baked milk or baked egg is associated with recovery later in life in raw milk and egg FA. In contrast, peanut and tree nut allergies diagnosed in the first years of life are mainly persistent, as are allergies to fish and seafood [14].

II. Food sensitization or allergy: What have we learnt from birth cohort studies?

Using a mathematical approach, Peters *et al.* recently studied the clinical expression of allergic diseases in the first 2 years of life with an emphasis on challenge proven FA [15]. They reported the results of a latent class analysis in 5276 infants (11 to 15 months old) of the

Australian HealthNuts cohort, integrating parental, infant and environmental risk factors. In this analysis, 14 and 9.4% of the children were respectively sensitized or allergic to egg, 8 and 3% to peanut, and 2 and 0.7% to sesame. Twenty-six percent had a history of atopic dermatitis (12% before 4 months of age) and 18% had presented with one or more wheezing episodes. The authors defined five phenotypes: no atopic disease (71%); atopic dermatitis without food sensitization (16%); egg allergy only (8%); multiple FA with predominant peanut allergy (3%); multiple FA with predominant egg allergy (2%). They demonstrated that filaggrin null mutations or a history of atopy in siblings were associated with atopic dermatitis without food sensitization or with egg allergy, rather than with multiple FA. Male gender and prematurity were risk factors for multiple FA. Children with parents born in Asia were at a higher risk of FA. Wheeze was equally reported in the five latent classes suggesting other mechanisms. Havstad *et al.* evaluated the significance of atopic phenotypes using ten allergen sIgE measurements including egg, milk and peanut in 594 children aged 2 years (WHEAL cohort, 62% Afro-Americans) [16]. No food challenges were done. Four latent classes were identified, including “low or no sensitization”, “high sensitization”, “milk and egg dominated” and “peanut and inhalants”. They showed that only high sensitization was associated with a risk of asthma. Food sensitization *per se* was not a risk factor. Early multiple sensitizations, including to food, associated with an imbalance of the Th2/Th1 cytokine ratio is a risk factor for severe atopic conditions and poor lung function. This was confirmed by a latent class analysis conducted on the Manchester Asthma and Allergy Study (MAAS) birth cohort and also on the Multizentrische Allergiestudie (MAS) and the Protection against allergy: Study in Rural Environments (PASTURE) birth cohorts [17, 18]. Gabet *et al.* also showed the role of early allergic sensitization in the risk of developing an allergic disease at 6 years of age in the Pollution and Asthma Risk: an Infant Study (PARIS), a population-based prospective birth cohort [19]. In all, 1525 infants (18 months old) were

evaluated. Sensitization to at least one allergen was found in 13.8% of the population, to food allergens in 12.3% and to inhalant allergens in 2.3%. Furthermore, 6.2% of the children were multi-sensitized either to several food allergens (81.3%) or to both food and inhalant allergens (18.7%). Concerned foods were mainly cow's milk (7.6%), egg white (6.6%), hazelnut (2.3%), and peanut (1.9%), accounting for 93.3% of all the cases of food sensitization. Cluster analysis revealed three profile groups: the first comprised children who were not, or rarely, sensitized (89.2%); the second, children sensitized to one to three allergens (9.2%); and the third, children who were polysensitized (1.6%). This study highlighted that early sensitization during infancy came before ingestion exposure to food allergens, and that the prevalence of asthma, rhinitis, eczema, FA and multimorbidity at 2 years increased from the first profile through to the third ($p<0.001$). At 6 years, symptoms of current asthma, rhinitis, atopic dermatitis and allergic multimorbidity were also significantly more frequent in the second and third profiles.

All these studies confirm that early multiple sensitization, but not isolated food sensitization, is a strong risk factor for atopic disease (table III). Improving our knowledge of the underlying mechanisms by using an integrative approach (mathematical, statistical modeling) may help to prevent or to treat FA.

III Determinants of severe food allergy phenotypes

III.1 Type of food: peanut and tree nut allergies, models of severe FAs

Peanut and tree nut allergies are increasingly prevalent in Western countries. These allergies convey a high risk of severe and even fatal anaphylaxis. The severity of peanut allergy was illustrated in the MIRABEL survey (France, Belgium, and Luxemburg) which

included 785 patients with peanut allergy (children: 85%) [20]. In this study, 30% of all the patients had a history of anaphylaxis. Data on nut allergies confirm the severity of this type of allergy, particularly for cashew nuts, as illustrated by the recent data published by Van der Valk [21]. Johnson *et al.* showed that cashew was the first cause of anaphylaxis in children seen at a pediatric emergency department in Stockholm [22]. Co-sensitization to peanut and several nuts are frequently observed. In view of the severity of these allergies, more studies are required to further characterize nut allergy phenotypes [23].

III.2 Atopic comorbidities: an important feature of severe and/or persistent FA

The expression of a FA varies according to its association with other allergic diseases which can significantly increase the risk of anaphylaxis. This has been demonstrated for peanut and nut allergies [table IV]. Summers *et al.*, in a study of 1,094 patients with tree nut and peanut allergies, demonstrated not only that severe asthma was associated with life-threatening bronchospasm induced by food ingestion in these patients, but also that severe pharyngeal edema was more common in patients with severe allergic rhinitis [24]. They also found that having severe atopic dermatitis was associated with a 3-fold increased risk of becoming unconscious during an acute allergic reaction, thus further highlighting the link between the severity of acute allergic reactions and the severity of co-existing atopic diseases. The role of these allergic comorbidities was also illustrated in the MIRABEL survey which found that patients often had allergic comorbidities, including asthma (58%) or atopic dermatitis (66%), frequently associated (40%) [20]. A combined factor and cluster analysis was used to study the phenotypes in a subgroup of 247 patients who had undergone an oral food challenge (OFC) [25]. Three independent phenotypes were identified and replicated in the whole population; the most severe phenotype was associated with allergic comorbidities

(asthma and atopic dermatitis in 100% of the cases) and with the lowest eliciting dose (ED) during the OFC. The weight of asthma as a comorbidity in severe phenotypes of peanut allergy in children is also highlighted by Cousin *et al.* [26]. They conducted a retrospective study in 317 children with proven peanut allergy followed in one French allergy center. They described three clusters. The most severe group comprised 71 children and was characterized by the highest frequency of asthma (80.6%) and allergic rhinitis (90.3%), the lowest EDs and the largest proportion of anaphylactic reactions (44.4%) on OFC. Finally, asthma and rhinitis comorbidities are also more frequent in cases of persistent cow's milk or hen's egg FA [5, 6], and asthma has been shown to be a risk factor of more severe reaction or failure of food oral immunotherapy (OIT) in patients with cow's milk allergy [27].

III.3 Exposure route and severity of FA

Patients who react to food allergens on inhalation may form a specific high-risk group with frequent severe reactions and low EDs upon OFC, as observed in the MIRABEL study [20]. Comstock *et al.*, reporting a high frequency of severe reactions to peanuts and tree nuts in 41 patients (history of asthma: 68%) during flights, also showed that 58% of reactions were induced by inhalation [28].

III.4 Demographic characteristics of FA: Influence of gender and ethnic background

Gender is a determinant of FA phenotype. Multiple FAs were more prevalent in the boys of the pediatric HealthNuts cohorts [15]. In the MIRABEL survey, the two severe peanut-allergic phenotypes were more frequent in girls [25], a finding that was also reported

by Cousin *et al.* [26]. In an adult population, anaphylactic reactions are more frequent in women [29]. The influence of sex hormones on allergy development has been suggested [30].

Ethnic background can also influence the FA phenotype and severity. This parameter was evaluated by Mahdavinia *et al.* in a retrospective cohort study of children aged 0-17 years consulting at two allergy tertiary care centers in the US. The African-American or Hispanic children were found to have more severe FA profiles more commonly associated with allergic comorbidities, than the non-Hispanic white children [31]. Corn, seafood and fish allergies were more frequent in African-American and Hispanic children, wheat or soybean allergies more common in those of African-American origin, and nut allergies in white children. These differences may be attributed to cultural dietary habits, food introduction practices, different living conditions and genetic backgrounds. In the HealthNuts study, the authors also showed that children of Asian descent were more likely to have an FA to egg or multiple FAs [15].

IV Cofactors and health condition as triggers in special phenotypes

IV.1 Anaphylaxis induced by exercise: a particular form of FA

Some cofactors or underlying diseases can promote the expression of FA. This is illustrated by wheat-dependent exercise-induced anaphylaxis which was first reported in 1980 by Sheffer and Austen. It is defined as the occurrence of symptoms of allergy or anaphylaxis during generally intense or prolonged exercise in the post-prandial period. Four criteria have been described: (1) episodes of collapse (syncope) and / or upper airway obstruction with recurrent flushing or generalized urticaria (at least twice); (2) urticarial papules greater than 10 mm; (3) no evidence of cardiac, pulmonary or neurological causes; (4) time to onset 15 to 30 minutes after the start of exercise [32]. The maximum interval between ingestion and

onset of symptoms can be up to 3 or 4 hours but is usually less. Prodromes are very common, namely pruritus of extremities, sneezing, cough, respiratory discomfort, flush, and abdominal pain. These manifestations usually regress if the patient stops exercise during the prodrome period. Omega 5 gliadin and molar fractions of high molecular weight glutenin subunits have been incriminated, but also many other foods mostly of vegetable origin [33, 34]. Diagnosis is based on medical history, results of the allergy tests, and if necessary the OFC which must be associated with intense physical exercise. The roles of cofactors – such as alcohol, aspirin and anti-inflammatory drugs – have also been highlighted though the pathophysiology remains poorly understood. Certain cofactors (alcohol, aspirin) could trigger anaphylaxis due to increased intestinal permeability and consequent absorption of partially digested foods including allergens. Unlike other foods, gliadin crosses the intestinal barrier and can be detected in the blood. Dysregulation of the autonomic nervous system has also been suggested. Treatment is based on avoidance of physical exercise within 3 hours after a meal, stopping the activity at the first sign of prodromes, and adrenaline if the manifestations occur.

IV.2 FA and liver transplantation

Liver transplantation in young children is associated with a very high risk of FA [35, 36]. Immunosuppressive therapy (tacrolimus-basiliximab combination) may modify immune tolerance and intestinal permeability in young children with chronic cholestasis who are on a very limited diet before transplantation. Substituting tacrolimus with ciclosporin, if tolerated by the liver, and avoidance of the food allergen are recommended treatment options. The food reintroduction under medical supervision depends on the kinetics of allergy tests after transplantation.

V: Role of allergen components

V.1 Allergen component reactivity and FA characteristics:

Vereda *et al.* used a molecular component diagnostic approach to describe patients with peanut allergy from the USA, Sweden, and Spain. The severity of the peanut allergy differed from one region to another with the Americans being the most severe followed by the Spanish and then the Swedish [37]. The ages at diagnosis were also different with earlier diagnosis in the USA than in Spain or Sweden. Furthermore, they reported that peanut allergy was linked to r Ara h 2 sensitization (90%) in the USA, to r Ara h 9 (60%) in Spain, and to r Ara h 8 (65%) in Sweden. In both the USA and in Western Europe, peanut allergy is known to be associated with a primary sensitization to the 2S albumin allergens r Ara h2, but also r Ara h 1, 3, and 6. In contrast, in Northern and Eastern Europe, peanut allergy has been shown to be frequently secondary to a sensitization to birch PR 10 pan allergen. Finally, in Mediterranean countries such as Spain, peanut allergy is often the expression of a primary lipid transfer protein (LTP) sensitization. Astier *et al.* (France) showed that polysensitized patients (r Ara h 1, 2, 3) were at higher risk of a severe reaction during the OFC than r Ara h2 monosensitized patients [38]. However, this has not been clearly established in other populations.

Diversity associated with geographic factors is also illustrated for nut allergy. The data of the EuroPrevall Cohort showed that the sensitization profiles of patients with hazelnut allergy differed according to the patient's country of residence (PR 10, LTP, oleosins or storage proteins) and age, with possible consequences on the phenotypic expression [39].

Specific molecular allergen sensitizations have been described in children with persistent cow's milk and hen's egg allergies [5, 6]. For example, children with long-lasting egg allergies had higher concentrations of IgE against sequential and native ovomucoid,

ovalbumin and specific ovomucoid epitopes than those who subsequently gained tolerance [40]. Cerecedo *et al.* showed differential recognition patterns for several epitopes between children who had persistent or cow's milk allergy and those who had developed tolerance [41]. This diversity of phenotypes is also shown by the tolerance of baked food compared to raw food, with significant specificities in the immune profile (IgE, IgG4, basophil activating test) [12, 13].

V. 2 Lipid transfer protein (LTP) syndrome

Scala *et al.* used a supervised hierarchical clustering approach to analyze LTP sensitizations in a large population of Italian patients with FA [42]. Eighty-two percent of the 568 patients (94% of those < 6 years of age) were sensitized to Pru p3, and 71% to Jug r3. Participants with more than five LTP sensitizations had the highest risk of systemic reaction. Only Art v3 and Pla a3 (LTP of artemisia and plane tree respectively) were associated with respiratory symptoms. Co-sensitizations to Par j2 (parietaria pollen LTP) and PR-10 or profilins were associated with a lower risk of severe allergic reaction. The authors emphasized that describing different phenotypes by a molecular approach to LTP sensitization may improve FA diagnosis, and subsequently orient dietary advice and FA management according to the risk. This work was conducted in the Mediterranean area and could be adapted for other FAs of plant origin and other geographical areas depending on the local molecular epidemiology.

V.3 Food-pollen allergy syndrome; latex fruit syndrome

Bet v 1-homologous proteins from plant-based foods lead to birch pollen-associated FAs, usually associated with mild to moderate symptoms (oral allergy syndrome) [43]. Severe manifestations remain rare but have been described and attributed to the quantity of food ingested (soy) [44]. Latex fruit syndrome has also been reported involving specific epitopes of latex allergen [45].

V. 4 The alpha-Gal syndrome

Numerous epitopes responsible for IgE-mediated FA have been described and are primarily protein-based. A recent work showed that sIgE antibodies for the carbohydrate, galactose-1,3-galactose (α -Gal), could trigger serious, and even fatal, allergic reactions. The α -Gal carbohydrate moiety is known to be present in multiple tissues from nonprimate mammals. The production of sIgE has been linked to bites from ecto-parasitic ticks, especially those of the Lone Star tick, *Amblyomma americanum*. FA by sensitization to alpha Gal is characterized by delayed-onset anaphylaxis (*i.e.*, 3-6 h) after ingestion of mammalian food products (e.g. beef and pork meat; milk; gelatin) [46]. Observations have been recorded in children [47]. Immediate-onset anaphylaxis during first exposure to intravenous cetuximab is another manifestation of the same sensitization [48].

VI Food allergy biomarkers attached to phenotypes

VI. 1 Biomarkers for the prediction of resolution with time of FA

Cow's milk and hen's egg allergies are known as transient FAs. However, some patients are characterized by persistent phenotypes, with a large part of them who tolerate extensively heated forms of milk and egg. Children who persist with cow's milk allergy have higher sIgE to casein than patients who grow out of it, as illustrated by Garcia Ara et al [49]. They described different cutoffs for sIgE to casein according to the age (13-18 months: 2 kU/L; 24-36 months: 9 kU/L; sensitivity = 95%, sensibility = 90%). Children who do not grow out of their hen's egg allergy have also higher sIgE to egg [5]. More recently, sIgE to casein and to ovomucoid- an egg allergen which is stable against heat and digestion by proteinases- have been assessed to distinguish patients who are tolerant to baked milk or egg. For Caubet et al, the cutoff for sIgE to casein with a 95% positive predictive value is 10 kU/L for a positive OFC to baked milk [149 patients; median age : 6.5 years (first cohort) and 7.5 years(second cohort), 2.1 to 17.3) [12]. For Lemon Mulé et al, the cutoff for sIgE to ovomucoid with more than 90% positive predicted values is 50 kU/L for a positive OFC to baked egg (117 patients; mean age of 6.9 years, range, 1.6–18.6 years) [13]. However, all these decision points have been largely discussed. To try to improve the prediction, IgE and IgG4 epitope diversity and IgE affinity have been assessed in different phenotypes of cow's milk allergy. Wang et al have shown that patients who were tolerant to heated milk (16 patients; mean age of 6.6 years, range, 4.8-11.3 years) had similar IgE binding patterns to those that had outgrown their allergy (8 patients; mean age of 7.9 years, range, 2.6-14.6 years) but IgG4 binding patterns that were more similar to those that have persistent allergy (17 patients; mean age of 7.7 years, range, 3.4-14.9 years) [50]. Furthermore, persistent allergic patients demonstrated a combination of high and low IgE binding, in contrast to those who had outgrown who had low affinity binding. These data are confirmed by Caubet et al

who showed that children with persistent CMA beyond 5 years of age have IgE and IgG4 antibodies that bound to CM peptides with greater intensity and broader diversity at baseline (median age = 10 months) than patients with transient CMA [51]. Regarding patients with egg allergy, ovalbumin and ovomucoid sIgE/sIgG4 ratios are higher in patients who do not tolerate baked egg [52]. Wright et al showed that higher ovomucoid sIgG4/sIgGE ratio were observed in patients who were good responders to OIT, and that it was not observed for egg white or ovalbumin [53]. New assays (polymer coated silicon microarray) with the simultaneous detection of food allergen-specific IgE and IgG4 are in development [54].

To further improve the monitoring for resolution of FA, T cell assays have been developed. Michaud *et al.* evaluated the sensitivity and specificity of ELISpot for the diagnosis of IgE mediated cow's milk allergy [55]. In this study, 29 children aged 3 to 10 years, who presented a diagnosis of cow's milk allergy received a double-blind cow's milk OFC. Peripheral blood mononuclear cells were cultured for 18 h in the presence of casein, α -lactalbumin, and β -lactoglobulin and the number of lymphocytes secreting IL-4 and IL-13 specific for these allergens was evaluated by the ELISpot test. Seventeen children (59%) had a positive OFC, confirming persistent allergy, and the remaining 12 children had a negative test showing that they had developed tolerance. The mean numbers of casein-specific IL-4 and IL-13 secreting T cells were higher in the allergic than in the tolerant children ($p = 0.009$ and 0.004 , respectively). In addition, these numbers were inversely correlated with the cumulative tolerated dose ($p = 0.003$ and 0.0009 , respectively). The prediction of the OFC reaction evaluated by the receiver operating characteristic curve of the combined threshold values of IL-4 and IL-13 was excellent with an area under the curve of 0.98 (95% CI $0.90 - 1.06$). This method could be a promising tool for improving the diagnosis of cow's milk allergy, and deserves to be assessed in larger populations and with other food allergens.

VI.2. Biomarkers for the severity of FA

Although some risk factors related to the severity of FA have been identified, no clear bio-markers are available to predict the risk or the level of ED in real life or during an OFC. Studies have shown contradictory results about sIgE and SPT and individual risk assessment [56]. Shreffler *et al* demonstrated that patients with a history of severe multisystem reaction have similar total recombinant peanut IgE than patients with only cutaneous symptoms but recognized significantly more epitopes of Ara h 1, 2 and 3 [57].

Analysis of basophil activation or lymphocyte function might constitute new tools. The basophil activation test has been used by different teams with conflicting results, probably related to the heterogeneity of the populations as well as to the OFC procedure. Blumchen *et al.* studied 63 children with a median age of 6.5 years (3.2 to 17.8 years) and OFC proven peanut allergy [58]. 86% of the children had a history of reaction in real life (median score of IV on a V-scale) and 54% had asthma. The authors showed that the OFC ED (according to a modified protocol with doses scheduled 2 hours apart) was inversely correlated with sIgE level, prick test size, basophil activation and production of Th2 cytokines by mononucleated blood cells stimulated *in vitro* with a peanut extract. However, none of these markers correlated with the severity of the reaction. Santos *et al.* conducted an OFC to confirm, or not, diagnosis of allergy in a population of children with peanut sensitization with different characteristics (median age 5 years, no history of previous ingestion of peanut before the OFC for 77%) [59]. They also studied the reactivity and the basophil allergen threshold sensitivity (CD sens) and measured the ED by OFC (nine doses including three placebo doses, cumulative dose of peanut protein of 9.35 g). The children were subsequently differentiated according to the severity of the OFC reaction - mild to moderate or severe. The authors showed that the basophil response was correlated with the severity of the reaction and that the CD sens was related to the ED. If these results are confirmed, combining basophil

reactivity and CD sens might be useful to describe a population of peanut allergic children with an “at risk” phenotype characterized by a low threshold and a history of severe reactions, and thereby personalize the management strategy.

VII. Consequences for medical practice and patients approach

Better definition of the phenotypes involved in FAs could help personalize the treatment, especially when it comes to the degree of avoidance, introduction of baked food in the daily diet, and the contents of the emergency kit. Dietary advice is crucial, ideally involving a multidisciplinary team including a dietician, and must be tailored, patient by patient taking into account the identified triggers of reaction. Cross allergies should be assessed to delineate the avoidance perimeter. Baked food might be tolerated by some patients allergic to milk or egg [60] or to PR10 allergen. Guidelines have recently updated the indications of adrenaline auto-injector [61] The emergency kit could be adapted to the FA phenotype as recently proposed by Fernandez Rivas *et al.* for LTP allergic patients in Spain [43]. In the proposed algorithm for patients with allergy to Rosacea fruits in Mediterranean countries, they distinguished patients with LTP and Bet v1/profilin sensitization and pollen allergy who developed mainly mild symptoms (oral allergy syndrome) from the patients with LTP but no Bet v 1/ profiling sensitization and no pollen allergy who are at risk of systemic reaction. Finally, all the allergic comorbidities – asthma, rhinitis, atopic dermatitis- have to be evaluated and adequately treated [24].

Some clinical trials have reported that milk, egg and peanut OIT is effective although mild or moderate side effects can occur [62-65]. To better establish the indications for OIT, biomarkers predicting the efficacy or the risk of side effects or failure are required. Martinez-Botas *et al.* recently designed a study to identify biomarkers associated with the efficacy of

OIT in 25 children who were allergic to milk [66]. They were compared with a control group of seven milk allergic children who were not treated. The treated group was divided into a low-, moderate- or high-risk group based on syndromic reactions during OIT (> 7 reactions) and the time required to complete the OIT (i.e., ingestion without reaction of at least 200 ml of milk). High-risk subjects had IgE directed to a statistically greater number of casein peptides (and, to a lesser degree, for IgG4). Cluster analysis identified 13 regions of casein in terms of IgE response that differed significantly between the high-risk group and the other groups. Bioinformatic analyzes were used to select two sets of 16 IgE-binding casein peptides before ITO that predicted tolerance ($R^2 = 0.858$) and efficacy ($R^2 = 0.732$) of OIT.

Treatment with omalizumab at the initiation of OIT can reduce the number of side effects, especially in severe allergic patients [67-68]. This treatment has also been tried in patients with multiple FAs who remain a challenging population for the clinician [69]. However, a number of important issues such as need to be addressed before applying this expensive strategy: for which patients? Which protocol of administration? Which aim? Up to now, omalizumab is in Europe only approved for severe resistant to treatment allergic asthma or chronic urticaria.

Finally, a better knowledge of the phenotypes could also lead to improved prevention strategies. The LEAP trial showed the positive impact of early introduction of peanut in the daily diet of infants at risk of peanut allergy [70, 71]. Guidelines have been recently updated defining risk categories, appropriate use of allergy tests including OFC and the timing and approaches for introduction of peanut-containing foods [72].

In conclusion, FAs are complex diseases. A phenotypic approach is the key for improving prevention and tailoring diagnosis and treatment. It seems to be important to take into account genetic factors and geographical and environmental features as well as dietary habits. More studies are required to better describe the diversity of FA and to find biomarkers. Finally, such an approach may help identify which patients are at the highest risk of severe, anaphylactic reaction who require follow-up in a tertiary care center.

References

1. Bousquet J, Anto JM, Sterk PJ, Adcock IM, Chung KF, Roca J, Agusti A, et al. Systems medicine and integrated care to combat chronic noncommunicable diseases. *Genome Med.* 2011; 3:43.
2. Just J, Bourgoin-Heck M, Amat F, Just J. Clinical phenotypes in asthma. *Clin Exp Allergy* 2017; in revision.
3. Hagl B, Heinz V, Schlesinger A, Spielberger BD, Sawalle-Belohradsky J, Senn-Rauh M, et al . Key findings to expedite the diagnosis of hyper-IgE syndromes in infants and young children. *Pediatr Allergy Immunol.* 2016; 27:177-84.
4. Torgerson TR, Linane A, Moes N, Anover S, Mateo V, Rieux-Laucat F, Hermine O, et al. Severe food allergy as a variant of IPEX syndrome caused by a deletion in a noncoding region of the FOXP3 gene. *Gastroenterology.* 2007; 132:1705-17.
5. Savage JH, Matsui EC, Skripak JM, Wood RA. The natural history of egg allergy. *J Allergy Clin Immunol.* 2007; 120: 1413-7.

6. Skripak JM, Matsui EC, Mudd K, Wood RA. The natural history of IgE-mediated cow's milk allergy. *J Allergy Clin Immunol.* 2007; 120: 1172-7.
7. Schoemaker AA, Sprickelman AB, Grimshaw KE, Roberts G, Grabenhenrich L, Rosenfeld L, Siegart S, et al. Incidence and natural history of challenge-proven cow's milk allergy in European children--EuroPrevall birth cohort. *Allergy.* 2015; 70:963-72.
8. Saarinen KM, Pelkonen AS, Mäkelä MJ, Savilahti E. Clinical course and prognosis of cow's milk allergy are dependent on milk-specific IgE status. *J Allergy Clin Immunol.* 2005;116: 869-75.
9. Wood RA, Sicherer SH, Vickery BP, Jones SM, Liu AH, Fleischer DM, Henning AK, et al. The natural history of milk allergy in an observational cohort. *J Allergy Clin Immunol.* 2013;131: 805-12.
10. Peters RL, Dharmage SC, Gurrin LC, Koplin JJ, Ponsonby AL, Lowe AJ, Tang ML, et al; HealthNuts study. The natural history and clinical predictors of egg allergy in the first 2 years of life: a prospective, population-based cohort study. *J Allergy Clin Immunol.* 2014; 133:485-91.
11. Sicherer SH, Wood RA, Vickery BP, Jones SM, Liu AH, Fleischer DM, Dawson P, et al. The natural history of egg allergy in an observational cohort. *J Allergy Clin Immunol.* 2014;133:492-9.
12. Caubet JC, Nowak-Węgrzyn A, Moshier E, Godbold J, Wang J, Sampson HA. Utility of casein-specific IgE levels in predicting reactivity to baked milk. *J Allergy Clin Immunol.* 2013; 131: 222-4.

13. Lemon-Mulé H, Sampson HA, Sicherer SH, et al. Immunologic changes in children with egg allergy ingesting extensively heated egg. *J Allergy Clin Immunol* 2008; 122: 977-83.
14. Byrne AM, Malka-Rais J, Burks AW, Fleischer DM. How do we know when peanut and tree nut allergy have resolved, and how do we keep it resolved? *Clin Exp Allergy*. 2010;40:1303-11.
15. Peters RL, Allen KJ, Dharmage SC, Lodge CJ, Koplin JJ, Ponsonby AL, Wake M, et al; HealthNuts study. Differential factors associated with challenge-proven food allergy phenotypes in a population cohort of infants: a latent class analysis. *Clin Exp Allergy*. 2015; 45: 953-63.
16. Havstad S, Johnson CC, Kim H, Levin AM, Zoratti EM, Joseph CL, Ownby DR, et al. Atopic phenotypes identified with latent class analyses at age 2 years. *J Allergy Clin Immunol*. 2014; 134: 722-7.
17. Simpson A, Tan VY, Winn J, Svensén M, Bishop CM, Heckerman DE, Buchan I, Custovic A. Beyond atopy: multiple patterns of sensitization in relation to asthma in a birth cohort study. *Am J Respir Crit Care Med*. 2010; 181: 1200-6.
18. Hose AJ, Depner M, Illi S, Lau S, Keil T, Wahn U, Fuchs O, et al; MAS; PASTURE study groups. Latent class analysis reveals clinically relevant atopy phenotypes in 2 birth cohorts. *J Allergy Clin Immunol*. 2016 Oct 19. pii: S0091-6749(16)31135-6.
19. Gabet S, Just J, Couderc R, Bousquet J, Seta N, Momas I. Early polysensitization is associated with allergic multimorbidity in PARIS birth cohort infants. *Pediatr Allergy Immunol*. 2016; 27: 831-837.

20. Deschildre A, Elégbédé CF, Just J, Bruyère O, Van der Brempt X, Papadopoulos A, Beaudouin E, et al. Peanut allergic patients in the MIRABEL survey: characteristics, allergists' dietary advice and lessons from real life. *Clin Exp Allergy*. 2016; 46: 610-20.14.
21. van der Valk JP, Dubois AE, Gerth van Wijk R, Wichers HJ, de Jong NW. Systematic review on cashew nut allergy. *Allergy*. 2014;69:692-8.
22. Johnson J, Malinovschi A, Alving K, et al. Ten-year review reveals changing trends and severity of allergic reactions to nuts and other foods. *Acta Paediatr* 2014; 103: 862-7.
23. Eigenmann PA, Lack G, Mazon A, Nieto A, Haddad D, Brough HA, Caubet JC. Managing Nut Allergy: A Remaining Clinical Challenge. *J Allergy Clin Immunol Pract*. 2016 Oct 25. pii: S2213-2198(16)30394-4.
24. Summers CW, Pumphrey RS, Woods CN, et al. Factors predicting anaphylaxis to peanuts and tree nuts in patients referred to a specialist center. *J Allergy Clin Immunol* 2008; 121: 632-638.
25. Just J, Elegbede CF, Deschildre A, Bousquet J, Moneret-Vautrin DA, Crepet A;Mirabel study group.. Three peanut-allergic/sensitized phenotypes with gender difference. *Clin Exp Allergy*. 2016;46:1596-1604.
26. Cousin M, Verdun S, Seynave M, Vilain AC, Lansiaux A, Decoster A, Sauvage C. Phenotypical characterization of peanut allergic children with differences in cross-allergy to tree nuts and other legumes. *Pediatr Allergy Immunol*. 2017 Jan 31.
27. Elizur A, Goldberg MR, Levy MB, Nachshon L, Katz Y. Oral immunotherapy in cow's milk allergic patients: course and long-term outcome according to asthma status. *Ann Allergy Asthma Immunol*. 2015; 114: 240-244.

28. Comstock SS, DeMera R, Vega LC, Boren EJ, Deane S, Haapanen LA, Teuber SS. Allergic reactions to peanuts, tree nuts, and seeds aboard commercial airliners. *Ann Allergy Asthma Immunol.* 2008; 101: 51-6.
29. Brown AF, McKinnon D, Chu K. Emergency department anaphylaxis: A review of 142 patients in a single year. *J. Allergy Clin. Immunol.* 2001;108:861–6.
30. Chen W, Mempel M, Schober W, Behrendt H, Ring J. Gender difference, sex hormones, and immediate type hypersensitivity reactions. *Allergy.* 2008;63:1418–27.
31. Mahdavinia M, Fox SR, Smith BM, James C, Palmisano EL, Mohamed A, Zahid Z, et al. Racial Differences in Food Allergy Phenotype and Health Care Utilization among US Children. *J Allergy Clin Immunol Pract.* 2016 Nov 22. pii: S2213-2198(16)30504-9.
32. Sheffer AL, Austen KF. Exercise-induced anaphylaxis. *J Allergy Clin Immunol* 1980; 66: 106-11.
33. Romano A, Scala E, Rumi G, et al. Lipid transfer proteins: the most frequent sensitizer in Italian subjects with food-dependent exercise-induced anaphylaxis. *Clin Exp Allergy* 2012; 42: 1643-53.
34. Brockow K, Kneissl D, Valentini L, et al. Using a gluten oral food challenge protocol to improve diagnosis of wheat-dependent exercise-induced anaphylaxis. *J Allergy Clin Immunol* 2015; 135: 977-84.
35. Needham JM, Nicholas SK, Davis CM. Food allergies developing after solid organ transplant. *Pediatr Transplant.* 2015; 19: 827-35.
36. De Bruyne R, Gevaert P, Van Winckel M, De Ruyck N, Minne A, Bogaert D, et al. Raised immunoglobulin A and circulating T follicular helper cells are linked to the

development of food allergy in paediatric liver transplant patients. Clin Exp Allergy. 2015; 45 : 1060-70.

37. Vereda A, van Hage M, Ahlstedt S, Ibañez MD, Cuesta-Herranz J, van Odijk J, et al. Peanut allergy: Clinical and immunologic differences among patients from 3 different geographic regions. J Allergy Clin Immunol. 2011; 127:603-7.

38. Astier C, Morisset M, Roitel O, et al. Predictive value of skin prick tests using recombinant allergens for diagnosis of peanut allergy. J Allergy Clin Immunol 2006; 118:250-6.

39. Datema MR, Zuidmeer-Jongejan L, Asero R, et al. Hazelnut allergy across Europe dissected molecularly: A EuroPrevall outpatient clinic survey. J Allergy Clin Immunol. 2015; 136: 382-91.

40. Järvinen KM, Beyer K, Vila L, Bardina L, Mishoe M, Sampson HA. Specificity of IgE antibodies to sequential epitopes of hen's egg ovomucoid as a marker for persistence of egg allergy. Allergy. 2007; 62: 758-65.

41. Cerecedo I, Zamora J, Shreffler WG, Lin J, Bardina L, Dieguez MC, Wang J, Muriel A, de la Hoz B, Sampson HA. Mapping of the IgE and IgG4 sequential epitopes of milk allergens with a peptide microarray-based immunoassay. J Allergy Clin Immunol. 2008; 122: 589-94.

42. Scala E, Till SJ, Asero R, Abeni D, Guerra EC, Pirrotta L, et al. Lipid transfer protein sensitization: reactivity profiles and clinical risk assessment in an Italian cohort. Allergy. 2015; 70: 933-43.

43. Fernández-Rivas M. Fruit and vegetable allergy. Chem Immunol Allergy 2015; 101: 162-

44. Kleine-Tebbe J, Vogel L, Crowell DN et al. Severe oral allergy syndrome and anaphylactic reactions caused by aBet v 1- related PR-10 protein in soy-bean, SAM22. *J Allergy Clin Immunol* 2002; 110: 797–804.
45. Barre A, Culerrier R, Granier C, Selman L, Peumans WJ, Van Damme EJ, Bienvenu F, et al. Mapping of IgE-binding epitopes on the major latex allergen Hev b 2 and the cross-reacting 1,3beta-glucanase fruit allergens as a molecular basis for the latex-fruit syndrome. *Mol Immunol*. 2009; 46: 1595-604.
46. Wilson JM, Schuyler AJ, Schroeder N, Platts-Mills TA. Galactose- α -1,3-Galactose: Atypical Food Allergen or Model IgE Hypersensitivity? *Curr Allergy Asthma Rep*. 2017;17:8..
47. Kennedy JL, Stallings AP, Platts-Mills TA, Oliveira WM, Workman L, James HR, Tripathi A, et al. Galactose- α -1,3-galactose and delayed anaphylaxis, angioedema, and urticaria in children. *Pediatrics*. 2013;131:e1545-52.
48. Berg EA, Platts-Mills TA, Commins SP. Drug allergens and food--the cetuximab and galactose- α -1,3-galactose story. *Ann Allergy Asthma Immunol*. 2014;112:97-101.
49. García-Ara MC, Boyano-Martínez MT, Díaz-Pena JM, Martín-Muñoz MF, Martín-Esteban M. Cow's milk-specific immunoglobulin E levels as predictors of clinical reactivity in the follow-up of the cow's milk allergy infants. *Clin Exp Allergy*. 2004;34:866-70.
50. Wang J, Lin J, Bardina L, Goldis M, Nowak-Wegrzyn A, Shreffler WG, Sampson HA. Correlation of IgE/IgG4 milk epitopes and affinity of milk-specific IgE antibodies with different phenotypes of clinical milk allergy. *J Allergy Clin Immunol*. 2010;125:695-702

51. Caubet JC, Lin J, Ahrens B, Gimenez G, Bardina L, Niggemann B, Sampson HA, Beyer K. Natural tolerance development in cow's milk allergic children: IgE and IgG4 epitope binding. *Allergy*. 2017 Mar 27.
52. Caubet JC, Bencharitiwong R, Moshier E, Godbold JH, Sampson HA, Nowak-Węgrzyn A. Significance of ovomucoid- and ovalbumin-specific IgE/IgG(4) ratios in egg allergy. *J Allergy Clin Immunol*. 2012;129: 739-47.
53. Wright BL, Kulis M, Orgel KA, Burks AW, Dawson P, Henning AK, Jones SM, et al; Consortium of Food Allergy Research. Component-resolved analysis of IgA, IgE, and IgG4 during egg OIT identifies markers associated with sustained unresponsiveness. *Allergy*. 2016; 71:1552-1560.
54. Sievers S, Cretich M, Gagni P, Ahrens B, Grishina G, Sampson HA, Niggemann B, et al. Performance of a polymer coated silicon microarray for simultaneous detection of food allergen-specific IgE and IgG(4). *Clin Exp Allergy*. 2017 Mar 27.
55. Michaud B, Aroulandom J, Baiz N, Amat F, Gouvis-Echraghi R, Candon S, et al. Casein-specific IL-4- and IL-13-secreting T cells: a tool to implement diagnosis of cow's milk allergy. *Allergy*. 2014; 69: 1473-80.
56. Santos AF, Brough HA. Making the Most of In Vitro Tests to Diagnose Food Allergy. *J Allergy Clin Immunol Pract*. 2017;5:237-248.
57. Shreffler WG, Beyer K, Chu TH, Burks AW, Sampson HA. Microarray immunoassay: association of clinical history, in vitro IgE function, and heterogeneity of allergenic peanut epitopes. *J Allergy Clin Immunol*. 2004;113:776-82.58. Blumchen K, Beder A, Beschorner J, et al. Modified oral food challenge used with sensitization biomarkers provides more real-life clinical thresholds for peanut allergy. *J Allergy Clin Immunol*. 2014; 134: 390-8.

59. Santos AF, Du Toit G, Douiri A, et al. Distinct parameters of the basophil activation test reflect the severity and threshold of allergic reactions to peanut. *J Allergy Clin Immunol* 2015; 135: 179-86.
60. Leonard SA, Caubet JC, Kim JS, Groetch M, Nowak-Węgrzyn A. Baked milk- and egg-containing diet in the management of milk and egg allergy. *J Allergy Clin Immunol Pract*. 2015; 3: 13-23.
61. Muraro A, Roberts G, Worm M, Bilò MB, Brockow K, Fernández Rivas M, et al; EAACI Food Allergy and Anaphylaxis Guidelines Group. Anaphylaxis: guidelines from the European Academy of Allergy and Clinical Immunology. *Allergy*. 2014; 69:1026-45.
62. Anagnostou K, Islam S, King Y, Foley L, Pasea L, Bond S, et al. Assessing the efficacy of oral immunotherapy for the desensitisation of peanut allergy in children (STOP II): a phase 2 randomised controlled trial. *Lancet* 2014; 383: 1297-304.
63. Peters RL, Dang TD, Allen KJ. Specific oral tolerance induction in childhood. *Pediatr Allergy Immunol*. 2016; 27: 784-794.
64. Vickery BP, Scurlock AM, Kulis M, Steele PH, Kamilaris J, Berglund JP, et al. Sustained unresponsiveness to peanut in subjects who have completed peanut oral immunotherapy. *J Allergy Clin Immunol*. 2014; 133: 468-75.
65. Nurmatov U, Dhami S, Arasi S, Pajno GB, Fernandez-Rivas M, Muraro A, Roberts G et al. Allergen immunotherapy for IgE-mediated food allergy: a systematic review and meta-analysis. *Allergy*. 2017 Jan 6.
66. Martínez-Botas J, Rodríguez-Álvarez M, Cerecedo I, Vlaicu C, Diéguez MC, Gómez-Coronado D, et al. Identification of novel peptide biomarkers to predict safety and efficacy of cow's milk oral immunotherapy by peptide microarray. *Clin Exp Allergy*. 2015; 45: 1071-84.

67. MacGinnitie AJ, Rachid R, Gragg H, Little SV, Lakin P, Cianferoni A, et al. Omalizumab facilitates rapid oral desensitization for peanut allergy. *J Allergy Clin Immunol*. 2016 Sep 5. pii: S0091-6749(16)30898-3.
68. Brandström J, Vetander M, Lilja G, Johansson SG, Sundqvist AC, Kalm F, Nilsson C, Nopp A. Individually dosed omalizumab: an effective treatment for severe peanut allergy. *Clin Exp Allergy*. 2016 Nov 24.
69. Bégin P, Winterroth LC, Dominguez T, Wilson SP, Bacal L, Mehrotra A, et al. Safety and feasibility of oral immunotherapy to multiple allergens for food allergy. *Allergy Asthma Clin Immunol*. 2014; 10: 1.
70. Du Toit G, Roberts G, Sayre PH, Bahnson HT, Radulovic S, Santos AF, et al; LEAP Study Team. Randomized trial of peanut consumption in infants at risk for peanut allergy. *N Engl J Med*. 2015; 372: 803-13.
71. Du Toit G, Sayre PH, Roberts G, Sever ML, Lawson K, et al; Immune Tolerance Network LEAP-On Study Team. Effect of Avoidance on Peanut Allergy after Early Peanut Consumption. *N Engl J Med*. 2016;374:1435-43.
72. Togias A, Cooper SF, Acebal ML, Assa'ad A, Baker JR Jr, Beck LA, Block J, et al. Addendum guidelines for the prevention of peanut allergy in the United States: Report of the National Institute of Allergy and Infectious Diseases-sponsored expert panel. *J Allergy Clin Immunol* 2017; 139: 29-44.

Figure 1: Major determinants of FA phenotypes

FA= food allergy, PFS = pollen food syndrome, NSAID= nonsteroidal anti-inflammatory drug

Table I: cow's milk allergy phenotypes: data of the cohorts

Study	Population	Aim	Main results	Conclusions
Baked/raw milk				
Caubet et al (12). (1 tertiary care center, USA)	225 patients 2.1 to 15 years of age baked milk reactive (N=61) baked milk tolerant (N= 149) Outgrown milk allergy (N=15)	To evaluate the clinical utility and added diagnostic value of measurements of sIgE and sIgG4 antibodies to CM, casein, and β lactoglobulin	- Levels of sIgE to CM, casein, and β lactoglobulin significantly higher in baked milk–reactive patients - Casein- and β lactoglobulin–sIgE/sIgG4 ratios significantly higher in baked milk–reactive subjects - Casein-sIgE : significantly greater accuracy for predicting baked milk reactivity	Quantitative measurements of casein-sIgE antibodies useful in the management of CM allergy and the tolerance of baked milk. Cutoff = > 20.2 kUA/L (unlikely negative OFC with baked milk): NPV=78%, specificity = 95%. Cutoff = < 0.94 kUA/L (unlikely positive OFC with baked milk): NPV=96%, specificity = 32%,
Raw milk				
Skripak et al (6). (2 pediatric allergy clinics, 1 private and 1 university-based, USA)	Retrospective cohort, 1993-2007, 807 patients Asthma=49% Allergic rhinitis=40% Eczema=71% Other FA =91%	To define the rate of allergy resolution over time To identify the clinical and laboratory features that may predict the outcome of CMA over time	Tolerance (OFC passed, OR no reaction in the past 12 months and sIgE < 3 kU/L) : at 4 years: 19%, at 8 years: 42%, at 12 years: 64%, at 16 years: 79% Predictors of persistence : co-existing asthma or allergic rhinitis, high peak sIgE level ($p<0.001$)	Prognosis for CMA is worse than previously reported. Follow-up and re-evaluation of CMA patients is important because late recovery may be observed Predictors of persistence : Peak sIgE level (>20 kU/L) and allergic comorbidities
Schoemaker et al (7) (EuroPrevall birth cohort in 9 European countries : Iceland, UK, Germany, Poland, Lithuania, Spain, Italy, Greece, the Netherlands)	9336 patients followed up from birth to 2 years of age	To define the overall and centre-specific incidence and natural history of CMA in children up to 2 years of age	- CMA suspected=358 - IgE and non IgE mediated CMA confirmed by a double-blind placebo controlled OFC =55 (median age=6 months) - Incidence=0.54% (95% CI 0.41-0.70) - 32/55 : evaluation 1 year after diagnosis Tolerance = 69% (100% of non IgE associated CMA, 57% of IgE associated CMA)	Challenge-proven CMA in <1% of children up to age 2. Good prognosis of CMA: two-thirds of affected infants becoming tolerant within one year after diagnosis. Better prognosis for non-IgE mediated CMA

Saarinen and al (8) (Finland)	Population-based cohort of 6209 full-term newborn infants Emergence of CMA prospectively studied and confirmed in 118 using an elimination challenge test Children followed until recovery	To investigate the development of tolerance and the risk for asthma, rhinoconjunctivitis, atopic dermatitis, and sensitization in children with CMA followed to school age	- IgE-mediated CMA in 86 (73%) - Persistent CMA at age 8.6 years: 15% who developed more allergic diseases and sensitization than control subjects - All children with IgE-negative CMA tolerant by age 5.0 years ($P < .0001$) - Risk factors for persistent CMA at age 2.0 years: sensitization to CM at age 1.6 years, urticaria at diagnostic challenge, CM exposure at the maternity hospital, early sensitization to egg - CMA: risk factor for sensitization to inhalant allergens	IgE-mediated CMA often persists to school age CMA is a risk factor for other allergic diseases
Wood et al (9) (5 sites ; USA)	Observational study 512 infants originally enrolled aged 3 to 15 months Subset of 244 infants with positive OFC or convincing history of milk allergy, with a positive cow's milk sIgE and/or SPT and/or (2) flare of AD associated with milk ingestion and sIgE>5 kU/L	To assess the natural history of milk allergy To determine the clinical factors predicting the resolution of milk allergy over the first 5 years of life	Resolution of milk allergy in 154 (52.6%) subjects at a median age of 63 months Characteristics most predictive of resolution: milk-specific IgE level (5.74 fold increase of resolving if < 2 kU/L: versus > 10 kU/L), milk SPT wheal size (3.65 fold increase of resolving if SPT < 5 versus > 10 mm) , and AD severity Baseline milk-specific IgG4 level and milk IgE/IgG4 ratio not predictive of resolution	Approximately one half resolved over 66 months of follow-up. Baseline milk-specific IgE level, SPT wheal size, and AD severity were all important predictors of the likelihood of resolution.

ED: eliciting dose; OFC: oral food challenge; AD: atopic dermatitis ; CMA: cow's milk allergy; CM: cow's milk; SPT: skin prick test; Ig : immunoglobulin

Table II: hen's egg allergy phenotypes: data of the cohorts

Study	Population	Aim	Main results	Phenotype consequences
Baked egg				
Lemon-Mulé et al (13). (1 tertiary care center, USA)	117 patients (mean age=6,9 years) with documented IgE-mediated egg allergy: OFC to extensively heated egg/regular egg if passed heated egg challenge AND heated egg incorporated in diet in heated egg-tolerant subjects	To confirm tolerance of heated egg in a subset of children with egg allergy, To evaluate clinical and immunologic predictors of heated egg tolerance, To characterize immunologic changes associated with continued ingestion of heated egg To determine whether a diet incorporating heated egg is well tolerated.	Heated egg tolerated in 64 patients, 23 tolerated regular egg, and 27 reacted to heated egg. Heated egg– reactive subjects: larger skin test wheals, greater egg white sIgE, ovalbumin sIgE, and ovomucoid-sIgE levels Continued ingestion of heated egg associated with decreased egg white skin test wheal diameters and ovalbumin-sIgE levels and increased ovalbumin-specific and ovomucoid-sIgG4 levels.	Extensively heated egg baked is tolerated in the majority (70%) of children with egg allergy, confirming that heating significantly diminishes the allergenicity of hen's egg white. Incorporating extensively heated egg products into the daily diet is well tolerated; Regular ingestion of heated egg is associated with modification of immunologic parameters.
Peters et al (10) HealthNuts cohort (Australia)	Population-based birth cohort 5276 infants (11-15 months) Baked egg substudy : 264 infants with challenge-confirmed egg allergy at 12 months; 140 infants with additional baked egg OFC	To assess the natural history of egg allergy at 2 years of age To identify clinical predictors for persistent egg allergy	- Egg allergy resolved in 66 (47%) infants by 2 years of age (95% CI, 37 to 56%) - Baked egg reactive versus baked egg tolerant at 1 year of age : resolution = 13% versus 56% Tolerance increased if frequent ingestion of baked egg (>5 times per month) Mutation in the filaggrin gene not associated with the resolution of either egg allergy or egg sensitization at age 2 years.	Phenotyping of egg allergy (baked egg tolerant / reactive) is essential for the daily management (diet restriction) and also in terms of prognosis, complications.
Raw egg				
Savage et al. (5) (1 tertiary care center, USA)	retrospective chart review 881 patients (median age=14 months) Diagnosis: history of allergic reaction to egg, and evidence of egg sensitization (egg IgE >0.35 kU/L or positive SPT)	To estimate the proportion of children with egg allergy who develop egg tolerance To identify predictors of tolerance development.	- Resolution in 4% of patients with egg allergy by age 4 years, 12% by age 6 years, 37% by age 10 years, 68% by age 16 years. - Persistent egg allergy: higher sIgE levels at all ages to age 18 years, allergic comorbidities and MFA	A majority of patients with egg allergy will develop egg tolerance. Most patients are likely to develop egg tolerance by late childhood, except patients with peak sIgE > 50 kU/L

Sicherer and al.(11) (5 tertiary care centers, USA)	Multicenter observational study 213 Children Diagnosis: convincing history of immediate allergic reaction and/or moderate-severe atopic dermatitis + positive SPT Resolution: successful ingestion.	To describe the natural course of egg allergy and to identify early prognostic markers.	Egg allergy resolved in 105 (49.3%), at a median age of 72 months Factors most predictive of resolution: initial reaction characteristics (isolated urticaria/angioedema), baseline egg-sIgE level, egg SPT wheal size, atopic dermatitis severity, IgG4 and IL-4 response	Approximately 50% of children with egg allergy egg tolerant by 6 years of age. Resolution highly associated with lower peak egg-sIgE level, acute reaction with only skin symptoms
---	--	---	---	---

OFC: oral food challenge; MFA: multiple food allergy ; SPT: skin prick test; sIgE : specific IgE

Table III: Impact of early food sensitization on the phenotype of the allergic disease

Study	Population	Aim	Main results	Conclusions
Havstad et al (16) WHEAL birth cohort (USA)	594 infants evaluation at 2 years (Afro-american=62%)	To determine atopic diseases phenotypes by a LCA, using sensitizations to 10 Ag including egg, cow's milk, and peanut measured at 2 years of age (No food challenge). To compare conventional (at least 1 sIgE level ≥ 0.35 IU/mL) to LCA classification	4 classes : - Low or no sensitization - High sensitization - Milk and egg dominant - Peanut and inhalant dominant	- Food sensitization per se : not a risk factor of asthma at 4 years of age, contrary to high sensitization (OR = 5.3; 95% CI: 1.6-17.5) - LCA classification more strongly associated to allergic diseases risk than conventional classification
Gabet et al (19) PARIS birth cohort (France)	1525 infants - Sensitization evaluation at 18 months - Allergic diseases evaluation at 6 years	To study by a cluster analysis the impact of early sensitizations on the risk of atopic diseases at 6 years 12 food sIgE : milk, egg white, peanut, mustard, fish, wheat, soy, kiwi, beef, hazelnut, sesame, shrimp	Sensitization to: - ≥ 1 food Ag = 13.8% (cow's milk: 7.6%, egg: 6.6%, peanut: 1.9%, hazelnut: 2.3%) - ≥ 1 inhalant Ag = 2.3% - Several foods or food + inhalant Ag = 6.2%.	3 profiles in infants: - No sensitizations = 89% - 1 or few sensitizations = 9,2% - polysensitizations = 1.6% Prevalence of allergic disease at 6 years increases from the class 1 to 3.
Hose et al (18) 2 birth cohorts MAS (Germany) and PASTURE cohort (5 european countries)	Analysis on 766 PASTURE cohort children and 680 MAS cohort children First 6 years	To study by LCA the sensitization profiles To relate it to asthma and allergic diseases - PASTURE: 6 foods (milk, white egg, soy, wheat, peanut, hazelnut sIgE) and 13 inhalant Ag, at 1, 2, 3, 5, 6, 7 years of age - MAS: 4 foods (milk, white egg, soy, wheat sIgE) and 6 inhalant-Ag, at 16, 54 and 72 months of age	MAS : one class sensitized to cow's milk or egg PASTURE: two classes : one sensitized to milk, the other to milk+other food Others classes = inhalant or severe atopy defined by multiple sensitizations including seasonal Ag	- Food sensitization : not associated to symptomatic allergic diseases - Inhalant sensitization : associated to benign allergic disease with normal lung function - Multiple sensitization : severe allergic disease and impaired lung function
Peters et al (15) Population based birth cohort : HealthNuts cohort (Australia)	5276 infants 11-15 months	To study by LCA expression of allergic diseases in the first 2 years Challenge proven FA	- Egg : S : 14% and A : 9.4% - Peanut : S : 8% and A : 3% - Sesame: S : 2% and A : 0.7% - 5 distinct phenotypes : no atopic disease; eczema without FA; egg allergy only; multiple peanut dominant; multiple egg dominant	Significant determinants : - Multiple FA: gender (male), prematurity - Egg allergy: parents born in Asia, filaggrin null, history of atopy in the siblings

LCA: latent class analysis ; FA : food allergy; sIgE : specific IgE; Ag= allergen; S= sensitization; A= allergy. OR= odds ratio

Table IV: peanut allergy phenotypes: data of the cohorts

Study	Population	Aim	Main results	Conclusions
Peanut				
Summers et al (24) (Tertiary Regional Allergy Center, Manchester , UK) - 1992 to 2004	- 1094 patients, with history of peanut and TN allergy - 76% : < 15 years - most severe allergic reaction: to peanut:80%, to TN: 20% (cashew nut:7%, brazil nut: 7%, walnut: 3%, hazelnut: 2%, almond: 1%, pine nut: 1%). - Severe reactions history: pharyngeal oedema=43% Acute A =35% reduced consciousness: 14%	to determine which clinical and laboratory parameters best predict the likelihood of severe allergic reactions.	- Allergic comorbidities increase the risk of severe reactions: Strongest predictor of severe upper airway obstruction: severe allergic rhinitis [OR= 3.8, 2.1-6.9], Strongest predictor of severe lower airway disease: A (OR: 6.8 , 4.1-11.3]), Strongest predictor of reaction affecting the systemic circulation: severe AD [OR=3.1; 1.1-8.4]. - Severe upper airway obstruction : 2.6 (1.8-3.7) fold more common after ingestion of TN than peanuts - No association between sIgE, skin prick wheal size and either severity of chronic atopic diseases or acute allergic reactions - Patients with serum angiotensin-converting enzyme concentrations <37.0: mmol/L : 9.6 (1.6-57) fold risk of severe pharyngeal edema.	Role of allergic comorbidities in the severity of allergic reaction to peanut or TN confirmed Specificities according to the features of the comorbidity. These comorbidities have to be controlled in patients with peanut/ TN allergy. Acute A is a feature of anaphylactic reaction and should be treated with adrenaline Suggested role of bradykinin metabolism in severe life threatening pharyngeal oedema; to be confirmed.
Just et al (25) MIRABEL study (France, Belgium, Luxemburg)	704 patients (<16 y: 88%) History of peanut allergy= 82%, Anaphylaxis=30% Oral food challenge in 247 patients ED available but no data on the reaction	To identify peanut-allergic/sensitized phenotypes Combined factor and cluster analysis (N=247)	3 clusters described: Cluster 1 = Severe peanut allergy with little allergic multi-morbidity (50%): medium level of ED at OFC (235 mg PP), low rates of allergic comorbidities Cluster 2 = Severe peanut allergy with allergic comorbidities (25%): lowest ED at OFC (112 mg PP), 100% with A + AD, 84% with A + AD + MFA. Cluster 3 = Mild peanut allergic / sensitized phenotype (25%): lowest mean age, lowest proportion of positive OFC (53%), highest ED (770 mg PP), medium	1. Confirmation of the diversity of peanut allergy in a large population based cohort 2. Differences between girls and boys: only severe phenotype in girls 3. Consequences on the management according to the allergic risk : 1/ for the OFC challenge 2/ for the daily life: diet, emergency kit, ...

			proportion of comorbidities (48% A + AD + MFA).	
Cousin et al (26) (France)	317 children, 7 years (SD=3.9) 12 years period 1 allergy care center Peanut allergy always confirmed by OFC (ED and symptoms score available) No data on the severity of the reaction	To identify phenotypes of peanut allergy To describe cross-allergy patterns and the possible risk factors for cross-allergy to TN and legumes. Hierarchical agglomerative clustering method	Cross-allergy to TN and/or other legumes = 43.2%, risk factor: AD (adjusted OR=16 [95% CI: 7.4–37]; p < 0.001) Main TN= Hazelnut = 21.8% Main legume = Pea = 3.2% 3 phenotypes described: Cluster 1 (n=72): highest level of rAra h 2, lowest ED at OFC (median= 41.5 mg PP), highest asthma rate : 80.6%, cross allergy to TN and/or legumes: 31.9%. Cluster 2 (n=93): highest ED at OFC (median=191.5 mg PP), lowest % of cross-allergy to TN and/or legumes (1.1%). Cluster 3 (n=152): highest % of cross-allergy to TN and/or legumes : 74.3% and AD (94.7%), median ED at OFC= 91.5 mg PP.	1. Confirmation of the diversity of peanut allergy, of the high frequency of TN cross allergy, and of the low frequency of legumes cross allergy. 2. Highest severity in girls (cluster 1 = 56%) 3. Consequences on the management: Asthma associated with the severity AD = risk factor of TN/legumes cross allergy Low risk of TN/legumes cross allergy if high ED at OFC

A = asthma, AD = atopic dermatitis, MFA = multiple food allergies, Ed = eliciting dose, OFC = oral food challenge; TN = tree nuts, OR = odds ratio

