Abstracts

Friday 15 September 2017
Poster discussion Session I
14:00 - 14:45
Poster Area

P01 Epicutaneous vaccine-adjuvanted micro-delivery for allergen-specific immunotherapy to food allergy

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Keywords: Epicutaneous Immunotherapy (EPIT), Food Allergy, Immunoglobulin E (IgE), Macrophages (MF), Microneedle (MN)

Introduction
Sublingual/oral immunotherapy is still the mainstay therapeutic approach for reversing peanut allergy in clinics. However, sublingual/oral immunotherapy can cause severe life-threatening allergic reactions in some patients and, therefore, painless and safer alternative routes of treatment strategies are in great demand. Here in this work,We sought to fabricate short-microneedles (MN) loaded with powdered vaccine-adjuvants (VA) for epicuatneous immunotherapy (μEPIT) in a peanut-sensitized murine model of intestinal hypersensitivity.
Methods
Newly fabricated short-microneedles (MN) patches loaded with powdered vaccine-adjuvants [peanut (PN), vitamin-D3 (VD3) and CpG-ODN] were prepared and applied epicutaneously onto the intact shaved skin of peanut allergy mice. MN-VA patch or intradermal (ID)-VA application was repeated six times at two treatment phases and its clinical severity, humoral as well as local immune responses were determined. Finally, induction of antigen-specific regulatory-T cells (t-regs) as well as IL-10+ and TGF-β1+ skin-resident macrophages (MΦ) was determined.
Results
We designed and fabricated short microneedles loaded with powdered vaccine-adjuvants. Further, peanut allergy mice treated with both MN-VA and ID-VA but not vehicle or ID-PN resulted in significant reduction in clinical severity, humoral PN-specific IgE and mucosa specific mast cells as well as eosinophils. Concomitantly, IgG2a levels in serum as well as T-regs response in spleen, mesenteric- and draining-lymphnodes were subsequently elevated. Further, epicutaneous immunization with adjuvants (VD3 and CpG) in combination but not alone showed profound increase in the migratory and immuoregultory MΦs in skin, which expresses anti-inflammatory IL-10 and TGF-β1 intracellularly.

Conclusion
Epicutaneous immunotherapy using short-microneedles loaded with powdered vaccine-adjuvants might serve as non-invasive and safe technology for IgE-mediated allergic diseases. Vaccine-adjuvanatated immunotherapy promotes immune tolerance initiated by skin-enriched CD11b+ macrophages, thereby favouring expansion of antigen-specific regulatory-T cells. Thus, μEPIT with vaccine-adjuvants combination could be a new and effective therapeutic approach by shortening the number of times of immunotherapy along with minimal risk of anaphylaxis.

P02 Tolerance to baked egg in children under 5 years old

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Introduction
The introduction of baked foods in allergic children's diet seems to allow a natural approach to an oral immunomodulation; not only improving their quality of life by decreasing the fear of possible adverse events, but also increasing food variety. Aim, to evaluate tolerance to baked egg (BE) in egg allergic children under 5 years of age
Methods
Prospective, descriptive study. Data were collected for demographics, adverse events at OFC with baked and cooked egg, specific-IgE (s-IgE) and skin prick test (SPT). Written informed consent was signed. Open OFC with BE were performed, each biscuit contained 0.22 gr of egg protein. If negative, patients underwent a cooked egg OFC. Immunological parameters were compared between groups of tolerant and non-tolerant patients using the non-parametric U Mann-Whitney test. ROC curves were performed.
Results
32 patients were included, 65% boys. Median age at OFC: 45 months (IQ range 31.2-57.7). Median SPT: egg-white (EW) 8.64 mm (6.05-11.86), yolk 6.67 mm (4.73-9.0), ovalbumin (OVA) 7.61 mm (4.87-10.34), ovomucoid (OVM) 8.84 mm (6.5-12.6). Median s-IgE: EW 3.45 KU/L (1.26-13.17), yolk 0.66 KU/L (0.26-2.03), OVA 1.72 KU/L (0.55-7.17), OVM 3.18 KU/L (1.4-12.5). 50% (16/32) were tolerant to BE (1.1 gr of protein by protocol, 5 biscuits). They underwent an OFC with cooked egg: only 1 patient tolerated a complete cooked egg omelet, 3 have not yet undergone the OFC and 11 were allergic to cooked egg. Significant differences between tolerant vs non-tolerant to BE, are seen in yolk and OVA SPT (p 0.023 and p 0.013) and s-IgE (p 0.015 and p 0.038). Area under the curve was: 0.73 (CI 95% 0.55-0.91) for Yolk SPT; 0.75 (CI 95% 0.58-0.93) for OVA SPT; 0.74 (CI 95% 0.55-0.92) for Yolk s-IgE; 0.74 (CI 95% 0.56-0.93) for OVA s-IgE; 0.72 (CI 95% 0.52-0.92) for OVM s-IgE. Cut-off values were determinate to predict allergy to BE: Yolk SPT ≥ 6.03 mm, 75% sensitivity (S) 68% specificity (E); OVA SPT ≥ 6.05 mm, 75% S 56% E; Yolk s-IgE ≥ 0.53 KU/L, 80% S 73% E; OVA s-IgE ≥ 1.5 KU/L, 80% S 73% E; EW s-IgE ≥ 1.1 KU/L, 86% S 26% E; OVM s-IgE ≥ 3.5 KU/L, 73% S 80% E.

Conclusion
Half of our egg-allergic patients under 5 years of age were tolerant to 1.1 gr of BE protein, allowing the inclusion of 5 biscuits in their diet and foods that can contain traces, improving quality of life. Yolk and OVA SPT and yolk, OVA and OVM s-IgE are good markers for prediction of patients allergic to BE, with a high sensitivity and specificity.

P03 What can oral food challenge teach us about tolerance in patients with cow's milk protein allergy?

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Keywords: Oral Food Challenge, Cow´s Milk Protein Allergy, Tolerance, Specific IgE, Food Allergy

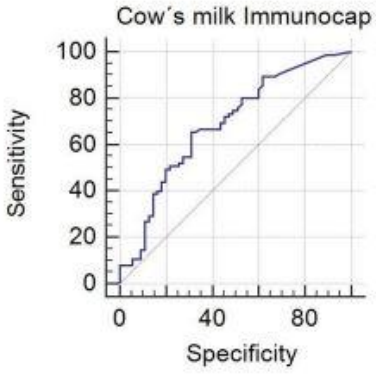
Introduction
Cow´s milk protein allergy (CMPA) is a commom cause of food allergy in children. IgE mediated CMPA occur often whithin few weeks after introdution of cow´s milk. Early reactions can occur after minimal ingest. The detection of specific IgE against milk may guide the diagnosis and prognosis of allergic patients and the oral food challenge (OFC) remains the gold standart for diagnosis and assessment the tolerance of CMPA. The objective of this study is to compare clinical and laboratorial profiles of patients who underwent OFC to assess tolerance to fresh cow's milk
Methods
Retrospective evaluation of OFC with fresh cow's milk performed to assess tolerance in patients with IgE mediated CMPA from September 2002 to March 2016 at a tertiary care pediatric allergy clinic. All procedures were performed in a hospital setting. OFC were considered positive if signs and symptoms described in medical history were reproduced. Patients who ingested 200 ml of milk without reaction were considered tolerant.
Results
188 OFC for cow's milk were performed to assess tolerance in patients with a confirmed history of IgE mediated CMPA. We excluded 54 patients because data on serum specific IgE to cow's milk was not available for the previous year. Of the 134 OFC, involving 94 patients (63M: 31F), 75 had a positive result, 55 were negative and 4 were inconclusive. The median of age those who had a positive test was 5.3 years and 4.79 for negative (p=0.82). The median of age at onset of symptoms was 0.33 years in patients with remaining allergy and 0.41 years in tolerant ones (p=0.14). Regarding family and personal history of atopy, values were similar between the two groups (75% and 70%, respectively). Prior anaphylaxis was present in 66% of patients with persistent CMA, and 58% of tolerant ones (p= 0.4). Comparing levels of specific IgE to milk and milk fractions performed on the year of OFC there were significant statistical differences to higher levels of cow´s milk and casein in patients with persistence of allergy (p=0.0003 / p=0.004). Similar results were found at skin prick test (p=0.0001 / p=0.001).

Conclusion

Clinical features could not establish differences between tolerant and allergic patients but lower levels of cow´s milk and casein specific IgE could indicate the best moment to submit patients to oral food challenge to determine cow´s milk tolerance

Recurrent oral food challenge to assess tolerance: relevant data

Characteristics	Values	
Number of OFC	64	
Number of enrolled patients	28	
Status of allergy	17 tolerants	9 persistents
Mean/median of age of first OFC	3,3 y	3,7 y
Mean/median of age to acquire tolerance	5,1 y	4,2 y
Number of OFC needed to acquire tolerance	2,35	
Mean/median of age of last OFC	8,5 y	8,7 y
Inconclusive OFC	2	



Sample size	130
Positive group [a]	75 (57,69%)
Negative group [b]	55 (42,31%)

Area under the ROC curve (AUC)

Area under the ROC curve (AUC)	0,687
Standard Error [a]	0,0478
95% Confidence interval [b]	0,600 to 0,766
z statistic	3,921
Significance level P (Area=0.5)	0,0001

Youden index

Youden index J	0,3442
Associated criterion	>1,84
Sensitivity	65,33
Specificity	69,09

P04 Pharmacy availability of epinephrine auto-injectors and knowledge of pharmacists regarding their use

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Keywords: Epinephrine Auto-Injectors, Anaphylaxis, Pharmacists

Introduction
Epinephrine is the treatment of choice for anaphylaxis. Patients that have experienced a life-threatening anaphylactic episode have to be trained on how to use epinephrine auto-injectors (EAI) and always carry it with them. Pharmacists may play a crucial role, not only supplying EAI but also to urgently assist patients in the absence of a medical assistance. The aim of our study was to examine the prompt access of patients in buying EAI devices from pharmacies and pharmacists' knowledge on its use.
Methods
The pharmacies of Nicosia were randomized and a team of medical students visited part of them. Pharmacists were asked on whether they had available ANAPEN (the only EAI device in Cyprus) or adrenaline ampules for medical use. Three questions were made on EAI's use; a) the site of application, b) which ANAPEN is recommended for a child of 27kg and c) contraindications for the use of epinephrine in the case of anaphylaxis.
Results
49 questionnaires have been completed, while 8 more pharmacies were visited refusing to participate in the study. Only 1/49 had ANAPEN (the 300μg device) and 3/49 had adrenaline for medical use. 12/49 replied accurately indicating thigh as the injection site, while also 9/49 replied "intramuscularly". Regarding the question on dose 3/49 replied 300μg correctly. 18/49 replied correctly that no contraindication on epinephrine's use exist, in the case of anaphylaxis. Impressive was the unwillingness of pharmacists to reply in most of the questions ranging 15-19/49.
Conclusion
In Nicosia patients shouldn't rely on purchasing EAI from a pharmacy in the event of anaphylaxis and should order and buy it soon after their doctor prescribes it. Information and training of pharmacists on EAI is inadequate.

P05 Cow's milk protein intolerance imitating septic shock in a small infant

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Keywords: Allergy, Enterocolitis, Cow'S Milk

Introduction
Food protein-induced enterocolitis syndrome (FPIES) is a non IgE-mediated type of allergy, characterized by a sepsis-like symptomatology after the ingestion of the triggering food.
Methods
We present the case report of a 4 months old infant that came to the emergency room with profuse vomiting and signs of hypovolemic shock. He was breastfed exclusively until the age of three months. At this time, it was made the first attempt to ingest milk formula but he vomited twice and had diarrhoea maintaining however a good general condition. By mother's choice, he maintained her milk exclusively. After two weeks the milk formula was restarted once a day. During the following two days he presented one vomit per day right after the milk ingestion. At day 3 he had persistent vomits, bloody diarrhoea, was becoming progressively more lethargic, being admitted in shock in the emergency department.
Results
To reverse the shock, the initial intervention was to give bolus of isotonic fluid (20ml/kg). Not knowing yet the cause of it, it was also given intramuscular adrenalin, methylprednisolone, and antibiotic therapy was initiated with ceftriaxone. Once admitted at the peadiatrics department he was fed with breast milk or with an amino acid-based formula and it was noticed a complete reverse of the symptomatology in less than 12 hours. Knowing the suggestive history and favourable clinical evolution after the suspension of the milk formula it was possible to conclude that he had a cow's milk food protein-induced enterocolitis syndrome (CM-FPIES). Two months later it was made the first attempt to initiate an extensively hydrolysed formula with recurrence of the initial symptomatology, but with good tolerance at the second try by the age of 12 months.
Conclusion
It is known that about 10-40% of the patients with CM-FPIES may not tolerate extensively hydrolysed casein-based formula and require an amino acid–based formula, this was the case with our patient.
The knowledge of this syndrome and the severity of its presentation allows the precocious eviction of the trigger and the unnecessary realization and administration of exams and drugs.

P06 Role of tropomyosin in determining cross reactivity between shellfish families during oral food challenges in children

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Keywords: Shellfish Allergy, Shellfish Crossreactivity

Introduction
In the Mediterranean, shellfish is a major cause of food allergy. Shellfish are divided in 3 families: crustacean, cephalopod and bivalve. Allergy to one family does not

necessarily imply symptoms will be present with the others. The pan-allergen tropomyosin, can help identify allergic patients. We aim to evaluate tolerance to shellfish in our population and identify immunological characteristics of patients associated to cross reactivity.

Methods

Prospective observational study at Sant Joan de Déu Hospital Barcelona, Spain. Children (≤18 years) with suggestive history of shellfish allergy and a positive skin prick test (SPT) and/or specific IgE (sIgE) to shrimp, squid and mussel extract were selected. Positive SPT wheal diameter ≥3 mm. Positive sIgE (ImmunoCAP) ≥0.35 kUA/L, upper limit 100 kUA/L. Positive molecular allergens (ImmunoCAP ISAC microarray) ≥0.3 ISU. Oral food challenges (OFC) were performed: controlled, single blind to shrimp, squid and mussel, to all patients except those with severe anaphylaxis to shellfish in the last 3 months.

Results

99 patients included, 68% male, mean age 8.8 years (± 4). In shrimp OFC, 53 patients were allergic and 46 tolerant. sIgE to shrimp differentiates between tolerant and non-tolerant patients, cutoff 7.11 kUA/L (Sensitivity:65% , Especificity:70%). sIgE to squid and mussel did not predict tolerance in OFC.[see table 1] Shrimp allergic patients were also allergic to squid in 58%; shrimp tolerant patients in only 4.3%. All patients who tolerated shrimp or squid also tolerated mussel. 8.7% of shrimp allergic patients and 12.9 % of squid allergic patients were also allergic to mussel. Patients allergic to shrimp were more likely to be allergic to squid (p 0.0001) and mussel (p 0.0047).

Pen m 1 differentiated between allergic and tolerant patients for shrimp (p 0.0031), squid (p≤0.0001), and mussel (p 0.0476). [see table 2]

Pen m 1 cut-off ≥ 12.6 ISU will distinguish between tolerant and allergic patients in all families. [see table 3] No differences were seen for Pen m 2 or Pen m 4.

Conclusion

Crustaceans are more allergenic than molluscs (squid and mussel). The best tolerated shellfish were bivalves (mussels).

Patients allergic to shrimp are more likely to also be allergic to squid and mussel. Higher levels of Pen m 1 were present in patients allergic to all shellfish families. In our population, sIgE to shrimp ≥7.11 kUA/L can predict allergy to shrimp.

Pen m 1 ≥12.6 ISU can predict allergy in OFC to different shellfish families.

Table 1: sIgE and SPT sensitization patterns in children allergic to shellfish.

Allergen	OFC status	% of patients (n)	Mean (±SD)	Median (range)	p
SPT shrimp extract (mm)	Tolerant to shrimp	46.5% (46/99)	2.3 (±29.4)	13.2 (0-111)	0.93
	Allergic to shrimp	53.5% (53/99)	23.6(±2.7)	16 (0-136)	
sIgE shrimp extract (kUA/L)	Tolerant to shrimp	-	10,2(±3.1)	1.78 (0.1-100)	0.006
	Allergic to shrimp	-	27,1 (±4,5)	12.85 (0.1-100)	
SPT squid extract (mm)	Tolerant to squid	67% (65/96)	24.8 (± 23)	18.4 (0-94)	0.026
	Allergic to squid	33%	14.2 (±18,6)	10.7 (0-92)	

		(31/96)			
sIgE squid extract (kUA/L)	Tolerant to squid	-	45.0 (±13,8)	0.76 (43-0)	0.575
	Allergic to squid	-	43.8 (±0,85)	1.66 (46-0.1)	
SPT mussel extract (mm)	Tolerant to mussel	95.6% (85/89)	3.75 (±3,7)	0 (0-15)	0.82
	Allergic to mussel	4.4% (4/89)	49.7 (±10)	0(0-58)	
sIgE mussel extract (kUA/L)	Tolerant to mussel	-	4.6 (±14,8)	1.1 (0-5.2)	0.11
	Allergic to mussel	-	70.3(±35,2)	5.95 (1.6-12)	

Table 2: Tropomyosin sensitization patterns in children sensitized to shellfish.

Allergen	OFC status	% of patients (n)	Mean (±SD)	Median (range)	p
Pen m 1 (ISU)	Tolerant to shrimp	42.2% (38/90)	6.9 (±1.9)	2.12 (0-55)	0.0031
	Allergic to shrimp	57.7% (52/90)	18.2 (±2.8)	14.7 (0-111)	
	Tolerant to squid	67% (58/87)	24 (±2.3)	2.7 (0-54)	≤0.0001
	Allergic to squid	33% (29/87)	82 (±12)	16 (0-111)	
	Tolerant to mussel	95.4% (84/88)	13.6 (±17,7)	6.21 (0-111)	0.0476
	Allergic to mussel	4.6% (4/88)	244(±12.2)	24.3 (13-36)	

Table 3: Best fit values in ROC curves of allergic vs. tolerant patients sensitized to Pen m 1.

OFC	Pen m 1 Cut-off value	Sensitivity	Specificity	Area under the curve	Standard error
Shrimp	12.7 ISU	54%	82%	0.72	0.055
Squid	12.6 ISU	75 %	79 %	0.81	0.049
Mussel	12.5 ISU	100 %	64 %	0.78	0.072

P07 Basophil activation tests based on surface expression of Basogranulin and CD63 and the release of Basogranulin and Histamine

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Keywords: Basophil Activation, Basogranulin, CD63, Histamine, Allergen

Introduction

Tests for basophil activation in vitro can provide useful means for investigating allergic sensitivity and even susceptibility to allergens. Methods adopted include measurement of allergen-induced membrane expression of CD63 and CD203c, or the release of histamine release. While close correlations have been demonstrated between findings with the two membrane markers, the extent to which they may be associated with mediator release is less clear. The advent of antibodies specific for basogranulin, a unique secretory product of basophils has opened the way for other means for examining basophil activation.

Methods

Blood was collected from subjects with a history of allergy to foods, grass pollen or house dust. Within two hours, either whole blood or basophils purified from blood using an immunomagnetic procedure were stimulated with specific allergen, anti-FcεR1 antibody or the peptide f-met-leu phe. Flow cytometry was performed with antibodies specific for CCR3 and CD63 or basogranulin, or cell supernatants were collected for measuring the release of histamine (by enzyme immunoassay) and basogranulin (by dot blotting with BB1 antibody). Comparisons were made between these methods for measuring basophil activation, and associations sought with clinical history, results of allergen challenge and skin prick test data

Results

Basophil activation in response to allergen was demonstrated by increased membrane expression of CD63 and basogranulin, and by the release of both histamine and basogranulin into cell supernatants. Bell-shaped or partial bell-shaped response curves were obtained with increasing concentrations of allergen. Flow cytometry with permeabilised basophils indicated that basophil activation was associated with a reduction in intracellular basogranulin content, increased cell surface expression (as indicated with non-permeabilised cells), and basogranulin release into supernatants. Measurement of basogranulin expression was as effective as that for CD63 as a means for investigating basophil activation to different allergens.

Conclusion

The methods developed for determining basophil activation in vitro may be valuable as new means for establishing allergic sensitivity.

P08 Beer Allergy: When malt is the answer

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Introduction

Beer is one of the most consumed alcoholic beverages around the world. In Portugal, its consumption may reach 52.4 litres per capita. Nevertheless, anaphylaxis with beer is rare. Our aim was to present a case of recurrent anaphylaxis to beer and the diagnostic work-up used to identify the allergenic protein.

Methods

A 71-year-old man presented in 2015 with three episodes of anaphylaxis. All included generalized urticaria, angioedema, dyspnea and wheezing. He was admitted to an intensive care unit after cardiac arrest during the last episode. Symptoms occurred immediately (<15 min) after the ingestion of the following foods, according to each episode: (1) beer and meat patties; (2) beer mixed with a soft drink; (3) beer and smoked pork meat. He subsequently tolerated pork, beef, egg, spices, cereals (wheat, barley, maize), wine and soft drinks.

Skin prick tests with standard aeroallergen extracts, cereals, profilin, Pru p 3 (nsLTP) and nuts and skin prick to prick tests with 8 different brands of beers, cereals (wheat and barley) and barley malt syrup were performed. Serum tryptase and serum specific IgE was measured for extracts from cereals, yeast (Sacharomyces cerevisae), hop (Humulus lupulus), wheat, barley, maize and barley malt. ISAC (Immuno Solid-phase Allergen Chip) was also performed. SDS-PAGE Immunoblotting with extracts from 4 different beers and barley malt as well as an immunoblot inhibition assay were carried out to assess possible common allergens shared between barley malt and beer.

Results

The SPPT were positive for barley malt and all the different brands of beer made from barley and/or wheat malt. No other sensitizations to the aeroallergens, foods, cereals or beer components tested were found. Immunoblot analysis detected IgE-binding bands at 17.5 kDa in the Carlsberg® and Franziskaner® beers and in the barley malt extracts, which could be a trypsin, oleosin or a heat shock protein. Another band with lower intensity was also identified at the 26-28 kDa range. Immunoblot inhibition assay showed that the barley malt syrup extract could inhibit the beer extract.

Conclusion

The recurrent, severe allergic reactions to beer may be explained by sensitization to a 17.5 kDa IgE-binding protein which is shared with barley malt. However, the patient

tolerates barley cereal ingestion. Malting process involved in converting barley into malt during the brewing could be responsible for the allergic sensitization in this case.

P09 Pollen food allergy Syndrome or food allergy?

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Keywords: Food Allergy, Pollen Food Allergy Syndrome

Introduction
Pollen-food allergy syndrome is a symptom complex almost exclusively localized to the oropharynx which is usually caused by certain fresh fruits and vegetables in individuals who are sensitized to pollens. Food allergy is an immunologic mediated adverse reaction after the ingestion of a food, clinical manifestations can be mild to lethal (anaphylactic shock).

Methods - Case Report
We present the case of a 21 year-old Mestizo boy. Medical student. Family history was positive for atopy. Since a toddler he presented respiratory allergy symptoms. Skin prick tests with commercial extracts were positive for multiple allergens. He was considered candidate for specific immunotherapy for house dust mite, cat and Bermuda grass. He presented adverse reactions to subcutaneous administration so route of administration was changed to Sublingual. He showed a significant response in terms of rhino conjunctivitis and asthma symptoms, which are both currently well controlled.

He also presented mild symptoms after ingestion of different food (mango, pineapple, chocolate). Skin prick-prick test was done but the patient presented acute respiratory and skin symptoms (anaphylaxis) so he was not considered candidate for food challenge and elimination diet was indicated.

Currently his main complains are still oral symptoms after ingestion (mainly fruits). Last event he presented pruritus and swelling of the mouth and throat just after ingestion of fresh mango (within 15 minutes) and a new non gastrointestinal symptom: pruritus of both arms, symptoms remitted with oral antihistamine. There is an overlap of symptoms between oral allergy symptoms and food allergy. Accurate diagnosis is necessary so an allergen component resolved diagnostics testing could be quite useful although there is limited availability of these tests in Mexican population. Knowledge of the specific sensitization of this particular case impacts both risk assessment and dietary management.

Conclusion
Within complex poly-sensitized patients affected by food allergy CRD show to be quite useful for identification within pollen food allergy and genuine food allergy.

P10 The effectiveness of probiotic use in children with Obstructive Bronchitis

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Keywords: Children, Bronchitis, Obstructive, Microflora, Probiotic

Introduction

High prevalence (250:1000) and mortality (2.6:100 000) rates of obstructive bronchitis (OB) among young children has revealed the necessity for an active search for new scientifically based measures of optimizing the diagnostic and treatment. The intestinal microflora is one of the unique systems that ensure the constancy of the macroorganism's internal environment. Due to the recurrent course of disease, early termination of breastfeeding, more frequent use of drugs, especially antibiotics, intestinal microbiocenosis often develop changes.

Methods

To investigate the qualitative and quantitative composition of the microflora of the intestines in young children with OB, has been conducted a bacteriological analysis of feces in 120 children aged 1month up to 3 years on the admission day and on day of discharge from the hospital. For 60 children with OB was additionally prescribed probiotic, contained Lactobacillus sporogenes и Bacillus coagulans. The average length of hospitalization was 10 days.

Results

With the symptoms of obstructive bronchitis, the majority of the examined patients (85.71%) have revealed clinical signs of dyspeptic syndrome. Regurgitation was noted in 33% of children, bloating - in 30 %, vomiting - in 18%, diarrhea was marked in 59%, periodic abdominal pain - at 49% patients with OB.

The study of intestinal microflora in children with OB revealed a violation of the qualitative and / or quantitative composition. There were significant changes in anaerobic flora, which was characterized by a decrease in the content of Bifidobacterii and Lactobacilli and an increase in the number of opportunistic bacteria (Proteus, Klebsiella, Enterobacter and Candida).

Studies have shown that inclusion of probiotic in the complex therapy of OB has contributed to the improvement of the microbiocenosis of the large intestine, especially in infants, as well as traditional treatment obviously intensified the phenomena of colon dysbiosis. In addition, inclusion in the treatment of OB in children probiotic has positively influenced the reverse development of inflammatory response by a decrease in the synthesis of anti-inflammatory (IL-4) cytokines in children of all age groups.

Conclusion

Taking into account the obtained data, our findings has shown the necessity for correction of microflora violations by assigning probiotic. Also, received results require further in-depth study of intestinal dysbiosis in in children with OB and their relationship with immunity disorders.

Friday 15 September 2017
Oral Abstracts presentation
17:45 - 18:45
Plenary Room

O01 Validation of a targeted mass spectrometry method for confirmation of peanut allergens in serum

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Keywords: Food Allergies, Peanut, Serum, Targeted Mass Spectrometry

Introduction

IgE-mediated reactions to foods (as diagnosed by food challenges) affect 0.9% of adults in Europe, and blood samples are commonly taken during these challenges. These samples can be used to assess whether the passage of allergen into the circulation may contribute to determining the severity of an allergic reaction by comparing uptake in healthy and allergic individuals undergoing food challenges. A targeted mass spectrometry (MS) method for analysis of clinically relevant peanut allergens has been developed to confirm the presence of peanut allergens in serum identified by other methods such as immunoassay and mediator release.

Methods

An LC-MS/MS method was developed, targeting peptides of 5 clinically relevant, peanut allergens (Ara h 1, Ara h 3 and Ara h 2,6 and 7), for the detection of peanut peptides in serum. Using serial isotopic dilution (SID) series and blank purchased serum spiked with a peanut protein extract, initial validation studies have been undertaken exploring the optimal methods of sample processing such as the use of a depletion column and an on filter digestion step. Data was acquired using a Waters Xevo TQ-S mass spectrometer interfaced with an ACQUITY UPLC M-Class equipped with an IonKey. Data analysis was undertaken using Skyline.

Results

Blank serum spiked with peanut protein and analysed after depletion showed differential matrix effects, with some peanut peptides having been lost during the depletion process. However depletion was shown to be an essential step resulting in less severe matrix effects in both the serum samples spiked with endogenous peanut protein and the SID series. The general limit of detection of peanut peptide targets was achieved in the femtomolar range, on column.

Conclusion
Further method development is required to allow detection of peanut at levels likely to be found in human subjects after ingestion of peanut, which is expected to be in the nanomolar range. This may be achieved through the use of Nano LC-MS/MS. This mass spectrometry method shows promise as a complementary tool to immunoassay methods to detect the presence of peanut allergen in serum of allergic subjects undergoing food challenges.

O02 Omalizumab-assisted oral tolerance in patients with severe LTP Syndrome

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Keywords: Omalizumab, LTP Syndrome, Tolerance, Pru P 3

Introduction
Peach non-specific lipid transfer protein (LTP), Pru p3, is the primary sensitizer in fruits/vegetables and responsible of severe allergic reactions in the Mediterranean area. Patients with LTP syndrome present food allergy symptoms with different fruits/vegetables, and in some cases a very restricted diet is required. Omalizumab has been shown to be an efficient treatment to diminish the risk and increase the success in oral immunotherapy to milk, eggs and peanuts.
Objective: To evaluate the efficacy of omalizumab in improving the tolerance to fruits/vegetables in patients sensitized to peach LTP (Pru p3) with severe systemic allergic reactions (SSR) to fruits/vegetables and a very restricted diet.

Methods
Retrospective study of patients with severe LTP syndrome and a restricted diet. LTP allergy was defined by a positive skin prick test to nPru p 3 (Bial-Aristegui-Spain), specific IgE >0.35 kU/l to rPru p3 (ImmunoCAP-ThermoFisher-Sweden) and severe systemic reactions with three or more non related fruits/vegetables. Patients were included from 2009 to 2016, with a follow-up between 1 and 7 years. Omalizumab was approved as off-label treatment and introduced, previous signed informed consent, 16-weeks prior to an oral-challenge performed in a day hospital setting to assess threshold of tolerance of walnut or commercial peach juice. Subsequently other fresh fruit/vegetables were gradually introduced in the diet.

Results
A total of 21 adult patients (18 women) were included in the study. In 50% of SSR a cofactor (mainly non-steroidal anti-inflammatory drug or exercise) was involved.The mean age was 33 (range 19-59). 23.8% had concomitant atopic dermatitis, 14.3% other food allergy different to LTP syndrome, 81% allergic rhinitis, 47.6% extrinsic asthma. After 16-weeks of omalizumab (average dose 300 mg/month) patients could gradually introduce new LTP containing foods (mainly walnuts and commercial peach juice) without major systemic symptoms in 82.6% of cases. Whereas before omalizumab 61.9% of the patients did not tolerate legumes, 76.1% fruit, 95.2% nuts, 61.9% vegetables, 28.6% cereals and 95.2% peach-pulp; after omalizumab reactions decreased to only 23.8%, 14.3%, 47.6%, 9.5%, 19% and 47.6% respectively.

Conclusion
Omalizumab increases tolerance of fruits/vegetables and improves the safety in

severe LTP allergic patients decreasing the number of SSR and daily symptoms, allowing a more varied and balanced diet.

O03 The impact of baked food matrices on structural and allergenic properties of food allergens

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Keywords: Food Allergy, Peanut, Ovalbumin, Gluten

Introduction
Several factors such as thermal processing and cooking practices may affect protein structure, digestibility of food allergens and their ability to stimulate the immune response. Peanut is one of the major food allergens affecting 1.6% of individuals in Europe1. It is rarely consumed in its native form and is usually roasted or processed into snacks and baked goods which may modify its allergenic activity. We propose that baking will affect digestion and bioavailability of allergens from peanut containing food.
To assess the effect of baking on the digestibility of peanut, gluten and egg proteins using a batch in vitro gastro-duodenal digestion model and to determine whether digestion changes immunoreactivity of proteolytic fragments of allergenic proteins.
Methods
Peanut containing baked muffin matrix was prepared according to recipe used for diagnosis and treatment of food allergy and contained 3.66% (w/w) peanut flour. Protein digestion was evaluated by SDS-PAGE and immunoblotting using different polyclonal antibodies specific for the 11S (Ara h 3) and 7S (Ara h 1) seed storage globulins of peanut, hen's egg ovalbumin and a monoclonal antibody specific for wheat gluten proteins.
Results
In the gastric digestion, peanut proteins (Ara h 1 and Ara h 3) were rapidly digested while ovalbumin was completely resistant, Gluten protein presented largely in the insoluble phase and was undigested. However, peanut proteins were digested during the duodenal phase; gluten was slowly digested from the insoluble phase, ovalbumin was stable to digestion with a lower molecular weight stable intermediate generated.
Conclusion
Baking causes the majority of allergen proteins from muffin to be largely insoluble and resistant to pepsin digestion. During subsequent duodenal digestion any remaining peanut allergens were broken down. In contrast ovalbumin passed into the soluble fraction and remained resistant to duodenal digestion.

O04 Epidemiology of food allergy in Czech Republic, results of DAFALL (Database of Food Allergy) registry after 3 years

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Keywords: Food Allergy, Epidemiology, Czech Republic

Introduction

Occurence of food allergy has significantly risen in recent decades with interesting differencies in incidence and types of food allergy in different areas. Aim of the study is to describe population of Czech food allergic patients.

Methods

Electronic registry DAFALL (Datase of Food Allergies) was founded in October 2014. Most common triggers of food reactions, threshold doses, processing of food allergens, laboratory test results including component resolved diagnosis, skin prick tests and food challenges results as well as allergology history of the patients were collected and put into the electronic database.

Results

During the first 30 months until June 2017, more than 1500 patients were enrolled from more than 30 collaborating allergology outpatient clinics, most of them children under age of 6 years (n=750), 25% children aged 6-18 years (n=380) and 370 adults. In children under 1 year of age, cow´s milk is the most frequent food allergen. 26 % of patients with CMA needed to be treat with amino acid formula. 60% of milk reaction were non-IgE mediated, compared to only 5% of egg allergic patients or 11% of hazelnut allergic patients with negative sIgE. Most common triggers of allergy in children between 1 and 6 years of age were milk, egg , tree nuts, peanut and fruits. In patients older than 6 years, significant allergens are tree nuts (hazelnut, walnut, almond), fruits (apple, peach, kiwi), vegetables, peanut and seeds, mainly the sesame seed and poppy seed. We found some interesting differencies in type of food specific for Czech population. We see relative low occurence of allergy to fish, shellfish and soy and on the other hand high numbers of patients reacting to seeds, especially poppy seed and sesame. About 50% of patient with peanut and tree nut allergies have been examined by CRD, so we are able to describe these patients from moleculary point of view.

Conclusion

DAFALL is the first project describing relevant data on food allergy in the Czech population. Projected period for data collection is 3 years, so we will finish at the end of the year 2017. More than 70% of patients enrolled are children so we will be able to give a detail description of the Czech food allergic children.

Saturday 16 September 2017
Poster discussion Session II
10:30 - 11:30
Poster Area

P12 Potential of the Basophil activation test in Pru P 3 sensitized population

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Keywords: Basophil Activation Test, Pru P 3, Ns-LTP

Introduction
A recent Western-European study showed that the majority of the positive sIgE to ns-LTPs is clinically irrelevant, which hampers ns-LTP related food allergy diagnosis. We aim at validating the basophil activation test (BAT) with the recombinant ns-LTP allergen from peach (rPru p 3). Secondly, we aim at evaluating whether the BAT could help to discriminate the different clinical phenotypes of positive sIgE reactivity to Pru p 3.

Methods
26 healthy controls and 83 patients with a positive sIgE (>0,10 kUa/L) to rPru p 3 were included in clinical centers in Barcelona (n=42) and Antwerp (n=41). 27 patients with a generalized reaction (GR), 22 with an oral allergy syndrome (OAS) and 34 tolerant (TOL) to peach. A basophil activation test (BAT) with rPru p 3 was performed (0,001;0,01;0,1;1 µg/mL).

Results
Dose-response curves showed 1 gr/ml with a cut-off of 8% activation to be most discriminative for healthy controls and Pru p 3 sensitized individuals with clear anaphylaxis, both in the Belgian and Spanish populations. A sensitivity of 95.12% (83.47-99.40%) and specificity of 100.00 % (86.77-100.00%) was reached. When comparing tolerant sIgE Pru p 3 positive patients to those with OAS/GR using the above mentioned stimulation concentration and cut-off value, the BAT shows to have a sensitivity of 73.47% (58.92%-85.05%) and specificity of 52.94 % (35.13-70.22%) for Belgium and Spain combined.

Conclusion
The BAT with rPru p 3 seems to be a good potential in the evaluation of clinical relevance of Pru p 3 sensitized individuals. Additional comparison between geographic regions and further validation is needed.

P13 Egg white allergy in children: comparison of oral food challenge and other diagnostic allergy tests

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Keywords: Hen'S Egg, Skin Prick Test, Oral Food Challenge, Allergen Components, Specific Immunoglobulins.

Introduction

Standardized double blind placebo controlled oral food challenges are considered to be a gold standard in food allergy diagnostics. However they require a lot of time, qualified personnel, are costly and may cause severe allergic reactions. Therefore there is a necessity to evaluate the value of alternative diagnostic methods in determining hen’s egg allergy diagnosis. Our study aim – to assess skin prick test (SPT), specific immunoglobulin E (IgE) to hen’s egg white and egg allergen components diagnostic value compared to oral provocation challenge (OFC) with hen’s egg white.

Methods

A retrospective data analysis of 42 patients aged 1-13 years old, who underwent open oral food challenge with pasteurized hen's egg white powder was made. Skin prick tests were considered positive when the wheal diameter was greater or equal to 3mm, sIgE concentration was considered elevated when was greater or equal to 0.35 kUA/L.

Results

21 (50%) of assessed patients were diagnosed with hen’s egg allergy. Compared to OFC with egg white powder, skin prick test with cooked egg white sensitivity was 73,3%, specificity – 78,6%, positive predictive value (PPV) – 78,6%, negative predictive value (NPV) – 73,3%. For SPTs with raw egg white the values were 100,0%, 35,7%, 57,1% and 100,0% accordingly. OFC results were positive to all patients whose SPT with cooked egg white wheal diameter was greater or equal to 6mm (p=0,01), and in 76,9% of those whose SPT with raw egg white wheal diameter was greater of equal to 7mm (p=0,02). Compared to OFC with egg white powder, specific IgE to egg white sensitivity was 77,8%, specificity – 64,7%, PPV – 70,0%, NPV – 73,3%. SIgE to ovalbumin values were – 87,5%, 100,0%, 100,0%, 75,0% and sIgE to ovomucoid – 52,6%, 88,9%, 83,3%, 64,0% accordingly. OFC results were positive to all patients whose sIgE levels were greater or equal to 3,5 kUA/L (p=0,01).

Conclusion

Skin prick tests with cooked and raw egg white, quantitative specific IgE to egg white and egg allergen components are valuable in aiding the determination of hen’s egg allergy, however they do not replace the necessity of oral food challenges. In patients with the suspicion of egg allergy, whose skin prick test with cooked egg white wheal diameter is greater or equal to 6mm and/or sIgE to egg white levels are higher or equal to 3,5 kUA/L, oral food challenges may be unnecessary to confirm egg allergy diagnosis.

P14 Food dependent exercise-induced anaphylaxis in a female triathlete - case report.

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Keywords: Anaphylaxis, FDEIA, Food Allergy, Exercise

Introduction
Food dependent exercise-induced anaphylaxis (FDEIA) is characterized by sequential food intake and exercise. Neither food intake nor exercise alone trigger the reaction. Pathomechanisms of FDEIA are largely unknown. Standardized diagnostic procedures for FDEIA have not been established yet.

Methods
25-year-old female triathlete, without any chronic health problems or diagnosed food allergy, experienced three episodes of FDEIA following running sessions. Two reactions were preceded by intake of salad containing lettuce, tomato and sunflower seed in 2015. In April 2016 after eating celery salad the patient developed again generalized urticaria, itching, swollen lips and tongue, hoarseness, hypotension and tachycardia. All three episodes occurred 2-3 days prior to menstruation however no NSAIDs were taken. Detailed medical review revealed frequent occurrence of intensive diarrhea and abdominal cramps as well as spontaneous itching during running sessions. The symptoms were induced only by running and never by biking or swimming, although performed often on the same day – due to triathlon sport activity.

Results
Physical examination and blood morphology did not reveal any abnormalities. In skin prick test and specific serum IgE antibodies sensitization to house dust mite, grass, oak and mugwort were found. The results of specific IgE with culprit and most common food allergens were negative. Tissue tranglutaminase and deamidated gliadin antibodies IgA- and IgG-class were <0,80 kU/I. The level of serum baseline tryptase was normal. The pulmonary function test was normal. Food challenge test and submaximal effort test were negative. The patient continues to take physical exercise daily avoiding any food 3-4 h before every training session. She has been provided with self-medication emergency kit and has been trained in self-management of anaphylaxis.

Conclusion
Although FDEIA is regarded IgE-mediated reaction the patient did not show sensitization to food allergens. Cross-reactive reaction to mugwort or grass pollens must be considered. The pathophysiological components of physical exercise such as increased gastric permeability, redistribution of blood flow or stimulation of proinflammatory response must be further investigated. This could contribute to the establishment of standardized protocols for diagnosis and management of FDEIA.

P15 Management of Acral Pruritus due to high nickel food with oral iron: a small study

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Keywords: Nickel, Acral Pruritus, Food Allergy

Introduction
Patient presenting with the symptom of Pruritus of Fingers and hands is not uncommon in clinical practice. When cause of the pruritus is not known, management become difficult. Rarely, food can be a cause of Acral Pruritus.
Methods
A small study was undertaken to know the cause of Acral pruritus involving fingers and hands.The study included seven patients (Age: 18-35 yrs; M: 2, F: 5) who had acral pruritus on and off without any skin manifestation, of different duration. All the patients were investigated thoroughly.
Results
Three patients were found to have Nickel allergy on Patch testing and two were found to have higher level of IgE. In the remaining 2 patients, no relevant cause was identified. All 3 patients with nickel allergy when challenged with Oral Nickel Sulfate, had aggravation of their acral pruritus. Blood Nickel level of these 3 patients were also found to be higher than the normal level. While taking the dietary history, it was found that all of them consume high nickel diet on and off.
These 3 patients were advised to take 15 mg of elemental Iron before Lunch and Dinner (Total 30 mg a day) for 8 weeks along with Low Nickel Diet(LND). Patients were asked to take oral antihistaminic on SOS basis only.
The frequency of occurrence of acral pruritus was reduced significantly in all three cases by the end of 2nd weeks. All patients were free from Pruritus by the end of the 5th week & they no longer needed to use oral antihistaminic agents. Blood Nickel level when tested again at the end of 8 weeks found to be with in normal level.
Conclusion
This small study showed that oral iron along with LND can cause an effective reduction in the occurrence of Acral Pruritus due to food in individual with Nickel allergy.

P16 Basophil Activation Test In Diagnosis Of Food Allergy

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Keywords: Basophil Activation Test, Oral Food Challenge, Total IgE, Specific IgE

Introduction
Food allergy impacts quality of life and can result in life threatening reactions. Currently, a double-blind placebo-controlled food challenge (DBPCFC) is advocated as the gold standard to confirm the diagnosis of food allergy. However, because of potentially life-threating reactions of in vivo testing, clinicians are trying to find suitable in vitro tests which could be used in a daily routine practice. Also, it is desirable to avoid the inconvenience and the high costs of treating patients who would be reasonably safe without any treatment and develop tolerance; therefore, a high

diagnostic specificity is required. Basophil activation test (BAT) is a functional test developed to monitor basophil activation upon allergen challenge by detecting the expression of membrane surface markers (CD63, CD203c) by flow cytometry. Our objective was to investigate the correlation between BAT results and conventional laboratory tests (total IgE, specific IgE (sIgE)) and severity of clinical symptoms to a certain food allergen.

Methods

A group of 98 children (65 male, 33 female, median age 7), with a history of food allergy was recruited. A clinician performed physical examination, upon which blood samples for total IgE, sIgE and BAT were taken. Blood samples for BAT were tested to three different allergen concentrations (0.225ng/ml, 2.25ng/ml, 22.5ng/ml)

Results

Patients were categorized into two groups according to severity of symptoms: 25.5 (25?) of them had severe food allergy symptoms. We found significant correlation between BAT and total IgE in medium ($p=0.011$, CI 0.05-0.43) and high allergen concentrations ($p=0.012$, CI 0.05-0.42). The correlation between BAT and sIgE level was significant for all allergen concentrations ($p_{low}=0.0004$, CI 0.16.0.51; $p_{middle}<0.0001$, CI 0.4-0.68; $p_{high} < 0.0001$, CI 0.39-0.67), as well as the correlation between BAT and severity of symptoms ($p_{low}=0.0006$, CI 0.15-0.5; $p_{middle}<0.0001$, CI 0.44-0.7; $p_{high} < 0.0001$, CI 0.52-0.75).

Conclusion

Our results show that BAT has a good correlation with severity of clinical symptoms and sIgE in food allergy diagnosis and can be used as a predictor of severity of allergic reactions. This implies that BAT could be a useful tool for assessement of patients suitable for oral food challenges.

P17 Clinical Features Of Pediatric Patients With "Fish–Chicken Syndrome"

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Keywords: Fish Allergy, Chicken Allergy, Fish-Chicken Syndrome

Introduction

In 2012, the first case of fish-chicken allergy was reported, demonstrating cross-reactivity between α and ß parvalbumin. In 2016, parvalbumin (Gal d 8) was detected in chicken legs and wings, and enolase (Gal d 9) and aldolase (Gal d 10) were identified in chicken breast. It was proposed that fish and chicken allergens are cross-reactive, hence the "Fish-Chicken Syndrome". The aim of our study was to characterize children with possible fish-chicken allergy

Methods

Retrospective, observational study, Sant Joan de Déu Hospital, Barcelona, Spain. Records were searched of patients with suggestive history of chicken allergy and positive sIgE to chicken from 2013-2017. Data on age of onset, sex and personal history of atopy was recorded. Patients with a history of chicken and fish allergy were

selected. These patients had had skin prick tests (SPT) to chicken and fish (hake, cod, sole, sardine) (wheel d≥3mm); sIgE to chicken and fish (hake, cod, sole, sardine) by ImmunoCAP (≥0.1 kUA/L positive) and ImmunoCAP ISAC (≥0.3 ISU positive). Single-blind, controlled, oral food challenges (OFC) to fish and chicken were performed

Results

56 patients had positive sIgE to chicken, 26 of them were excluded (incomplete data). History of 30 patients was recorded, of those, 9 had symptoms with fish and chicken (Table 1) 77.8% (n=7) were female. Personal history of atopic dermatitis 66.7% (n=6), respiratory allergy 33.3% (n=3), other food allergies 88.9% (n=8), the most common egg and shellfish both in 44.4% (n=4). Age of onset to fish allergy mean 33 months (9-60), and chicken: 64 months (12-168). Symptoms with fish were predominantly cutaneous 88.9% (n=8) and 2 cases of anaphylaxis. Symptoms with chicken: cutaneous 66.7% (n=6), 1 vomit, 1 anaphylaxis and 1 oral allergy syndrome (OAS). Clinical reactivity to chicken meat was confirmed by OFC in 2 of 6 patients (33.3%) with OAS and urticaria. OFC to fish was positive in 4 patients (66.7%) with urticaria and anaphylaxis. sIgE for fish and chicken were positive in all patients. SPT to fish were positive in all patients and 7 also to chicken. 4 patients had positive Gad c 1, 4 Pen m 1 and 1 Gal d 5

Conclusion

A relationship between patients allergic to fish and chicken can be explained by cross-reactive allergens. Fish–chicken syndrome is found in our population and should be assessed in the clinical practice. In almost all of our patients, the initial symptoms were reported for fish and developed later for chicken, suggesting primary sensitization to fish

Table 1: Clinical Features, Associated Findings, And Laboratory Results Of Patient With Chicken-Fish Syndrome. Sant Joan de Déu Hospital, Barcelona Spain.

Case	Sex*	Symptoms /age of onset (month)		IgE (KUA/L) / Prick (mm)					Gal D5 (ISU)	Pen m1 (ISU)	Gad c 1 (ISU)
		fish**	chicken**	chicken	Cod fish	Sole fish	Hake	Sardine			
1	F	U / 12	OAS / 72	2.8 / Neg	ND /ND	ND / 10	100 / 13	100 / 9	Neg	1.16	78
2	F	U / 9	OAS / 168	0.5 / 4	ND / ND	83.7 / 10	100 / 11	ND / 9	Neg	4.5	100
3	F	U, Br /60	U / 72	16.2 / 15	ND / ND	10.8 / 10	22 / 5	18.5 / 7	Neg	Neg	Neg
4	F	U /36	U / 72	1.9 / 5	0.5 / 4	ND / 5	0.64 / 4	ND / 4	Neg	2.3	Neg
5	F	U /48	U / 49	0.3 / Neg	ND / ND	ND / ND	1.7 / ND	ND / ND	ND	ND	ND
6	F	U /48	U / 12	2.5 / 5	5.2 / 12	2.6 / 13	4.7 / 16	4.5 / 16	ND	ND	ND

7	F	U, AE / 9	AE, Br / 48	0.3 / 4	ND / 12	10.6 / 6	14 / 10	ND / 7	ND	ND	ND
8	M	Ap OAS / 60	V, D / 60	2.6 / 5	5.8 / 8	ND / 6	5.7 / 12	2.5 / 14	Neg	Neg	1.8
9	M	U / 12	U, Co / 60	13.5 / 8	ND / 4	ND / 6	ND / 6	ND / 4	19	63	23
* F: female, M: male, ND: no data; ** U: Urticaria, Br: bronchospasm, Ae: angioedema, AP: abdominal pain, OAS: oral allergy syndrome, V: vomiting, D: diarrhea, Co: conjunctivitis, *** Neg: negative											

P18 Can Profilin be associated with severe symptoms in tomato-allergic patients?

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Keywords: Anaphylaxis, Immunoblotting, Profilin, Tomato Allergy

Introduction
Tomato can be associated with oral allergy syndrome in pollinic patients, due to profilin sensitization. Recent data suggests that profilin can be associated with severe systemic reactions. Aim: To describe a case of tomato anaphylaxis, where profilin seems to be the allergen involved.

Methods
The patient was submitted to skin-prick test(SPT) with aeroallergens and food extracts and to culprit food prick-prick test(PPT). Specific IgE(sIgE) were evaluated by ImmunoCAP and ISAC. SDS-PAGE Immunoblotting assay was performed.

Results
67 years old male, with ischemic heart disease(IHD) and seasonal rhinitis, was attended in emergency after a sudden episode of dyspnea, palate angioedema, generalized urticaria and palmar-plantar itchiness, 1 h after a meal with cod, tomato salad and melon with improvement after IM adrenaline. Later, had a 2nd episode of generalized urticaria, eyelid edema and dyspnea, 40 mins after eating chicken, sausages, tomato and lettuce salad and a 3rd episode of palmar itching 1 h after a Bolognese meal, with spontaneous resolution. No NSAID, exercise or infection associated. SPT revealed hypersensitivity to grass, plane, olive, plantain, palm profilin Pho d2 and to tomato extracts and were negative to Pru p3(nsLTP). PPT were positive to fresh tomato pulp(8x8mm) and peel(5x5mm) and to baked tomato pulp(7x7mm)

and peel(5x4mm). He had positive sIgE(> 0.35 kU/L) to grass and olive pollens and negative to tomato. ISAC showed moderate/high levels of nSal k1, low levels of nCyn d1, rOle e1, rBet v2, rMer a1 and nMUX F3. The immunoblotting detected a 15 kDa-IgE binding band in tomato pulp and no band in the peel. A similar molecular mass band was detected with an anti-watermelon-profilin rabbit serum, suggesting that is a profilin. A SDS-PAGE Immunoblotting-inhibition with tomato pulp solid phase and inhibitory phases of pollen extracts(Olea europea, Plantago lanceolata, Salsola kali, Platanus acerofolia) and of purified profilins(Art v4, Pho d2, Ole e2) showed total inhibition on the 15-kDa-IgE band tomato pulp by all. An anti-Pru p 3 assay with tomato-seed extract, detected a band that did not match with the one detected with the patient's serum and that also appeared in the negative control with smaller intensity. Given patients' IHD an oral Pho d 2 provocation was not made.

Conclusion

We report a case of a severe systemic reaction to tomato where in-vitro study suggest that this reaction might be linked to a profilin sensitization.

P19 Food protein-induced allergic Proctocolitis to multiple foods

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Keywords: Food Allergy, Allergic Proctocolitis, Non-Ig E Mediated Gastrointestinal Allergies

Introduction

Food protein-induced allergic proctocolitis is a non-immunoglobulin E mediated gastrointestinal allergy. It is a benign condition commonly found in a well-appearing infant.

It manifests with bloody stools and the typical age of onset is usually between one and four weeks. Usually infants may react to cow milk, soy, egg, and/or wheat in maternal diet through breast milk. Diagnosis of food protein induced allergic proctocolitis is based on the recognition of specific symptom patterns. A trial of elimination diet is part of the diagnostic criteria. The gold standard that confirms the diagnosis after symptom resolution is the food challenge. Eczema is present in approximately 22% of the breastfed infants. Our objective is to describe a case of food protein induced allergic proctocolitis to multiple foods.

Methods

The patient is a 5-month-old male child, who was referred to the pediatric clinic with erythematous, intensely pruritic skin with scratch lesions localized on his face and limb extension surfaces and yellow stools with low consistency, absorbed in diapers, with mucus and blood, 2-3 times a day, with good general condition. He is breastfed. These symptoms were present after the first month of life. Allergy investigations including serum milk-specific immunoglobulin E and total immunoglobulin E were performed.

Results

The serum milk-specific immunoglobulin E and the total immunoglobulin Ig E were negative. Gradually cow milk, beef, egg, carrot, tomato and potato were removed from the mother's diet. At 6 old months, the dermatitis lesions are improved, the

consistency of the stools is normalized, the pathological elements of mucus and occult bleeding have disappeared. After 6 weeks, we tried to reintroduce sequentially in the mother's diet the food culprits; the food challenge led to rectal bleeding in 24 hours. At 1 year old he has tolerated carrot, tomato, egg and potato, but not cow milk and beef. The patient at 2-year-old is still reacting to cow milk and beef. He has not yet achieved tolerance and the serum milk-specific immunoglobulin E is still negative.

Conclusion

Non-immunoglobulin E mediated gastrointestinal allergies are relatively common to infants, but are likely under-diagnosed. Food protein-induced allergic proctocolitis is benign and it resolves usually before the age of 1 year old for most patients, after maternal elimination of the offending food.

P20 Autoclaving treatments applied to peanuts to reduce final immunoreactivity

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Introduction

Peanut allergy is one of the most frequent allergies especially affecting developed countries. In order to reduce the risk of eliciting undesired reactions, a number of technological approaches have been devised to inhibit/remove allergens in order to deliver a hypoallergenic food. In the present work we investigated alternative strategies based on thermal treatments like autoclaving to decrease peanut immunogenicity.

Methods

Raw and autoclaved peanuts were extracted, separated on SDS-PAGE and further submitted to immunoblot analysis using sera of allergic patients. Each individual extract was further analysed by ELISA in order to estimate the residual immunoreactivity of the processed peanuts. The most resistant allergenic proteins displayed in the gel were finally identified by LC-HRMS analyses.

Results

A progressive reduction in the intensity of the major allergenic bands was highlighted in autoclaved samples; such behaviour was even more evidenced with a total disappearance of the major allergenic proteins when samples were preliminary exposed to hydration. These data also confirmed results obtained by ELISA analysis. Raw and treated peanut material were finally submitted to immunoblot analysis in order to assess the residual immunogenicity of the treated peanuts.

Conclusion

Hydrating peanut seeds prior to autoclaving increased the efficacy of the thermal treatment contributing to the disappearance of the main allergenic proteins and

reducing significantly the final immunoreactivity, as assessed by ELISA tests and immunoblot analysis.