nitric acid synthase). Asthma is defined as a chronic inflammatory disease of the airway and is the most common chronic disease in children characterized by wheezing. Wheezing may occur even once it is up to 50% of all infants and children under 3 years old and topical dermatitis affecting over 10% of children.

**Objective:** To define incidence and prevalence of asthma and other allergic diseases in population of less than 5 years of age in Juarez Hospital Mexico.

**Results:** We evaluated 11,346 allergic patients on 4 years period (January 2007 to December 2010). Two thousand three hundred ninety six were ≤5 years (21.11%); 10.2% a year old or younger, 16.7% 2 years old, 18.5% had 3 years, 24.7% of 4 years and 30% 5 years old. Nine hundred ninety two two patients (41.4%) were females and 1,404 (58.6%) male. One thousand two hundred sixty eight patients (51.7%) had only one diagnostic finding as most frequent cause of consultation allergic rhinitis (59.6%), followed by asthma (23%), atopic dermatitis (5.1%), prurigo by insect (4.3), immunodeficiency (1.2%) the rest had various pathologies such as contact dermatitis, gastro esophageal reflux, oral allergy syndrome, and others. The remaining patients 48.3% (1158) had more than one diagnosis. The most common associations were asthma and rhinitis 716 (30%), 72 patients (3%) with asthma and atopic dermatitis, 48 patients (2%) with some immune deficiency and asthma and/or rhinitis, 12 (0.5%) with purigo and atopic dermatitis, 8 (0.3%) with urticaria and 11 patients (4.5%) with other diagnoses such as reflux or infections.

**Conclusions:** Allergic diseases are common on pediatric population and children under 5 years old asthma and rhinitis is the principal cause of consultation.

**FOOD ALLERGY**

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**Effect of Hydrolysis and Polymerization on Bovine Beta-lactoglobulin Immunoreactivity**

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**Methods:** The β-Lg from milk (IgE 216.20 ug mL−1) was polymerized in presence of Cys it decreased considerably after pepsinolysis. After modiﬁcation the samples were submitted to in vitro digestion, simulating gastric and duodenal conditions (Moreno, 2005; Martins et al, 2010). The characterization of the samples was performed by electrophoresis (SDS-PAGE) and Reversed-phase high performance liquid chromatography (RP-HPLC). The allergenicity of the protein was measured by ELISA, using sera of milk allergic patients.

**Results:** The untreated β-Lg was resistant to pepsin while the samples polymerized by TG showed an increased in the susceptibility to pepsin, since a predominance of low molecular mass (MM) peptides, 3.0 to 6.0 kDa (by SDS-PAGE) and peptides with low hydrophilicity (by RP-HPLC) were detected. After duodenal digestion, the polymerized samples showed an increased in the intensity of the peaks with high hydrophilicity, indicating a potential susceptibility of polymerized β-Lg to GI digestion. Immunoreactivity to IgE from sera of allergic patients was retained for β-Lg polymerized after heat treatment, even after in vitro gastric digestion; while for the sample polymerized in presence of Cys it decreased considerably after pepsinolysis. After duodenal digestion, both polymerized samples showed an important decrease in the immunoreactivity response, compared to untreated β-Lg.

**Conclusions:** These findings showed that the polymerization alters the susceptibility of β-Lg to GI digestion and could have implications in the allergenic characteristics of this protein.

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**Immunoreactivity of Polymerized and Digested Beta-lactoglobulin to IgE from Milk Allergic Patients**

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**Methods:** The β-Lg (7% w/v), donated by Davisco Inc., was modiﬁed by 2 different methods (1) heat treated (80°C/60 min) and polymerized by TG (10 U g−1 protein), and (2) polymerized by TG in the presence of the reducing agent Cysteine - Cys (0.1 mol L−1). After modiﬁcation the samples were submitted to in vitro digestion, simulating gastric and duodenal conditions (Moreno, 2005; Martins et al, 2010). The characterization of the samples was performed by electrophoresis (SDS-PAGE) and Reversed-phase high performance liquid chromatography (RP-HPLC). The allergenicity of the protein was measured by ELISA, using sera of milk allergic patients.

**Results:** The untreated β-Lg was resistant to pepsin while the samples polymerized by TG showed an increased in the susceptibility to pepsin, since a predominance of low molecular mass (MM) peptides, 3.0 to 6.0 kDa (by SDS-PAGE) and peptides with low hydrophilicity (by RP-HPLC) were detected. After duodenal digestion, the polymerized samples showed an increased in the intensity of the peaks with high hydrophilicity, indicating a potential susceptibility of polymerized β-Lg to GI digestion. Immunoreactivity to IgE from sera of allergic patients was retained for β-Lg polymerized after heat treatment, even after in vitro gastric digestion; while for the sample polymerized in presence of Cys it decreased considerably after pepsinolysis. After duodenal digestion, both polymerized samples showed an important decrease in the immunoreactivity response, compared to untreated β-Lg.

**Conclusions:** These findings showed that the polymerization alters the susceptibility of β-Lg to GI digestion and could have implications in the allergenic characteristics of this protein.

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**Identification of Novel Allergens in the Fish Parasite Anisakis simplex**

Christiane Fæste Kruse, PhD,1 Maaike Dooper, PhD,1 Alvaro Daschner, MD,2 Wolfgang Egge-Jacobsen, PhD,3 and Ellian Egaas, PhD1

**Methods:** The nematode Anisakis simplex is a marine parasite that causes allergy as well as anisakiasis in human. Here, we describe the identification of 4 novel allergens in anisakis.
Methods: Binding of human IgE to anisakis and house dust mite proteins was investigated by immunoblot with serum from individuals sensitized to anisakis or shrimp. IgE binding patterns in the immunoblots were used for the identification of major Anisakis allergens, which were analysed by mass spectrometry-based proteomics in ESI-Orbitrap, after separation on SDS-gel.

Results: Four new allergen candidates were identified. The first identified allergen was enolase, which is related to the cockroach allergen enolase. The other allergens were Heat Shock Protein-70 (HSP 70), tubulin, and glutathion-S-transferase, which are also present as allergens in house dust mite.

Conclusions: Here we describe the identification of 4 novel IgE binding allergens in anisakis. The allergens might explain IgE cross-reactivity between anisakis and house dust mite or cockroach.

416 Milk Components as a Tool in Predicting Tolerance in Cow’s Milk Allergy
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Background: One important point in cow’s milk allergy (CMA) is to establish predictive factors in acquiring tolerance. The aim of this study is to evaluate if a ratio of milk components: alpha-lactalbumin (α), beta-lactoglobulin (β), casein (C) and whole milk (WM) can contribute to predict tolerance development.

Methods: It was a retrospective study that included patients with previous diagnosis of CMA evaluated at 6 years old. CMA was defined as a positive double blind placebo-controlled food challenge, open challenge or confirmed anaphylaxis plus positive specific IgE to cow’s milk (higher than 3.5 kU/L or positive skin prick test), and tolerance was defined as acceptance of cow milk without previous symptoms. Specific IgE analysis to WM, α, β, and C were performed through Immunocap (Phadia AB). Ratios of milk components and whole milk (α/WM, β/WM, C/WM) were calculated and compared the results in 2 study groups: tolerant and persistent at 6 years old. Since values from both α/WM and β/WM ratios didn’t follow a normal distribution, Mann-Whitney test was used to compare groups. For C/WM ratios, Student’s t test was used as values were normally distributed.

Results: It included 49 patients (27 male/22 female), 24 tolerant and 25 persistent. Average age of Immunocap test was 2.7 years (SD = 1.4). Comparing the results from 2 groups, persistent and tolerant it was detected: no difference in α/WM (P = 0.055, Mann-Whitney test), higher levels of β/WM in persistent group (P = 0.023, Mann-Whitney test) and also higher levels of C/WM in persistent group (P = 0.004, Student’s t test).

Conclusions: Higher ratios involving beta-lactoglobulin or casein components were detected in persistent patients. Thus, evaluating these markers precociously can be helpful in predicting CMA evolution.

417 Uncommon Occupational Allergy to Rice: as a Food Allergen
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Background: Rice (Oryza sativa) belongs, with other cultivated cereals, to different tribes of the Poaceae family. It is one of the most widely produced and consumed cereals in the world but hypersensitivity reactions to this grain are uncommon. Most reports describe an immunologically-mediated urticaria due to contact with raw rice or reactions after the inhalation of rice fumes, whereas reports of immediate hypersensitivity reactions after ingestion of rice are scarce.

Methods: Patient 1 (P1): A 40-year-old-man, a professional cook, presented 2 episodes of generalized urticaria minutes after rice ingestion. He tolerated the inhalation of vapours during rice-boiling, but reported itchy skin and erythema after rice handling. Patient 2 (P2): A 30-year-old-woman, pizzeria worker for the last 10 years, complaint of sneezing and rhinorrea after handling rice for the last 2 years, and presented diarrhea and dysphagia after rice ingestion during the last year. One week before consulting she presented eyelid angioedema, chest tightness and abdominal cramping after doing exercise right after eating rice. None of the patients reported any additional atopic background. Skin prick tests with common inhalants and cereals extracts, Pru p 3 extract, prick-by-prick test with rice and rice flour and specific IgE determinations to rice were carried out in both patients.

Results: Skin prick tests to rice were positive in both patients (wheat diameter >3 mm). Skin prick-by-prick with rice and rice flour was also positive in patients 1 and 2. Serum specific IgE determinations against rice showed values of 0.8 kU/L and 1.48 kU/L for P1 and P2, respectively, out from a total IgE of 32.8 UI/mL and 23.7 UI/mL, respectively. SPT to common inhalants, to the rest of the cereals and to Pru p 3, showed a negative result.

Conclusions: We present 2 work-related cases of rice allergy with an unusual display and different clinical manifestations (urticaria, rhinitis and anaphylaxis) in 2 patients without atopic background and who worked handling rice and rice flour. No cross-reactivity with usual panallergens as LTP seemed to be involved.

418 Frequency of Food Hypersensitivity Mediated by IgG in Patients Received in a Venezuelan Laboratory During 2011
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Background: Foods could cause adverse reactions, which manifested with similar symptoms that could complicate the diagnosis, that is, gastrointestinal disease, rashes, edema, eczema, asthma, anaphylaxis, others; these reactions may be mediated or not by immunological processes. In the past years there were many research studies related with different types of immunological reactions, like for example those mediated by IgG, which are characterized with delayed and insidious manifestations. These reactions could appear in hours or days after ingestion of a particular food. The intake of “toxic food” can lead to immunological reactions, which includes the formation of immune complexes, able to increase the development of gastrointestinal, dermatological, neurological, muscular and respiratory disorders. The objective of this study was to evaluate the frequency of significant specific IgG titers against some foods in patients referred to Coppodiagnostics Laboratory, (Caracas, Venezuela, an ISO 9001:2008 certified laboratory) from January to August 2011.

Methods: There were a total of 148 patients referred for serum specific IgG evaluation against foods. We measured patient’s specific IgG titers against 45 foods using a commercial direct ELISA method (Dr. Fooke Labs, Germany), which is designed for the detection and quantification of specific IgG.

Results: We found detectable levels of specific IgG titers in highest frequency for: milk 69%, cheese 67%, egg 64%, gluten 54%, sugar cane 51%, followed by wheat meal, rye Meal, and other foods in smaller proportions. Moreover, in 46 patients with known clinical history, including gastrointestinal symptoms, respiratory symptoms, pervasive developmental disorder (PDD) and autism, we detected levels of specific IgG to a significant number of foods simultaneously.