Poster Session 28

From bench to bedside: new agents and components

857
Cross-reactive carbohydrate determinants in the diagnosis of occupational allergy

Wiszniewska, M[1]; Zgorzelska-Kowalik, J[2]; Palczynski, C[2]; Nowakowska-Swirta, E[2]; Walusiak-Skorupa, J[2]
[1]Nofer Institute of Occupational Medicine, Department of Occupational Diseases & Toxicology, Lodz, Poland; [2]Poland

Background: Cross-reactive carbohydrate determinants (CCDs) are carbohydrate structures of plant and animal glycoproteins. The role of CCDs in diagnostics of occupational allergy remains unclarified and its clinical relevance is still questioned. Specific IgE to CCDs can be highly cross-reactive and frequently occurs in patients with polyvalent not occupational sensitization. The aim of the study was to evaluate the prevalence of CCDs in the subjects with suspected occupational allergy and its role in differential diagnostics.
Method: The study group included 201 patients. They underwent clinical examination, skin prick tests (SPT) to common and occupational allergens, allergen-specific IgE (asIgE) determinations, spirometry and specific inhalation test. Moreover, asIgE to CCDs (CAP-RAST) were evaluated in all subjects.
Result: Specific serum IgE to CCDs were found in 14 subjects (7%). In most cases, levels of asIgE to CCDs remained in class 1, only in three sera the level reached class 2. Positive SPT to common allergens were found more frequently in CCDs positive subjects (64.3% of the group), while occupational SPT were found rarely in that group (28.6% of the group). Isolated allergy to occupational allergens was found only in one patient with asIgE to CCDs. In all subjects with asIgE to CCDs, the asIgE to occupational allergens were detected.
Conclusion: Specific IgE to CCDs were found in 7% subjects with suspected occupational allergy. The results indicate that detection of CCDs in serum is not helpful in differentiation diagnostics of occupational and non-occupational allergy.

858
Gene expression signatures in BALB3T3 fibroblasts exposed to cobalt micro/nano-particles and cobalt ions

Di Gioacchino, M[1]; Petrarca, C[2]; Verginelli, F[3]; Sabbioni, E[4]; Mariasni-Costantini, R[3]
[1]G. d'Annunzio University, Allergy and Clinical Immunology, Chieti, Italy; [2]G. d'Annunzio University Foundation, Allergy and Immunotoxicology, Chieti, Italy; [3]G. d'Annunzio University Foundation, Molecular Pathology, Chieti, Italy; [4]ECSIN, Rovigo, Italy

Background: Nanoscale materials are increasingly employed in several industrial applications, for instance by metals extraction and processing factories, as well as in biology and medicine, by drugs and prosthesis, and throughout large use products, as sunscreen and food package. Despite their wide predicted utilizations, little research has been carried out on the potential toxicity of nanomaterials. To explore possible biological effects relevant to toxicity, that could depend on the physical characteristics of compounds, we investigated the global gene expression patterns of BALB3T3 A31-1-1 cells exposed or not to 1 mM of cobalt as microparticles (Co-micro), nanoparticles (Co-nano), and ions (Co 2+).
Method: All exposures were carried out at semi-confluence for 72 h, as determined by toxicity assays for toxic metals. Global gene expression analyses were carried-out using 70mer murine 13.443 oligonucleotide microarrays (Operon version 1.1). Data were normalized and elaborated using MIDAS and SAM programs. Molecular interactions networks were constructed applying gene ontology, canonical pathway and functional network analyses with the IPA tools software (http//www.ingenuity.com).
Result: From this analysis emerged that treatments mainly affected different pathways: interferon, apoptosis, JAK/STAT and death receptor signalling in respect to the nanoparticles exposure; nucleotide sugar metabolism, cell cycle check point G1/S, hipoxia signalling and ubiquitination in respect to the microparticles; no statistically significant pathway was fund after ions treatment. Real Time-PCR analysis confirmed that exposure to Co-micro, -nano, and -ions down-modulate Rab18, a member of the RAS oncogene family, and CAV2, which encodes a key constituent of caveolae, integral membrane components. These two genes represent candidate biomarkers of specific effects on the regulation of intracellular membrane trafficking after Co-micro, -nano, and -ions exposure.
Conclusion: Our results shown that treatment with Co-micro and Co-nano differentially activates cellular pathways that control defense and repair mechanisms.

859
Combined nasal exposure to sodium hypochlorite and ovalbumin induces airway hyperresponsiveness in mice through activation of the TRPA1 channel and mast cells

Hox, V[1]; Vanoirbeek, J[2]; Callebaut, I[1]; Bobic, S[1]; De Vooght, V[2]; Vennekens, R[3]; Liston, A[4]; Ceuppens, J[1]; Nemery, B[2]; Hellings, P[1]
[1]KULeuven, Laboratory of Experimental Immunology, Leuven, Belgium; [2]KULeuven, Research Unit of Lung Toxicology, Leuven, Belgium; [3]KULeuven, Laboratory of Ion Channel Research and Gene Expression Unit, Department of Molecular Cell Biology, Leuven, Belgium; [4]VIB and KULeuven, Leuven, Belgium

Background: According to recent epidemiologic studies, attendance to chlorinated swimming pools has been associated with airway problems as allergies, bronchial hyperresponsiveness and asthma. Sodium hypochlorite (NaClO), the main pool disinfectant has been suggested as a possible etiologic factor.
Aim: To investigate the effects of NaClO on airway function and allergic sensitization in mice.
Method: Naive mice received 1–7 nasal instillations of ovalbumin (OVA) on alternate days with/without prior NaClO (3 ppm active chlorine) instillations. 48 h after 1, 3, 5 and 7 instillations, airway hyperreactivity (AHR) was evaluated by the flexivent® technique, differential cell counts were performed on cytospin preparations of broncho-alveolar lavage fluid (BALF), serum OVA-specific IgE and lung Th2 cytokine levels were measured. The role of substance P (SP) in the airway response was investigated by administration of the neurokinin (NK) 1 receptor antagonist RP67580. To check the involvement of the transient receptor potential (TRP) channels, the protocol was repeated in $TRPA1^{-/-}$ and $TRPV1^{-/-}$ mice. The role of mast cells was assessed by the use of mast cell deficient $Kit^{W\text{-}sh}/Kit^{W\text{-}sh}$ mice.

Result: Combined nasal NaClO-OVA exposure induced airway hyperreactivity (AHR) to methacholine in the absence of airway inflammation and systemic OVA specific IgE. AHR was already induced after a single combined exposure to NaClO-OVA and this was not observed after either OVA or NaClO alone. The AHR response was absent after pretreatment with RP67580. NaClO-OVA induced AHR in $TRPV1^{-/-}$ mice, but not in $TRPA1^{-/-}$ mice. Mast cell deficient mice did not develop an AHR in response to NaClO-OVA exposure.
Conclusion: Repeated exposure of the upper airways to low concentrations of NaClO in combination with OVA does not facilitate sensitization in mice. However, combined nasal NaClO-OVA exposure induces AHR in the absence of allergic inflammation. This effect appears to involve TRPA1, mast cells and release of substance P, suggesting a neuro-immune interaction.

860
The prevalence of occupational rhinitis in Hungary

Endre, L[1]; DuBuske, L[2]
[1]Heim Pal Children Hospital, Budapest, Hungary; [2]Immunology Research Institute of New England, Gardner, MA, United States

Background: Occupational rhinitis (OR) is an inflammatory disease of the nose, which is characterised by intermittent or persistent symptoms, arising out conditions attributable to a particular work environment and not to stimuli encountered outside the workplace. Symptoms of OR including nasal congestion, sneezing, rhinorrhea, itching, nasal airflow limitation, are very similar to the symptoms of allergic rhinitis caused by allergens. According to the Hungarian governmental 27/1996 NM Departmental Order, OR in Hungary is a reportable disease.
Method: Data regarding reporting of occupational rhinitis and asthma was surveyed from the official records of the National Labor and Hygiene Institute, and of the National Statistical Institute of Hungary between 1997 and 2009.
Result: During the period 1997–2009, not one case of OR was reported in Hungary. During the same time period, only 100 cases of occupational asthma (OA) were reported in Hungary. According to international literature OR is three times more frequent than the OA. OR is not unknown in other European countries. According to European literature, the prevalence of OR among bakers ranges from 18 to 29%, and among workers with diisocyanate exposure (painters, urethane mould workers) ranges from 36 to 42%. Risk factors for OR include pre-existing atopy, high concentrations of the irritant agent and multiple irritant agents in the air of the workplace.
Conclusion: There is likely a marked underdiagnosis of OR in Hungary. Appropriate diagnosis requires a clinical and occupational history, and is enhanced by specific allergen or irritant respiratory challenge. There is a need for better surveillance of Hungarian workers for OR and subsequent assessment of these OR patients for OA. As management of OR requires environmental intervention such as increasing ventilation, decreasing the time of exposure, and substitution for the irritant agent, it is likely that failure to diagnose OR in Hungary is leading to progressive exposure of Hungarian workers to adverse occupational conditions perhaps leading to progression of OR to OA.

861
Characterisation of inflammatory cell asthma phenotypes in dental health care workers using cytokine profiling

Singh, T[1]; Bello, B[2]; Jeebhay, M[3]
[1]National Institute for Occupational Health, NHLS, Immunology, Johannesburg, South Africa; [2]National Institute for Occupational Health, NHLS, Epidemiology, Johannesburg, South Africa; [3]University of Cape Town, Centre for Occupational and Environmental Health Research, School of Public Health and Family Medicine, Cape Town, South Africa

Background: Cytokines play an important role in the inflammatory process of asthmatic airways. Inflammatory biomarkers can be useful in revealing diseased airways that are not easily detected using conventional parameters such as symptom experience, serial PEF, spirometry and NSBH. The aim of this study was firstly to identify subject groupings on the basis of common patterns in inflammatory responses. Secondly, to determine the relationship between these identified patterns and clinical asthma outcomes.
Method: A cross sectional study was conducted of 454 dental health care workers (HCWs) including dental students in five academic dental institutions in South Africa. A self-administered, modified ECRHS questionnaire eliciting the health and employment history was used. Subjects (n = 76) with self-reported doctor diagnosed asthma and/or asthma-like symptoms underwent spirometric testing and their sera analysed for atopy (Phadiatop), and 12 cytokines (IL-1β, 3, 4, 5, 6, 7, 8, 10, 12p70, eotaxin, GM-CSF, TNF-α). Data clustering and factor analysis was used to identify inflammatory cluster patterns of the cytokines. Multivariate logistic regression was used to determine the association between the cluster groupings of cytokine patterns in relation to relevant clinical health outcomes.
Result: The classification of asthma subtype based on cytokine patterns among asthmatic dental HCWs demonstrated both eosinophilic and neutrophilic inflammatory responses. Four phenotypically distinct subgroups relating to severity of inflammation (acute or chronic) of the cell types were finally identified. The most important determinants for the neutrophilic subtype were IL-1β, 6, 8, 10, 12p70 and TNF-α and for the eosinophilic subtype IL-3, 4, 5, 7, eotaxin and GM-CSF. The multivariate models showed a significant association between work-related chest symptoms and all four inflammatory patterns. However, a stronger association was observed for the acute neutrophilic response (OR = 4.99, 95% CI: 1.32–18.92).
Conclusion: This study demonstrated that neutrophilic inflammatory cell asthma phenotypes coexist with eosinophilic inflammatory phenotypes suggesting a possible dual pathway for asthma probably due to mixed asthmagenic exposures. These findings also demonstrate that factor analysis can provide a robust tool for characterising inflammatory cell asthma phenotypes when the mechanism for the asthma in certain occupational groups is unclear.

862
Identification of occupational allergens in coffee dust

Petersen, A[1]; Hohn, M[2]; Oldenburg, M[2]; Baur, X[2]; Jappe, U[2]
[1]Research Center Borstel, Clinical and Molecular Allergology, Borstel, Germany; [2]Germany

Background: Occupational exposure to coffee dust can cause severe health risks. In a cross-sectional study employees in a coffee haulage company (n = 24), a coffee silo (n = 19) and a decaffeinating company (n = 17) were mainly affected by erythematous skin lesions and rhinoconjunctival symptoms. The present study aimed at the identification of the culprit single coffee allergens.
Method: The sera of three workers with IgE antibodies to green coffee in the Immuno-CAP assay were used to identify IgE-reactive components in extracts obtained from the dust of the different coffee plant species *Coffea arabica* (from Brazil respective a mixture from different tropical countries) and *Coffea robusta* (from Vietnam). The dust was separated by SDS-PAGE and used for Western blotting. IgE-reactive protein bands were isolated and N-terminal protein sequencing was performed. For sequence homology screening Genebank and EST (expressed sequence tag) database were used.