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**679 Methodology for Predicting Air-Borne Pollen in Canadian Cities** *JL Van Adrichem, LL Coates, FA Coates, WH Yang* Aerobiology Research Laboratories, Nepean, Ontario, Canada

Access to tomorrow's pollen levels is important to allergy sufferers, clinicians and researchers. The air-borne pollen of ragweed, grass, maple, oak, birch and alder was collected daily in 12 Canadian cities over a six year period using rotation impaction sampling equipment set to collect one minute of every ten minute interval over a twenty-four hour period. These samples were analyzed to determine the number of particles per cubic meter of air sampled. The data for each species was combined with the weather data for each city and subjected to several statistical analyses including multiple linear regression, cluster analysis, and multivariate analysis to identify relationships within the data base and between the pollen counts and weather conditions. These relationships were used to establish predictive models. After development several models ran concurrently on a daily basis to provide predictive values for specific pollen. The resulting values were then compared to subsequent pollen counts using linear regression to determine which of these statistical models produced the most accurate predictions for each species. The predicted values for grass, ragweed and birch were highly correlated with subsequent pollen counts whereas none of the methods above produced equivalent correlations for either maple or poplar. Evaluation and modification of the models is performed daily as an ongoing operation of the laboratory. The triggers which determine the date for these species to commence pollination have yet to be further defined and quantitated and may represent a viable focus for future research.

**680 Causative Aeroallergens of Bronchial Asthma in a Desert Environment** *CI Ezeamuzie\*, S Al-Ali†, A Dowaisan†, M Khan†, Z Hijazi‡* \*Dept. of Pharmacology & Toxicology, Faculty of Medicine, Kuwait University, Kuwait †Al-Rasheed Allergy Center, Ministry of Public Health, Kuwait ‡Dept. of Pediatrics, Faculty of Medicine, Kuwait University, Kuwait

Kuwait is a desert country where the prevailing high temperatures, low humidity and scant vegetation might suggest a low prevalence of allergy. On the contrary, bronchial asthma and other allergic diseases are quite common. What is uncertain is the panel of causative allergens. This study investigated the sensitizing allergens in Kuwaiti patients with extrinsic asthma. A total of 553 asthmatics [male-female ratio: 1.4; mean age 31.7 years (range 3 - 76)] and 112 matched controls were studied. Sera from all patients/subjects were tested by the CAP-RAST method for specific IgE to 14 locally relevant inhalant allergens. Specific IgE to at least one allergen was detected in 87.2% of the patients compared with 24.1% in controls,  $p < 0.001$ . Seventy-one patients (12.8%) were negative to all 14 allergens. Among the sensitized patients, the sensitization rates were: pollens (87.1%), house dust (76.1%) and moulds (30.3%). The 4 most prevalent individual allergens were: *Chenopodium album* (70.7%), Bermuda grass (62.9%), *Prosopis juliflora* (62.7%) and cockroach (58.5%). Positivity to any of these 4 allergens was 90.0%. Allergens with intermediate prevalence were: Cajput tree pollen (42.9%), house dust mites (38.6%-46.5%), Eucalyptus tree pollen (42.9%), Date palm pollen (39.6%) and cat dander (30.9%). Moulds had the lowest rates: 14.1% for *Cladosporium*, 14.7% for *Alternaria* and 22.9% for *Aspergillus*. The 5 most sensitizing pol-

lens were from local horticultural plants which were imported and cultivated for the purpose of "greening" the desert. The relatively high prevalence of mite sensitization was surprising given the high humidity and moderate temperatures required for mite growth. It is likely that the extensive use of air conditioners in homes has created a conducive environment for mite growth. For all allergens, except the moulds, the prevalence rate was higher in males than females, but age had only a weak effect. Severe asthma occurred significantly more common among mould-sensitized patients. These results show that even in a desert environment, pollens and house dust allergens can be important sensitizing allergens. They also illustrate how practices that make the desert habitable (horticultural practices and air-conditioning) can impact on public health.

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**681 Cloning of Two Allergens From the Dust Mite *Lepidoglyphus destructor* Using Phage Display Technology** *Tove L. J. Eriksson, MSc\*, Omid Rasool, PhD\*, Paul Whitley, PhD†, Reto Cramer, PhD‡, M van Hage-Hamsten, Associate Professor\** \*Division of Clinical Immunology, Karolinska Institute and Hospital, Stockholm, Sweden †Department of Biology and Biochemistry, University of Bath, Bath, England ‡Swiss Institute for Allergy and Asthma Research, Davos, Switzerland

Dust mites are world wide a major cause of allergy. The dust mite *Lepidoglyphus destructor* is a common species in Europe and a predominant cause of storage mite allergy. At least 20 IgE binding components have, by SDS-PAGE and immunoblotting, been identified in *L. destructor* whole extract. Lep d 2 is one of the major allergens which has been cloned, sequenced and expressed earlier by us. In this study we constructed a cDNA phage display expression library in order to identify and clone additional allergens from *L. destructor*. Messenger RNA was isolated from live mites and cDNA was synthesised. After adaptor ligation, the cDNA was PCR amplified and cloned into the pJuFo phagemid vector. The construct was transformed into competent *E. coli* cells which thereafter were infected by the helper phage VCSM13 (Stratagene). The primary size of the pJuFo phage display library was  $1.4 \times 10^6$  independent clones. A modified biopanning, using human anti mouse IgG coated magnetic beads, mouse anti-human IgE monoclonal antibody and serum IgE from *L. destructor* sensitised subject, was used to select phages expressing IgE binding proteins. Five rounds of selection were performed to enrich the IgE binding phages. After the fifth round of selection, plasmid DNA from colonies containing pJuFo derived phagemides was prepared and the cDNA inserts sequenced. Sequence analysis showed homology with other known mite allergens from group 5 and 13. Lep d 5 was partially determined (327 bp) and showed 29% identity with Der p 5 from *Dermatophagoides pteronyssinus* and 43% identity with Blo t 5 from *Blomia tropicalis*. The mite group 5 allergens are major allergens but the function of the proteins are not yet known. The other allergen, Lep d 13, showed 78% identity with Blo t 13 and 60% identity to Aca s 13 (from *Acarus siro*) as well as high sequence homology to other fatty acid-binding proteins (FABP) from different species. Lep d 13 is 393 bp in length and the calculated MW is 14.7 kDa. There is a cytosolic FABP signature at position 6-24 in the predicted amino acid sequence and there are no potential N-glycosylation sites.

Both Lep d 5 and Lep d 13 will be expressed in *E. coli* and the IgE binding frequency among sensitised subjects determined.