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Abstracts

1. INFLUENCE OF METEOROLOGICAL CONDITIONS ON THE CONCENTRATION OF AIRBORNE FUNGAL FRAGMENTS. **E. Levetin**[1], C. Owens[1], H. Weaver[2], and W. E. Davis[2]. The University of Tulsa, Tulsa, OK[1] and Ochsner Clinic Foundation, New Orleans, LA[2]

Previous studies have shown the allergenic importance of fungal fragments; however, there is little information available on the day-to-day occurrence of these bioaerosols. This study examined the presence of fungal fragments in the atmosphere for two bioclimatically different locations, Tulsa, Oklahoma (TUL) and New Orleans, Louisiana (NO). The atmosphere in TUL and NO was monitored using Burkard spore traps from Aug 2007 through July 2008. Sampling and analysis were conducted using standard methods and results expressed as spores/m^3 or fragments/m^3. Meteorological data were obtained from NWS offices in TUL and NO. Fungal fragments were present in the atmosphere on 99% of the days in both cities. Concentrations in TUL were higher with a mean concentration of 257 and peak of 2326 fragments/m^3. In NO the mean was 81 and peak 965 fragments/m^3. Highest concentrations occurred in the fall and lowest in late winter and early spring. The ratio of spores to fragments was 29:1 in TUL and 77:1 in NO. Fragments showed a significant correlation ($p<0.05$) with total spores and many individual spore types. In TUL, fragments showed significant positive correlations with maximum, minimum, and average temperature, and significant negative correlations with rainfall and average wind speed. In NO significant positive correlation occurred with maximum temperature and negative with average wind speed and relative humidity.

2. THE AIR SPORA AROUND A COMPOST FACILITY: PHASE II. **N. Abel**, M. Buchheim, and E. Levetin. The University of Tulsa, Tulsa, Oklahoma

Previous studies showed elevated *Aspergillus fumigatus* levels 470 meters downwind of a large compost facility in northeast Oklahoma. The present study is being carried out to determine if the airborne *Aspergillus fumigatus* is genetically identical to compost isolates. Compost samples were dilution plated onto malt extract agar containing streptomycin and incubated at 45 C for 48 hours. *Aspergillus fu*migatus colonies were subcultured and then grown in malt extract broth; DNA was extracted using DNeasy kit. Air samples were collected from 5 random locations around the facility with Anderson single stage samplers onto MEA plus streptomycin plates. Control samples were collected from five locations in Tulsa, approximately 144 kilometers upwind of the compost site. Plates were incubated at 45 C and all *A. fumigatus* isolates were subcultured as above. The DNA from isolates was amplified and fingerprinted using STRAf2Aforward Custom Oligo kit. Phylogenetic analysis of the ITS-1 gene from the C1S1 (compost) isolate confirmed our identification as *Aspergillus fumigatus*. Furthermore, the C1S1 isolate exhibits up to 3 nucleotide substitutions relative to other published sequences from *Aspergillus fumigatus*. Preliminary microsatellite analyses indicated that variability exists across a sampling of compost and air samples. Results from a more complete microsatellite analysis will be discussed.

3. DRIVING WITH GERMS: A POTENTIAL RISK FACTOR FOR ASTHMATICS. **Dhar Minati G[1]**, Dhar Aveek[2], Portnoy Jay[1] and Barnes Charles[1]. [1]Children's Mercy Hospital, Kansas City, Missouri,. [2]Shawnee Mission East High School, Kansas

The purpose of this study is to determine the relative amount of fungal allergen in household automobiles. In this study, the relative number of mold allergens was determined since mold spores can trigger respiratory allergic reactions such as asthma and other respiratory distress. From a clinical stand point, four major groups of molds, Phycomycetes, Ascomycetes, Basidomycetes, and Dueteromycetes are important as potential allergens. A high amount of mold allergen inside an automobile can be a potential risk factor for Allergic/ Asthmatic kids and adults who spend many hours in the car commuting to different places. The hypothesis of this study was that if inside of the automobile can be cleaned on a regular basis then there will be a lesser chance of the development of allergens inside the automobiles. In this study, several variables were considered including different areas of the vehicles such as dashboard, steering and radio, inner door handles and seat-belts, and seats. Swipes were taken in these areas, and then smeared on malt extract agar plates following incubation at room temperature for 72 hours. After incubation, the microorganisms developed in the plates were transferred onto microscopic slides and stained to identify different kinds of molds and the concentrations were counted under a microscope. The results suggested that mold concentrations are at maximum in the dashboards and the seats of the automobiles. The cars which were cleaned on a regular basis showed a less concentrations of molds. This study concluded that presumably a cleaned automobile has a lesser chance of respiratory allergens due to less mold concentration inside.

4. SIGNIFICANTLY DIFFERENT ANALYTICAL RESULTS FROM THREE US, AIHA ACCREDITED LABORATORIES FOR *LEGIONELLA* ANALYSIS OF SPLIT POTABLE WATER SAMPLES; IMPLICATIONS TO PUBLIC HEALTH; A CASE STUDY. **R. Garrison.** IAQ Consultants, Inc., Southlake, Texas

Twenty six (26) water samples were collected in order to determine the potential presence of viable *Legionella* bacteria in the potable water system of a high security government facility, a neighboring facility and from fire hydrants proximate to the facility. The overall objective of the water quality study was to compare the *Legionella* results from three (3) separate, AIHA accredited laboratories in the US. The laboratory results for *Legionella* from Laboratories 1 and 2 yielded reasonable agreement for all twenty six (26) split water samples. Laboratories 1 and 2 only recovered less than 2 CFU/ml of viable *L. pneumophila* in two (2) of the twenty six (26) split water samples. Lab 3 results indicated viable *Legionella bozemanii* in twenty three (23) of the twenty six (26) split water samples submitted. Viable *L. bozemanii* recoveries ranged from 1 CFU/ml to 560 CFU/ml as reported by Laboratory 3. The risk of building occupants contracting legionellosis from pathogenic *Legionella* was minimal based Laboratories 1 and 2 results; however, the risk of legionellosis was significant based on Laboratory 3 results. The conflicting quantitative analysis in *Legionella* results between the AIHA accredited laboratories directly affected the consulting aerobiologist's ability to provide reasonable, cost effective remedial recommendations.

5. STUDIES ON AIRBORNE FUNGAL SPORES FROM EIGHT HISTORICAL AND MODERN BUILDINGS IN AMHERST, MA. **M. Joy**, M.L. Muilenberg, and C.A. Rogers. University of Massachusetts, Amherst, MA, USA

Historical buildings are more than simple functional spaces where we live and work; they are an important part of our cultural heritage. While generally considered to be of higher quality construction than modern buildings, historic structures are subject to natural deterioration simply because of their longevity or to disrepair as a result of lack of proper maintenance over time.

Eight buildings, four historical (1906-1910) and four modern (1985-1997), were surveyed to establish background spore concentration estimates and profiles for structures of similar historic age and architecture. The frequency of appearance and concentration of airborne fungal spores from the buildings were measured using Allergenco spore trap cassettes. A minimum of four rooms on each floor were sampled in each building. Direct microscopic examination of the whole trace was used to analyze each sample.

A total of 23 spore taxa were recorded from the historical buildings and 20 from the modern. In both the historical and modern buildings a relatively high frequency of *Cladosporium*, *Penicillium/Aspergillus*-like, and some unidentified mitospores were recorded. Total spore counts for the historical and modern buildings were compared resulting in a statistical difference p-value 0.0541 with means 111.62+/-20.887 and 70.065+/-4.2968 respectively. These results will provide steady state concentrations for historical buildings to aide in the future assessment of moisture related damage, use, and occupancy.

6. FURNACE FILTER SAMPLES TO SCREEN FOR ABNORMAL MOLD CONDITIONS. **S Flappan** Flappan Consulting, Inc.**.** Overland Park, Kansas 66213

Oftentimes, a homebuyer wants to know if there is a significant mold problem in a house before purchasing it. The seller may forget to disclose or be unaware of past water incidents. Inspecting the premises for warped floors, stained ceilings/walls, peeling wallpaper, or visible discolorations can reveal prior or current moisture problems. Taking a few random representative air samples in the house can give a snapshot view of what is present in a few limited areas of the residence. These measures are useful but can easily miss hidden mold problems behind walls, under carpet, above ceilings, in ducts, etc. Or a significant problem on sheetrock walls may have been washed away and painted over (covered up).
A reliable screening tool is to take a surface sample from the currently used furnace filter to check for abnormalities. Data and examples will be presented.

7. FUNGAL CONCENTRATIONS IN FIRST NATIONS HOMES, JAMES BAY, ONTARIO **Mike Muilenberg**[1], C Rogers[1], and S Davies[2] Aerobiology Instruction and Research, LLC, Amherst, MA, USA[1], MouldClean, Lively, ON, Canada[2].

Two Canadian First Nations communities are situated in low lying areas near a river outlet on James Bay, Ontario. Spring flooding, infrastructure issues, sub-par housing, in addition to social challenges, contribute to fungal concentrations considered to be elevated by some published guidelines.

In Oct '07, 177 Allergenco-D airborne fungal samples were collected in 134 homes in Community A; 2 samples were collected in homes with basements. Samples were collected ~1 m above the floor in the main living area of each home. In Dec '07 and Jan '08, 48 homes (59 samples) were collected in Community B. Recoveries were analyzed from at least 25% of each particle deposit, using 400X magnification.

Outdoor median total spore concentration was 720 spores/m^3 in A and 33 in B. Indoor median concentration in A was 1,400 sp/m^3 (min/max; 99/1.36x10^6), and in B, 900 sp/m^3 (min/max; 27/4.98x10^5). In Community A, over 50% of the homes were above Baxter's (JOEH 2005) potentially moldy threshold of 1300 sp/m^3 (San Diego homes); in Community B, less than 40% were above this threshold. Median Pen/Asp concentration Community A was 594 (min/max; 0/5x1.36x10^6) and in B, 198 (min/max; 0/4.97x10^5). About 40% of the homes in both communities were above Baxter's Pen/Asp threshold of 900/m^3.
Fungal spore concentrations in First Nation housing are higher than in most homes in the U.S. and Canada. The possibility that this situation might result in adverse respiratory outcomes should be investigated.

8. PREVALENCE OF IGE REACTIVITIES TO AIRBORNE PARTICULATE BY ASTHMATIC SUBJECTS.
Félix E. Rivera Mariani, B.S., Benjamín Bolaños, PhD. Department of Microbiology, School of Medicine, University of Puerto Rico – Medical Sciences Campus

The atmosphere contains many biological and non-biological airborne particulate. Many studies have found that exposure to high concentrations of these, such as fungi, pollen, and inorganic particles can induce episodes of asthma. However, there few studies describing to what airborne particles asthmatic patients may be reacting to. The purpose of this study was to examine the IgE antibody reactivity of serum from asthmatic patients against air samples.

A piece of Fungi TapeTM, with the adhesive side facing upward, was attached to a microscope slides and placed in the Allergenco ® (MK3) spore trap to capture 24-hr air samples with 10 minutes of air sampling and 110 minutes of relapse. Every 24-hr the slide with the piece of Fungi Tape was exchanged for a clean one for the period of one week. 10-cm squares of Mixed Cellulose Ester (MCE) protein-binding membranes (PBM) were sealed with pieces of the Fungi Tape with the collected air sample. Sera from 12 asthmatic subjects and one control (with no skin reactivity to allergens nor signs of asthma) were collected and their reactivities against air samples examined by the Halogen Immunoassay. From the processed MCE-PBM, the total particles collected by the air sampler and the haloes of IgE reactivity were counted and the reactive particle described.

From the total sera collected, 10 of the 13 (77%) sera reacted to the air sample. Most of the particles with haloes were not able to be identified; however, 9 of the 13 sera (69%) reacted to ascospores and 6 of 13 (46%) to basidiospores. There was no correlation between total particulate and the amount of haloes of IgE reactivity, haloes to ascospores , or haloes to basidiospores; however, there was a correlation between total haloes and haloes corresponding to ascospores and basidiospores.This study suggests that the reactivity of asthmatic patients to airborne particles may be dependent more on the type of particle rather than the concentration of particles to which it is exposed.

9. AIRBORNE RAGWEED ALLERGEN DETECTION USING MULTIPLEX ARRAY. **Charles Barnes**, A Heisler, F Pacheco, J Portnoy, Children's Mercy Hospital, Kansas City, MO

Current visual techniques for approximating airborne allergen levels are labor intensive and may miss allergen carried on unrecognizable particles. We investigated a process measuring multiple outdoor allergen levels simultaneously using multiplex array technology. Carboxylated fluorescent microspheres were purchased from Luminex Corporation and coupled to rabbit anti ragweed antibodies obtained from Greer Laboratories. Air samples were collected from August through September using an Omni 3000 spinning concentrator. Anti Ragweed antibody coupled microspheres, along with biotinylated anti ragweed antibody was used to determine the relative amount of ragweed present in airborne samples using a detection system manufactured by the Luminex Corporation.
Ragweed concentrations at noon were determined for 45 different days. Airborne concentrations ranged up to 671 ng/M^3 and airborne ragweed counts ranged up to 566 grains for a 20 minute collection. Concentrations determined from the Luminex analyzer were consistent with results obtained from visual enumeration of airborne ragweed pollen grains. Additionally, the immunoassay results displayed early high airborne allergen levels not shown in the traditional method of allergen detection.
Development of the ragweed immunoassay on the multiplex array demonstrates that simultaneous detection of multiple airborne allergen levels is feasible.

10. EFFECT OF ORIFICE SIZE ON SAMPLER EFFICIENCY WITH THE BURKARD SPORE TRAP. **M. Cullen** and E. Levetin. The University of Tulsa, Tulsa, OK

The Burkard 7 day spore trap has been utilized extensively to sample bioaerosols. It can be fitted with a standard or an alternate orifice, which has a higher efficiency for collecting small spores. However, a preliminary study from our lab found that the concentrations of some spore types were significantly higher with the standard orifice and others with the alternate orifice. The present study was undertaken to expand the previous comparison for a one year period. The atmosphere was monitored with two Burkard spore traps; both placed on the roof of Oliphant Hall and fitted with the different orifices. Analysis of all common airborne spores was performed microscopically at 1000X, and raw spore counts were converted into daily concentrations and expressed as spores/m^3. Concentrations were log transformed and analyzed statistically using repeated measures MANOVA and T-tests. MANOVA analysis of 3 months showed that there were significant differences among spore types, no significant difference between orifice types, and a significant interaction between orifice and spore type. Further analysis showed the mean concentrations of ascospores and *Penicillium/Aspergillus* spores were significantly higher with the alternate orifice, while the concentration of *Alternaria* was significantly higher with the standard orifice. Completion of a full year's analysis is needed in order to verify the results of the preliminary study.

11. **HYPERSENSITIVITY PNEUMONITIS (HP): FIFTEEN CASES CAUSED BY RESIDENTIAL BIOAEROSOL EXPOSURES**. **Jeffrey C. May, M.A.** May Indoor Air Investigations LLC, Tyngsborough, MA 01879

In 15 homes where occupants had physician-diagnosed hypersensitivity pneumonitis (HP), we determined some of the bioaerosol present. Occupants were referred by pulmonologists, a nurse and an allergist. Two-thirds of the residential HP cases were females. Air and dust samples were obtained from mechanical equipment and various areas of the home. In two cases, cockatiels were present and in four cases, feather bedding or cushions. Home exposures to bioaerosols included *Aspergillus* and *Penicillium* spp., pet and wool dander, and insect fecal material. Remediation included removal of all feather products and pet birds, cleaning of basements and mechanical systems, elimination of contaminated carpeting and furniture, and installation of allergen encasings on pillows and mattresses. After remediation,12 out of 15 HP cases experienced dramatic improvements. One case refused to give up a mold-contaminated feather pillow, another did not remove carpeting, and a third continued to suffer due to as yet unknown causes.

12. RAGWEED POLLEN IN THE AIR OF KANSAS CITY – A 10 YEAR HISTORICAL PROSPECTIVE. BARNES, Charles DHAR, Minati, PORTNOY, Jay Children's Mercy Hospital, Kansas City, Missouri.

Ragweed pollen is the major fall aeroallergen in the Midwestern United States. Pollination begins as the days begin to shorten in late summer and terminates around the second week of October. To study the impact of weather parameters on airborne ragweed pollen concentrations we conducted the following studies.
Ragweed pollen was collected using a Hirst style spore trap on the roof of a 5 story building in Kansas City, MO. Slides were stained with Calberlas stain and pollen grains were enumerated microscopically every 4 hours from August 1 to October 31 during 10 successive ragweed seasons. Weather parameters including outdoor temperature were recorded hourly on an Automated Weather Station. Both sets of data were entered into an Access database for analysis.
The highest daily mean ragweed concentration was 814/M^3. And the highest mean airborne concentration for any 4 hour period was 2040/M^3. Ragweed pollen counts were diminished at temperatures lower than 56°F and at temperatures greater than 94°F. Geometric mean ragweed count on sunny days (> 50% maximum sun) was 3.14 and on cloudy days was 1.84 ($p < 0.001$). Geometric mean ragweed count on rainy days (> 1.0 inches rain) was 2.52 and on dry days 2.84 (NS).
Ragweed is a sun loving plant that thrives in areas of the country with warm temperatures and abundant sunshine during the pollination season.

13. INTERNATIONAL COLLABORATION FOR FORECASTING ALLERGENIC POLLEN – ICAP. **A. Ariatti**[1], S. Isard[1], R. Gehrig[2], and J. Russo[3] . [1]Pennsylvania State University, University Park, PA, USA, [2]MeteoSwiss, Zürich, Switzerland, and [3]ZedX Inc., Bellefonte, PA, USA

The International Collaboration for Forecasting Allergenic Pollen is an initiative that has emerged from the Pan-American Aerobiology Association Symposium held at Pennsylvania State University in 2007. At the symposium, a ragweed discussion group met with the goal of promoting an international collaboration for the creation of an Internet-based platform to forecast aerial concentration of ragweed pollen in North America and Europe.
Following the meeting, a restricted access pilot website was developed and has been operative in 2007 and 2008. Over the two growing seasons, phenological observations for ragweed (*A. artimisiifolia, A. Psylostachia, A. trifida*) have been collected from five countries in Europe (France, Hungary, Italy, Switzerland, Serbia), five states in United States (California, Florida, Pennsylvania, Massachusetts, Missouri), and two provinces in Canada (Ontario and Quebec) .

In the framework of ICAP, a set of short term and long term objectives was delineated. The short term objectives focus on the development of a restricted access platform interface to access temporally and geographically referenced aerobiological, phenological, and land cover databases for the major allergenic pollen producing plants for North America and Europe. Registered participants have tools for data input, datasets mining, maps and models displays. Phenological maps and weather model outputs will be available for the specialists and pollen forecasters.

The long term objectives include the development of a public platform interface to access pollen forecasts for selected allergenic pollen. The pollen types initially included are ragweed, grasses, oak, and cedar for North America and ragweed, grasses, birch, and olive for Europe. The pollen forecasts provided on the public website will be based on simulations of both the local development of pollen-producing plants and the long-range transport of pollen. Online tools will enable a direct interaction with the allergic sufferers for the collection of allergy symptom data and additional generic information about pollen biology, interpretations of the pollen simulations, and reported symptoms in map and graphical form will be made available.

Since the beginning of 2009 and thanks to the continued support of ZedX Inc. - a private IT company collaborating with CEAL (Computational Epidemiology and Aerobiology Laboratory) at the Pennsylvania State University - an upgrading of the pilot website is underway, in order to meet the short term goals.

14. ANALYTICAL PRECISION OF COMMERCIALLY AVAILABLE SPORE TRAP SERVICES. **Robertson, L.D.**[1], PhD, CIAQP, CIEC, CMRS. Brandys, R.[2], PhD, MPH, PE, CIH, CSP, CMR. [1]Indoor Environmental Consultants, Inc., Austin, Texas 78735. [2]Occupational and Environmental Health Consulting Services, Inc. , Hinsdale, Illinois, 60521

The general absence of "actual" fungal bioaerosol data in common environmental settings precludes the ability to evaluate the accuracy of spore trap analytical methods. Therefore, precision, or the comparison between values within a set of data, offers the only means by which the reliability of environmental sourced spore trap data can be determined. This study evaluates the precision of spore trap analyses of seven (7) commercial laboratories currently providing spore trap analytical services in the United States. Each laboratory had been deemed proficient in the identification of fungal spores in the Environmental Microbiology Proficiency Analytical Testing (EMPAT) program offered through the American Industrial Hygiene Association (AIHA). Simultaneous air samples were collected on four (4) commercially available spore traps (Cyclex-D, Allergenco D, Micro 5, Air-O-Cell) in four (4) different and individual collection conditions. In the opinion of the authors, the analysis of spore trap samples are ultimately subjective decisions based on the individual skill and experience of each microscopist, which may not lend itself to traditional statisical analysis of random environmental samples. However, given that precept, traditional statistical analysis reveal data that seriously question the validity of spore trap science as it exists to date. Extreme variability was observed in all data. Only a 56% precision was observed in the identification of total fungal spores, with individual fungal spore categories well below this value. The results also revealed questions about the validity of individual fungal spore categories that are traditionally utilized in current commercial spore trap services. The authors conclude that the results of current spore trap analysis are highly variable that should not be utilized as a sole method in assessing fungal spore concentrations and populations until improved analytical precision can be demonstrated for this methodology and documented with each analysis.