Integrated Healthy Homes Assessment and Intervention for Children in Memphis

Presented by:
Pratik Banerjee
Assistant Professor
Division of Epidemiology, Biostatistics, and Environmental Health
School of Public Health
University of Memphis
The Problem

• Two pediatric health issues
  • Allergic asthma and Lead poisoning

Poor housing
The Problem

• Memphis has one of the highest percentages of substandard housing in the nation

• Memphis was named as the nation’s top 3 “Asthma Capital” for five consecutive years (2011 to 2015)

Mold exposure causes asthmatic and respiratory reactions in children and infants
Why bother about molds?

- Uniquely adaptable to temperature variances, and environmental terrains
- Does cause respiratory diseases and certain species can have toxic effects in humans, especially the vulnerable population
- Respiratory diseases/illnesses include but not limited to...Asthma, Allergies/Allergic Rhinitis, Idiopathic Pulmonary Haemosiderosis, Chronic Obstructive Pulmonary Disease, Eye irritation, Cancer, Sudden Infant Death Syndrome, etc...
- Some pathogenic species include: *Alternaria alternate*, *Aspergillus fumigatus*, *Cladosporium herbarum*, *Penicillium spp.*, *Stachybotrys chartarum*
Justification

• Mold exposure does not necessarily have to be from the visible mold; exposure can also come from the invisible mold such as microscopic mold spores (Reponen et al., 2010)
• Developing a quantitative method to assess the burden of indoor mold and bacteria exposure
Specific Objective

• To evaluate the diversity and burden of major bacteria and mold genera in indoor dust samples collected from homes with or without visual molds in Memphis metropolitan area.

Technological Innovation

We have developed a “one stop” Environmental Monitoring Platform

• Rapid detection of multiple Indoor Environmental Quality (IEQ) parameters simultaneously
Methods - Field Sampling

- Dust samples from total 20 households
  - 10 homes with visible mold
  - 10 homes with NO visible mold
- 1 week sampling period in each home
- Home walkthrough questionnaire survey was administered
Methods - **Post Field Sampling**

**Molecular Detection and Next-Generation Sequencing (NGS)**

- **Samples Collected from Homes and stored in 4°C**
- **Extraction of DNA (Quigen DNA PowerSoil kit)**
- **Purification of DNA (Epigentek DNA Concentrator kit)**
- **PCR content cleanup (GenElute™ PCR Clean-Up Kit)**
- **Gel Electrophoresis of PCR content**
- **Endpoint PCR using ITS and 16S (universal mold and bacteria detection) primers**
- **Additional DNA concentrating (Quigen DNA PowerSoil kit)**
- **PCR content cleanup**
- **Gel Electrophoresis of PCR content**
- **Endpoint PCR using ITS and 16S (universal mold and bacteria detection) primers**
- **Additional DNA concentrating (Quigen DNA PowerSoil kit)**
- **Final DNA product sent for Amplicon Sequencing (Eurofins)**
- **Gene targets for NGS using Illumina MiSeq Platform**
Results - **PCR ITS (molds)**

- **DNA ladder**
  - Homes with visible mold

- **NTC**

- Homes with NO visible mold
Results - NGS

Diversity of major bacteria and mold genera in indoor dust

Panels A and B show relative abundance of most common bacterial genera. While panels C and D represent the relative abundance of major mold genera. Microbial diversity was estimated by high throughput genetic sequencing techniques targeting bacteria and mold-specific genes from DNA samples extracted directly from dust.
Results - **NGS**

**Concentration of mold species known to cause respiratory diseases**

The heatmap shows concentrations of major respiratory disease causing molds found in dust samples. Mold concentration was estimated by high throughput genetic sequencing techniques targeting mold-specific internal transcribed spacer (ITS) genes from DNA samples extracted directly from dust.
Summary and Conclusions

- Pathogenic molds such as *Stachybotrys spp.* (black mold), *Alternaria alternate*, *Aspergillus niger*, *Chaetomium globosum*, *Cladosporium sphaerospermum* were detected in both types of homes.

- Molecular techniques can reveal mold contamination in homes which the conventional visual inspections can not detect.

- Continuous mold monitoring using sensitive and accurate methods (such as sequencing or PCR) that can detect potential mold contamination in homes regardless of the visual inspections.
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