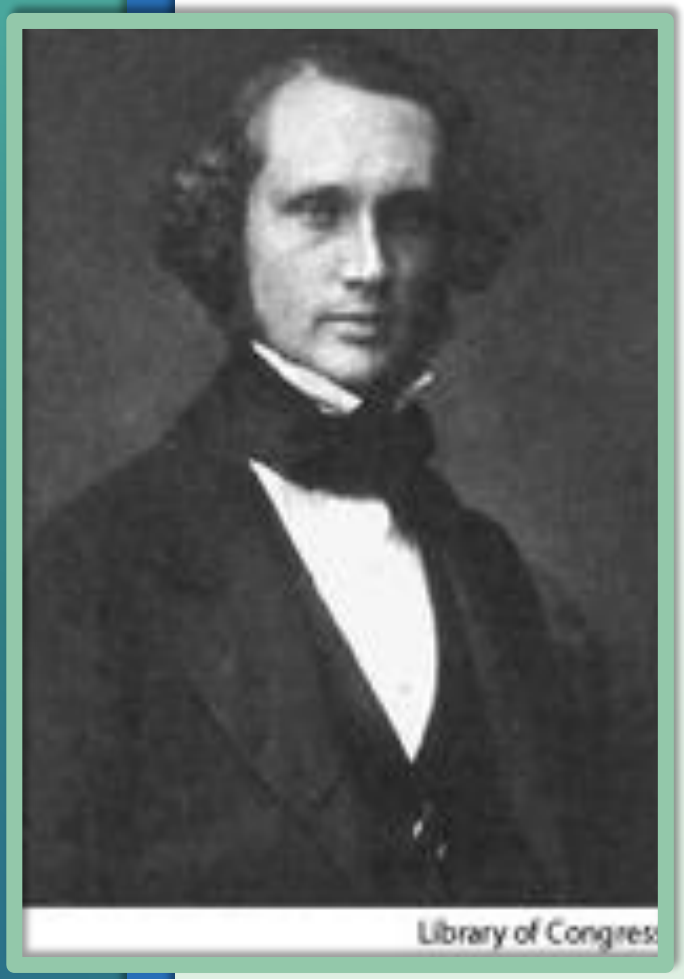
United States Environmental Protection Agency

Quantifying fungi/mold in the epidemiology of asthma and chronic rhinosinusitis

Steve Vesper

The U.S. Environmental Protection Agency (EPA) through its Office of Research and Development collaborated in the research described here. Although this work was reviewed by EPA, it may not necessarily reflect official EPA policy. Mention of trade names or commercial products does not constitute endorsement or

William Thomson, 1st Baron Kelvin



"When you can measure what you are speaking about, and express it in numbers, you know something about it, when you cannot express it in numbers, your knowledge is of a meager and unsatisfactory kind; it may be the beginning of knowledge, but you have scarcely, in your thoughts, advanced to the stage of science."

Traditional Mold Analysis

1. Visual and Olfactory Inspection

- Assumes you can see or smell the mold
- Varies depending on the skill of the investigator
- Mold is not always obvious; might be hidden in walls (as shown)
- Not quantitative

Traditional Mold Analysis

2. Microscopic Counting

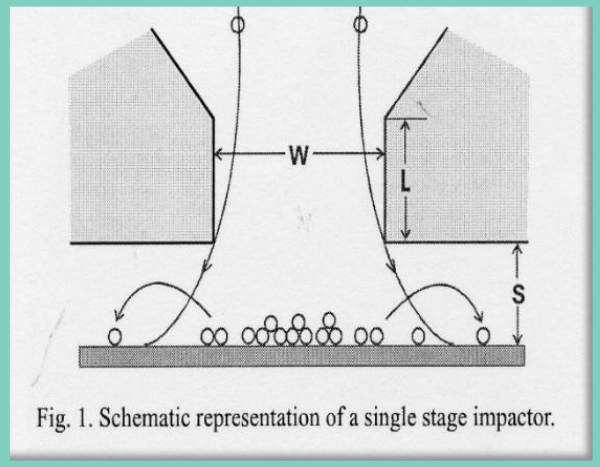
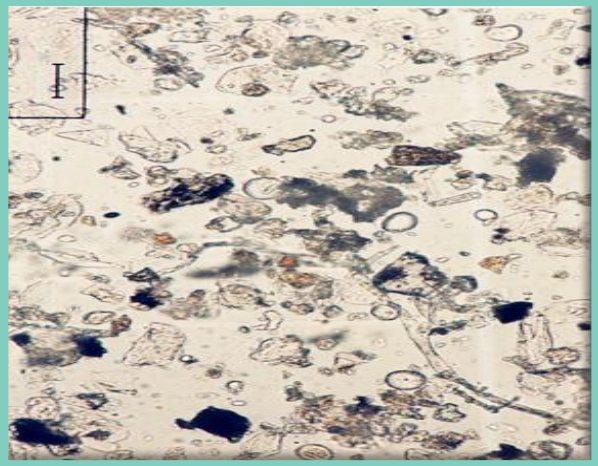


Fig. 1. Schematic representation of a single stage impactor.



- **Collection Problems**
  No standardization; short sampling times; each sampler has issues like "impact bounce", slit size etc.

- **Microscopic ID Problems**
  Most spores can't be identified to species: limited by analyst
  Example- Penicillium and Aspergillus (Pen/Asp)

Traditional Mold Analysis

3. Culturing

- Not all molds grow on the same medium.
- Plates can be overgrown; limited to short samples.
- Requires significant expertise to identify molds.
- Quantifies viable cells only; but "dead" cells may still be allergenic.

WHO- Guidelines for Indoor Air Quality: Dampness and Mold (2009)

Assessment

- Occupants of moldy buildings are at increased health risk
- Exposures to mold should be “avoided or minimized”

Need: Data Interpretation

HUD- Report to Congress (2005)

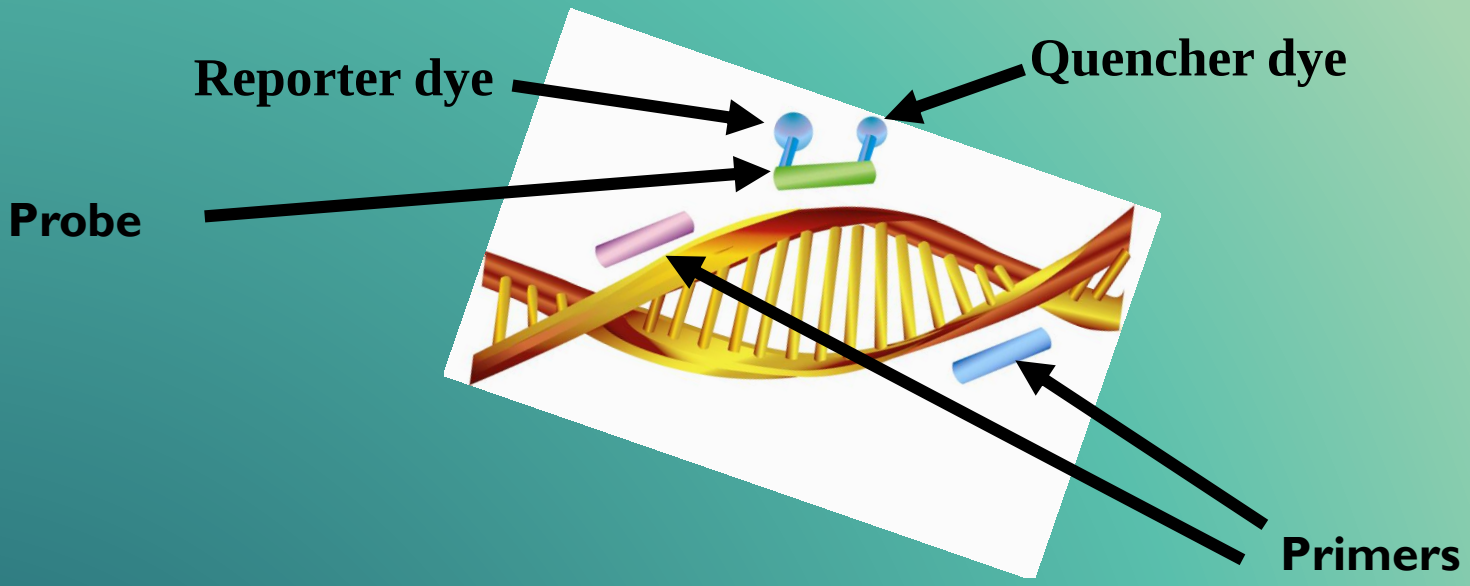
"Another problem is the difficulty in interpreting test results, since mold spores are ubiquitous and there is no consensus among experts regarding what constitutes acceptable indoor spore concentrations in indoor air or house dust, or which species are most problematic."

Need: New method

IOM- DAMP INDOOR SPACES AND HEALTH (2004)

"Committee identified need to developed improved exposure assessment methods… for specific microorganisms that used DNA-based and other technology…"

Development of Quantitative PCR (QPCR)



Sequence Detector instrument monitors the PCR reaction

American Healthy Home Survey -2006

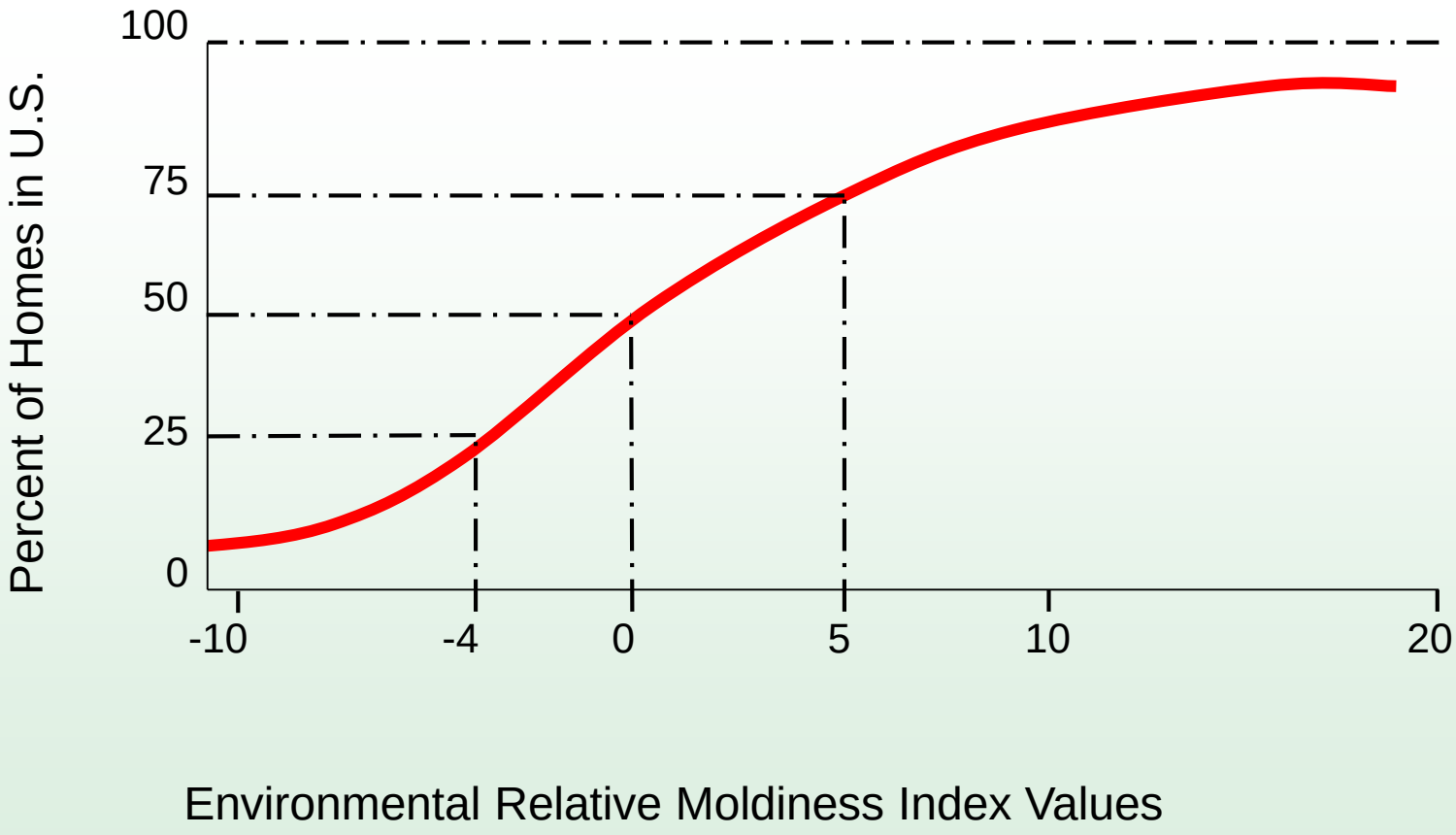
- HUD utilized a standard settled-dust sample – composite living room plus bedroom.
- Cannot measure all molds but we sought an "indicator" set of molds that would allow us to quantify mold exposures nationwide and develop an Environmental Relative Moldiness Index for US homes.

Development of the Environmental Relative Moldiness Index (ERMI)

Steps:

1. Hud collected dust samples from a nationally representative 1100 homes
2. Quantify 82 species using qPCR in 5 mg of sieved dust from each sample
3. Identified 36 “indicator” species from the 82
4. Divided 36 into 26 indicators of water-damage (Group 1) and 10 common to all homes (Group 2)
5. Mathematical calculation used to define ERMI

ERMI Scale


Percent of Homes in U.S.
100
75
50
25
0
-10
-4
0
5
10
20
Environmental Relative Moldiness Index Values

ERMI Method Assessments

Lessons from the AHHS

- Every home in the AHHS had some level of mold
- 50% of the time occupants were unaware of and inspectors failed to find mold problems, i.e., homes had ERMI value >5
- Indicator molds had no geographic bias

ERMI and Asthma Studies

Prospective Study in Cincinnati-

Results

- Only the home's ERMI value correlated with the development of asthma
- The adjusted relative risk for a 10-unit increase in ERMI value was 1.8 (95% CI, 1.5-2.2).
- Three specific Group 1 molds linked to development of asthma but no Group 2 molds

All ERMI and Asthma Studies

	Mean	SE
Asthma	7.97	0.33
Control	3.57	0.41
Difference	4.40	0.53
p-value		<0.001

Limitations

- Small number of studies which used ERMI
- Most tesing done in US; others- in Scotland, ERMI was relevant to asthma but not in Finland
- Can’t quantify all fungi
- Expensive compared to spore trap/culture

Current Studies

1. Assessment of mold exposures in difficult-to-treat asthma cases
2. Intervention studies in schools using HEPA filtration
3. Assessment of home after flooding or hurricanes
4. 2018 American Healthy Homes Survey

QPCR Analysis in CRS

Goal

To measure the populations of 36 fungi in the homes and sinuses of chronic rhinosinusitis (CRS) and non-CRS patients.

Methods

Samples from CRS (n = 73) and non-CRS patients (n = 16) using qPCR. Single-blind cross-sectional study.

Results

- Seven fungi discovered at very high concentrations in some CRS patients.
- Four CRS patients with marked elevations of fungal populations in their sinus samples underwent endoscopic sinus surgery.

Conclusions

- Seven fungi were found in very high concentrations in the sinuses of some CRS patients.
- Surgical treatment reduced by several orders of magnitude.

Our Goal

We hope to someday have enough data on the relationship between ERMI values and asthma that physicians, parents and patients can use the information to reduce asthma's prevalence and symptoms.