

Interactions between indoor environmental conditions and indoor microbial ecology

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**Environment and Health 2013:
Bridging South North East and West
Basel, Switzerland
19-23 August 2013**

Acknowledgments

- Shela Ray, Research Associate, and Gina Bendy, Assistant, Building Ecology Research Group
- Jonathan A. Eisen (P.I.) and David Coil, UC Davis, microBEnet grant team
- Janet Macher, California Department of Public Health
- Christopher Petty, database consultant
- Funding by the Alfred P. Sloan Foundation program: Microbiology of the Built Environment

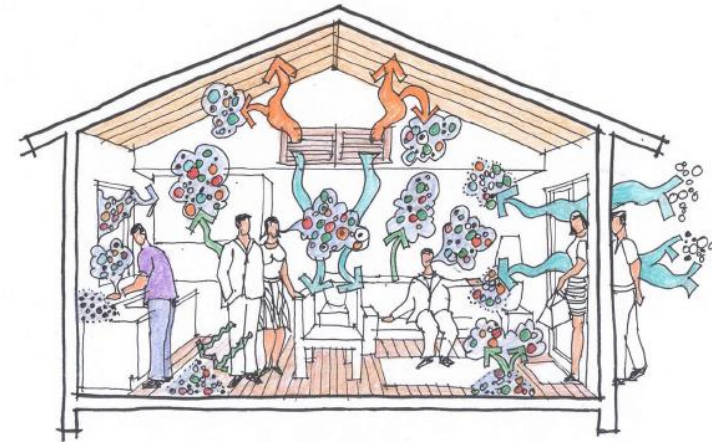
Aims and Methods of this Paper

Aims:

- Ascertain key environmental variables relevant to indoor microbiome
- Develop literature database: association of ind env – microbiome
- Guidance: minimize risks of exposure to health-relevant organisms.

Methods:

- A selective review of peer reviewed literature was used to create database of over 370 articles. Still growing
- The database enables identification of dominant patterns of associations between reported environmental factors and organisms by level of identification.
- Emphasis is placed on organisms by frequency of occurrence and/or health relevance.



Results

- Database of publications connecting indoor microbiome and indoor environmental conditions: now >370 (and growing)
- DB soon to be posted in SQL format on <http://microbe.net>
- Accessible to all, no cost. (Awaiting approval -extension of grant)
- Confirmed accepted beliefs (based on prior indoor air research):
 - Temperature
 - Humidity
 - pH
 - Ventilation,
 - Sources (including people).
- New hypotheses:
 - Humans and especially human skin as important source
 - Communities may be more important than individual organisms or diversity *per se* (Quorum Sensing – chemical communication among microbes in biofilms)
 - Need for more recognition and assessment of role of communities
 - Need to understand unidentified complex surface films (UCSF)

Background

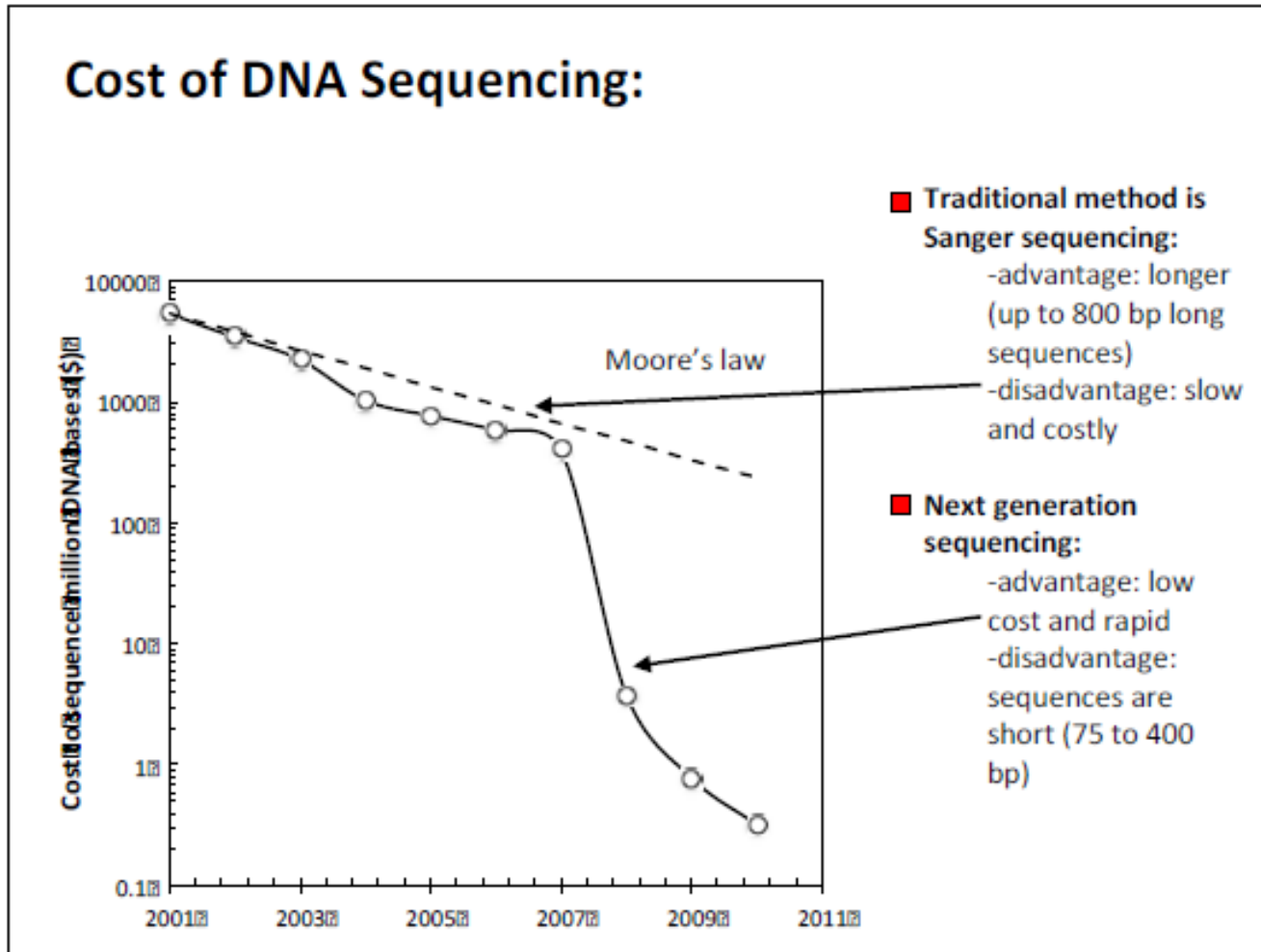
- Sloan Foundation Indoor Environment program funding important research on microbiology of built environment,
- Gene sequencing samples from indoor environment by many of the best-known microbial ecologists in America
- Culture-independent microbiology: huge increase in number of organisms found
- Identification not always at species or even genus level
- Building Ecology Research Group role: bring building science to microBEnet project – networking microbial ecologists and building scientists
- Building Ecology: an architect's perspective on building and environment
 - Factors often of little interest to M.E. researchers have important outcome impacts.
 - Always looking for mechanisms and connections or patterns.
 - Finding more connections than mechanisms.

Microbial ecology studies of Indoor microbiome

- Microbial ecologists: background in studies of natural environment – oceans, forests, and wilderness (samples collected from air, soils, and objects).
- Framework of ecology, interest is in “who is there” and “what are they doing.”
- Results are usually expressed in terms of relative abundance of various organisms. Diversity is regarded as an indicator of a health ecosystem.
- Developments in gene sequencing analysis has multiplied number of identified micro-organisms by 10x compared to a decade or two ago, primarily by culture-dependent analytical methods.
- Sample collection from surfaces is relatively simple: swabs samples quickly collected, easily stored/transported to laboratory.
- Since gene-sequenced based analysis can used for living or non-living samples, analysis results in a large number of organisms being identifiable.
- Also during the past few years, analytical costs have dropped dramatically to far more affordable levels.
- Acquisition of a large amount of data relatively easily and quickly is the basis of very rapid growth in the use of gene sequencing .
- Interpretation of results is still an evolving and developing field.

Cost of DNA Sequencing:

Dropped precipitously in the past decade, especially during the past six years (source: Jordan Peccia)



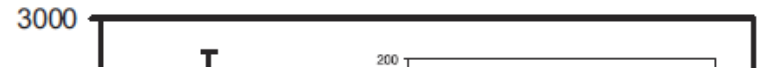
Hypotheses formation: Building ecology perspective of findings in the literature review

- Key factors are inter-dependent: Temperature, humidity, occupancy, ventilation, time – because of the way building environments are used, and how they are ventilated and thermally-controlled
- Communities of microbes dominate evolution, *e.g.*, “quorum sensing,” (bacteria growing on fungi found on wet wood, infectious, toxic or pathogenic doses)
- Long dormancy (“stationary phase”) important for many organisms
- Human-indoor environment interactions important: source, perturbation, modifier of conditions
- Indoor environmental surfaces covered with “undefined complex surface films” (UCSF)
- Indoor microbial ecology” needs to evolve to apply ecosystems concepts and building science to the built environment:
- Buildings are ecosystems, understanding key factors and interactions critical to knowing what and how to sample.

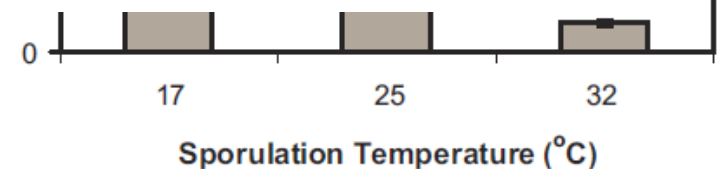
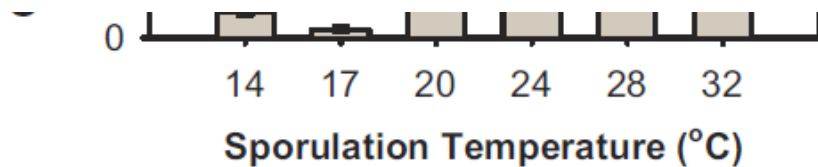
The allergenicity of *Aspergillus fumigatus* conidia is influenced by growth temperature

Culturability of *A. fumigatus* spores grown under different conidiation temperatures (\pm standard error, n = 3 experiments).

Allergenicity per 10^7 spores of *Aspergillus fumigatus* conidia cultivated under different sporulation temperatures. Error bars represent standard error values over three independent experimental replicates. (Inset) Protein content per 10^7 spores in conidia produced at 17 C, 25 C, and 32 C.



“...results of this study indicate that environmental conditions at growth significantly influence the allergenicity of this common mold through the differential production of allergenic proteins, and highlight the importance of in vivo or in vitro allergenicity”



Changes in atmospheric CO₂ influence the allergenicity of *Aspergillus fumigatus* (Lang-Yona et al, 2013)

- Hypothesis that environmental conditions linked to global atmospheric changes can affect the allergenicity of *Aspergillus fumigatus*, a common allergenic fungal species in indoor and outdoor environments and in airborne particulate matter.
- Fungi grown under present-day CO₂ levels (392 ppm) exhibit 8.5 and 3.5 fold higher allergenicity compared to fungi grown at preindustrial (280 ppm) and double (560 ppm) CO₂ levels, respectively.
- Corresponding trend observed in the gene expression encoding for known allergenic proteins and in the major allergen Asp f1 concentrations,
- Possibly due to physiological changes such as respiration rates and the nitrogen content of the fungus, influenced by the CO₂ concentrations.
- Increased carbon and nitrogen levels in the growth medium also lead to a significant increase in the allergenicity.
- Propose that climatic changes such as increasing atmospheric CO₂ levels and changes in the fungal growth medium may impact the ability of allergenic fungi such as *A. fumigatus* to induce allergies.

The different CO₂ concentrations for each treatment in the CO₂ chamber experiment

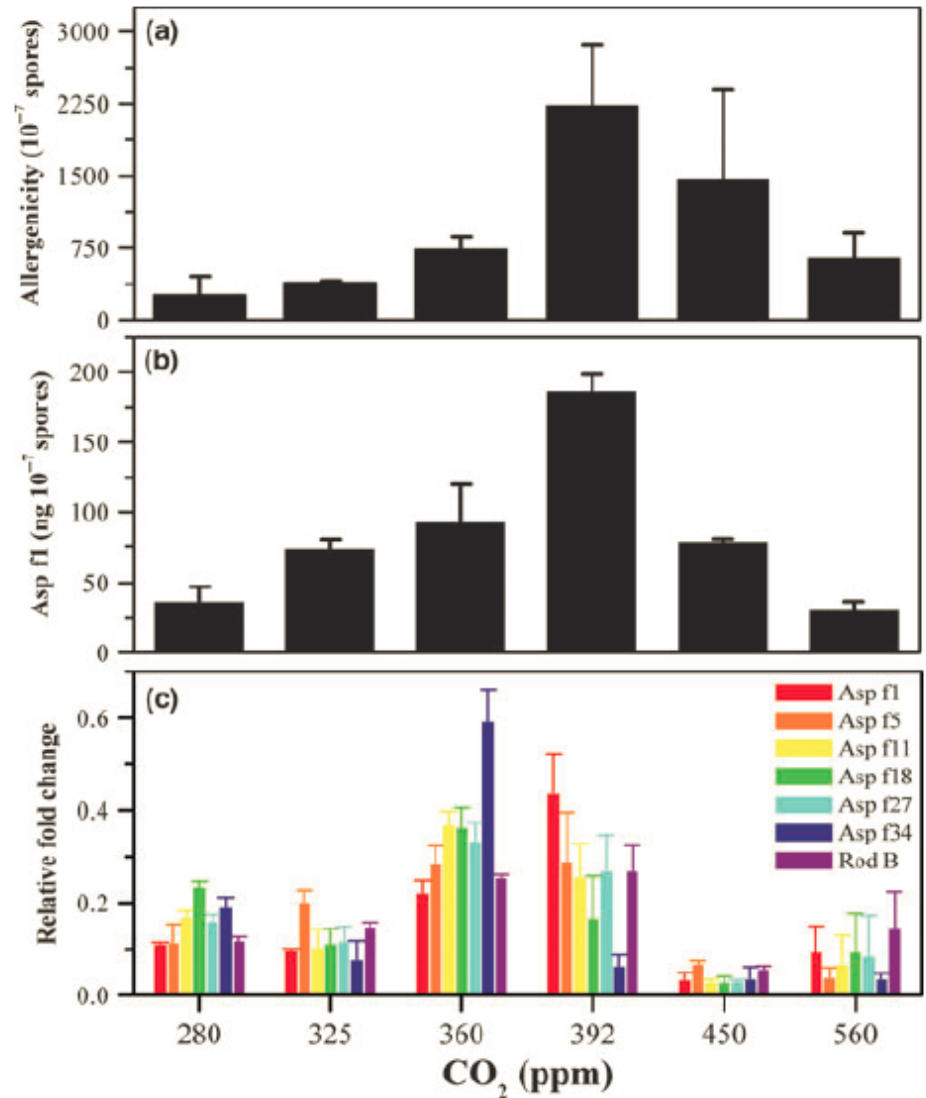
Treatment	CO ₂ concentration (ppm)	Remarks
1	280	Preindustrial equivalent concentration
2	325	Atmospheric CO ₂ levels during the year of 1968 (Tans, 2008)
3	360	Atmospheric CO ₂ levels during the year of 1993 (Keeling, 1999; Tans, 2008)
4	392	Atmospheric CO ₂ levels during the year of 2010 (Keeling, 1999; Tans, 2008)
5	450	Future prediction for the year of 2030 according to the A1F1 model (Johns <i>et al.</i> , 2003)
6	560	Future prediction for the year of 2050 according to the A1F1 model (Johns <i>et al.</i> , 2003)

The effect of changing CO₂ concentrations on the total allergenicity per 10⁷ spores of *Aspergillus fumigatus*

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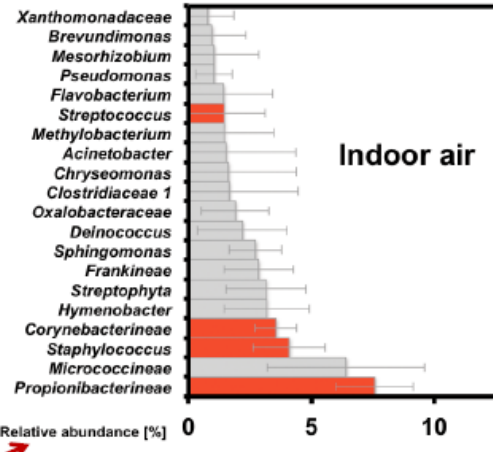
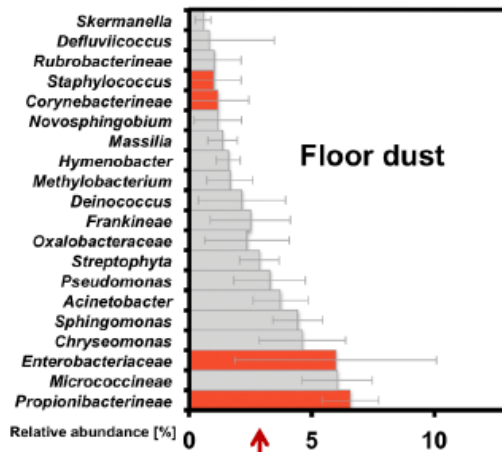
a), the major allergen Asp f1 concentration in ng per 10⁷ spores (b), and the gene expression by RT-PCR (c). The error bars represent the SE of the mean.



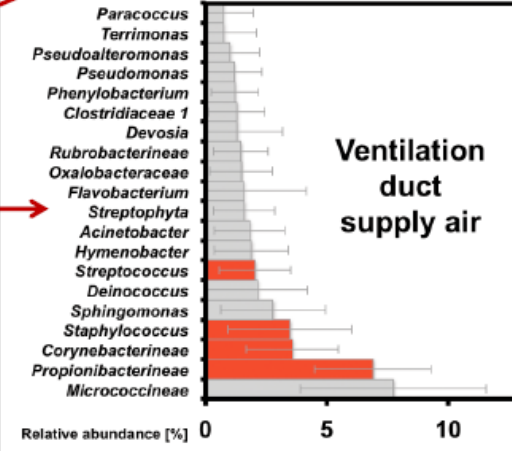
Humans contribute a significant fraction of bacteria found in indoor air and dust

Slide courtesy of Charles J. Weschler from Plenary lecture, 22 Aug 2013

Gene sequence analysis: indoor air & dust



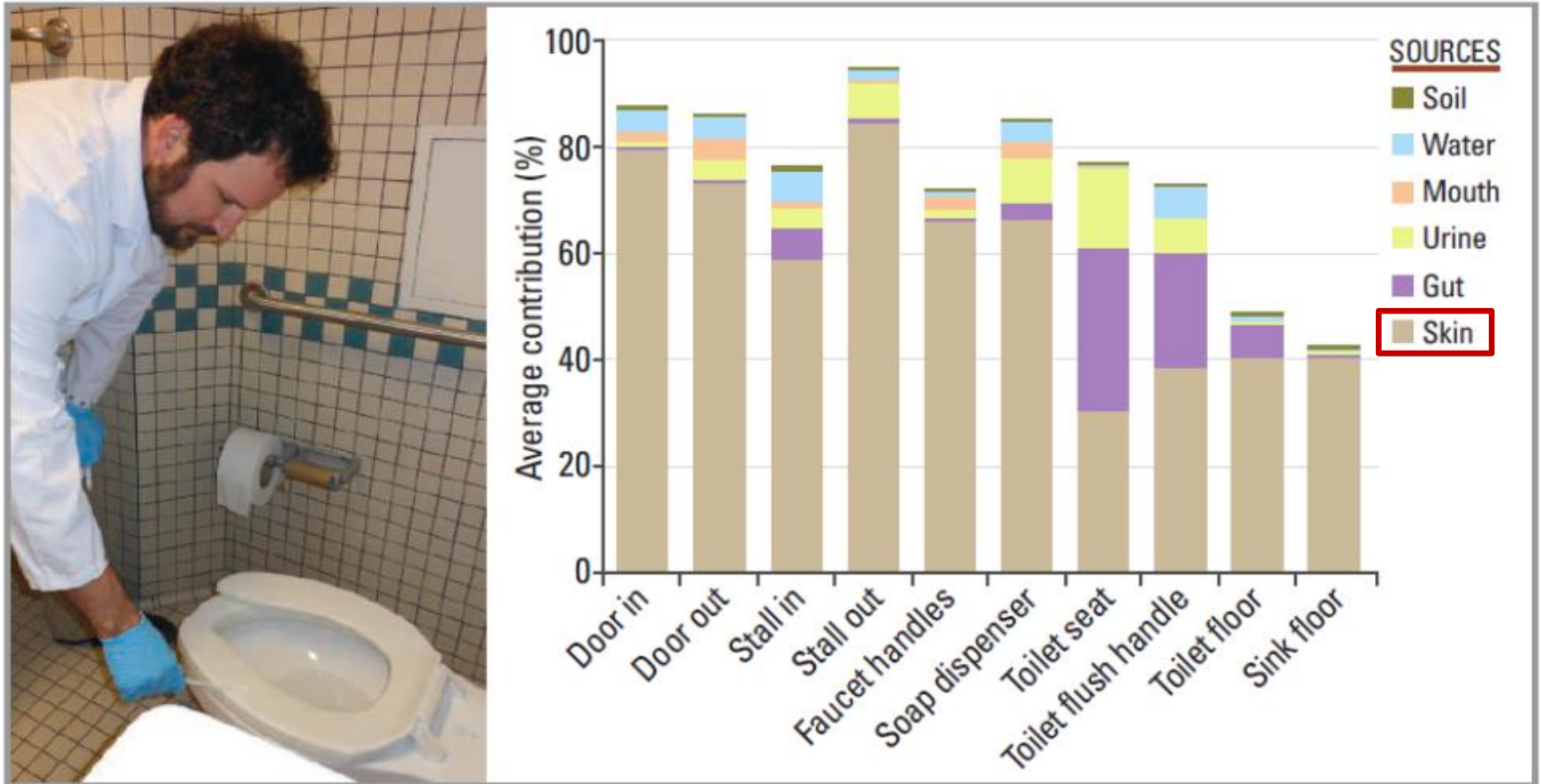
Red bars: bacteria associated with human skin, hair & nostrils – roughly 20% of all bacteria in indoor dust and air



- Shedding of skin cells and subsequent resuspension contribute to airborne bacteria
- Occupants commonly inhale microbes shed by others

further evidence of microbes on all indoor surfaces – “unidentified complex surface films” - UCSF

Gene sequence analysis: further evidence for soiling by squames
Slide courtesy of Charles J. Weschler from Plenary lecture, 22 Aug 2013



Sources of microbes found on different surfaces in public toilets

Unidentified complex surface films (UCSF)

Every indoor surface is coated with a film of organic chemicals, water, and particles including inorganic and microbial particles in a dynamic, interactive community or ecosystem

Exposure science is
not so simple!



Very short Summary

- Study of microbial ecology of the indoor environment has advanced in recent years
 - due in part to steep decline in cost of analysis
 - due in part to substantial injection of research funding by the Alfred P. Sloan Foundation.
- The results to date indicate that we are just beginning to recognize the nature of microbes role in the indoor environment.
- There is a lot of work to do and it will be fun and exciting.