The emerging importance and challenges of the human mycobiome

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Advances in genomics have significantly expanded our ability to examine human microbes. These studies on the human microbiome are elucidating the integral relationships that hosts have with their microbial communities. Many microbiome studies including large-scale microbiome efforts of the National Institutes of Health’s Common Fund Human Microbiome Project and MetaHIT have most often focused on bacteria, with fewer efforts focused on examining human fungi. Fungi comprise a smaller proportion of human microbial sequences as compared with bacteria, but remain an important component of the human microbiome. Fungi have been studied for well over a century with research ranging from the role of fungi in human diseases to the influence of fungi on bacterial growth, and from the interactions between fungi and host immunity to the biologic activities of fungal metabolic products. The emergence of genomic methods to study fungi has the potential to expand our understanding of the contributions of human fungi to health and disease. This issue of Virulence reviews literature on the human mycobiome, focusing on distinct body sites.

Studies of the bacterial microbiome of the gastrointestinal tract have formed the foundation of next-generation sequencing investigations of the human microbiome. Far fewer studies have examined the mycobiome of the gut in health and disease or investigated its functional impact on the host. While bacteria outnumber fungi in the gut, there are a significant number of fungal taxa in the gut and the majority of them can only be detected with non-culture-based techniques. Although establishment of the mycobiome of the gut is not as well-studied as the bacterial microbiome, it appears that the first fungi are transmitted by the mother. Dietary intake then becomes the main driver of the gut mycobiome. Diet shapes the bacterial microbiome primarily by providing nutrients. In contrast, fungi primarily enter the gut via food colonization. Fungal species that enter the gut through the diet may become resident or be eliminated. Vegetarians appear to have different mycobiomes than those consuming a Western diet, but few studies of the influence of dietary interventions on gut fungal communities have been performed. The interactions of gut fungal communities with bacteria and viruses are increasingly being explored, and the potential implications for understanding and treating disease remain an important unanswered question.

Recent data support a role of the gut mycobiome in altering the immune response and affecting both local and systemic disease, and we expect that future work will continue to elucidate the contribution of the gut mycobiome to various diseases.

Studies of the lung microbiome have lagged behind those in the gut in large part because the lung was thought to be sterile. A growing body of literature now highlights the bacterial communities in the lung in the healthy host and in individuals with lung disease, but as with the gut, the fungal composition of the lung is not as well-understood. The lung comes into contact with environmental fungi routinely during respiration, and oral fungi may be aspirated into the lungs. The most common fungi in the normal lung are the family Davidiellaceae, and the genera Cladosporium, Eurotium, Penicillium, and Aspergillus, but composition is highly variable between individuals. Because fungi can trigger both the innate and adaptive immune systems, it is hypothesized that fungal dysbiosis may be a component of chronic lung diseases such as asthma, cystic fibrosis, and chronic obstructive pulmonary disease (COPD). Use of immunosuppression such as that during transplantation may also alter the lung mycobiome and further contribute to underlying disease. In general, lung disease is associated with lower fungal diversity; however, the relative paucity of current literature makes it premature to...
generalize. Understanding of the lung mycobiome has also been hampered by technical issues. Some of these issues are those associated with any study of the mycobiome (i.e. lack of reference genomes), but some are lung-specific. For example, the lung is more difficult to access than other sites such as the skin or gut, and the low biomass of the lung leads to susceptibility to contamination using next-generation sequencing (NGS) techniques. Despite these difficulties, studies of the lung mycobiome in both humans and animal models focusing on the composition and function of the lung mycobiome as well as its effects on the host are critical to advance the field.

Fungi are an integral component of the vaginal microbiological ecosystem in many healthy women, with early studies reporting cultivation of fungal isolates from vaginal samples. In particular, the opportunistic pathogen Candida albicans asymptomatically colonizes 20% of women. Implementation of sequencing methods has led to greater understanding of the complexity of these vaginal fungal communities. These mycobiome studies have shown that Candida can be found in up to 70% of samples and that the vaginal fungal diversity has been underestimated. Work is ongoing to uncover the mechanisms of the balance between commensalism and pathogenicity. Known factors contributing to vulvovaginal candidiasis include immune defects, microbial dysbiosis, and epithelial disruption. Studies of the pathogenicity of Candida have also examined morphogenesis genes, proteolytic enzymes, and biofilm formation as microbial factors that play a role in vulvovaginal candidiasis. Interactions between vaginal bacteria and fungi may also play a role in maintaining balance in the vaginal microbial ecosystem. Understanding the physical, physiologic, and immunological interactions as well as metabolites and small molecules produced by microbial communities will be important in shedding light on vaginal health and disease and may result in therapeutic advances.

Cultivation studies have previously established the presence of skin fungi in healthy individuals and patients with skin diseases. These surveys have commonly isolated Malassezia spp. Studies of the skin mycobiome, though less numerous than sequencing studies of skin bacterial communities, have also identified the predominance of Malassezia in most skin sites and have highlighted the complexity of skin fungi. Existing skin mycobiome studies have examined the biogeography of fungal communities, showing that some regions of the skin surface (scalp, forehead) have higher relative abundances of M. globosa and other areas (back) have higher relative abundances of M. restricta. Recent studies of genomic differences in different species of Malassezia suggest that genes such as the fatty acid synthase gene can affect niche adaptation. In addition to skin site location, age and gender may influence fungal skin communities which may be an important consideration when designing clinical studies and when examining skin mycobiome differences in a particular disorder. The roles of fungi in skin diseases have been based primarily on culture studies. Recent studies have examined the mycobiome in skin disorders indicating that fungal diversity is inversely correlated with disease severity in atopic dermatitis and psoriasis. Additional investigations of fungal communities in these diseases as well as in immunodeficiency syndromes have the potential to expand our understanding of the host-microbial interactions.

The majority of culture-based oral fungal research has often focused on Candida and oral candidiasis. Some oral cultivation studies have isolated non-Candida fungal organisms, particularly in immunosuppressed patients, but have questioned whether these fungi result from potential environmental sources or contamination. The incorporation of genomics-based approaches to studying oral fungal communities has highlighted some of the limitations of cultivation approaches and the potential environmental sources or contamination. These sequencing analyses have been similarly identified in sequencing studies, yet mycobiome studies have also shown that a fungus that had not been previously cultured from the oral cavity, Malassezia, is often present and in high relative abundances. The several studies to date have used different methodologies which can significantly influence the resultant DNA yields and subsequent analyses. These examples cite a need for additional studies for optimization of methods to sufficiently capture the complexity of the oral mycobiome. While existing efforts to standardize human microbiome studies are often based on a more robust analysis pipeline for bacterial communities, the relatively early stages of oral mycobiome studies provides an opportunity to overcome challenges to the field. These areas for development apply to other regions of the human mycobiome and include validated protocols and analytical tools, different marker genes for amplification and sequencing, nomenclature, and curated databases. In conjunction with these efforts, deeper mechanistic studies will be required to understand the role of the mycobiome in oral health and disease.

In summary, study of the mycobiome of various body sites and in human health and disease remains in its infancy. The mycobiome field needs expansion of methodological studies, sequenced isolates, curated reference databases, and mechanistic studies, including research on the interactions of fungi with bacteria and viruses. These investigations highlight the challenges of working with microbes that require distinct cultivation conditions.
and demonstrate the importance of the complementary approaches of culture- and genomics-based investigations. We are only beginning to understand how the mycobiome functions to both maintain health and to cause disease, but fungi may be an underappreciated component of many illnesses.

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