Review

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Future MICROBIOLOGY

The hidden pathogenic potential of environmental fungi

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Invasive fungal infections are a growing threat to immunocompromised patients, highlighting the importance of monitoring fungal pathogens. Global warming (including climatic oscillations) may select for environmental species that have acquired thermotolerance, a key step toward pathogenesis to humans. Also, important virulence factors have developed in environmental fungi, because they are essential for yeast survival in the environment. Thus, fungi traditionally regarded as nonpathogenic to humans have virulence factors similar to those of their pathogenic relatives. Here, we highlight the emergence of saprophytic environmental fungi – including species of *Cryptococcus*, *Aspergillus*, *Penicillium*, *Candida* and *Scedosporium* – as new human pathogens. Emerging pathogens are, in some cases, resistant to the available antifungals, potentiating the threat of novel fungal diseases.

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Fungal infections on the rise

Over the last decades, the world has experienced an increase in the incidence and spread of emerging fungal infections. These events may be associated with several factors, including the increase in life expectancy, which generates older individuals with a weaker immune response, as well as increased epigenetic abnormalities that may favor disease establishment. In the case of fungal infections, the reduction in the host's natural defenses (caused by disease or immunosuppressive medication) meant that numerous pathogens previously considered harmless gradually became disease agents, causing serious infections that are both resistant to antifungal and fatal. The incidence of pathogenic fungal infections has risen enough to constitute a fundamental change the epidemiology of invasive fungal diseases, especially in immunocompromised individuals, such as those with HIV, cancer or subjected to transplants [1–5].

The distribution of fungal species on the planet was determined, broadly, by the following effects: the separation of fungal species through continental drift; the European colonial expansion, when various fungal diseases were brought from the old world to the new world; and human displacement (emigration, migration, immigration) [6]. Enviromental fungi that are pathogenic to humans exist in a broad range of geographic areas, but they are more common in warmer regions of the planet, probably due to temperature and humidity restrictions on their growth or propagation. This is the case for endemic species such as *Blastomyces* spp., *Coccidioides* spp., *Histoplasma* spp. and the species *Cryptococcus gattii* [7]. In contrast, environmental fungi capable of growing at wide temperature ranges are ubiquitous and cosmopolitan [8].

Natural disasters are clear examples of environmental conditions changing the dynamics of fungal populations. The link between natural disasters – which alter the local environmental conditions – and the subsequent occurrence of a typical fungal infections in disaster-affected individuals has been increasingly recognized [8], and highlights the importance of environmental factors on the dispersion of fungal diseases. For example, after the Japan earthquake and tsunami in 2011, fatal cases of pulmonary multiorgan dissemination by *Aspergillus fumigatus* and *Scedosporium apiospermum* were described, while these fungi do not typically cause invasive infections in immunocompetent hosts [9,10].

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Global warming & the emergence of novel fungal pathogens

The emergence of new diseases most frequently results from changes in host or pathogen ecology, from environmental alterations, or from interactions of all of these components. Increased environmental pressures – such as global warming and higher ultaviolet radiation levels – may result in stronger pathogens with advanced virulence mechanisms that favor survival in harsher environmental conditions [11].

Climate change is a consequence of biotic and abiotic processes, such as variations in solar radiation and human activities, which have been identified as the primary causes of global warming. Evidence suggests that the increase in the Earth's temperature over the last centuries has already resulted in marked ecological changes, including alterations in the seasons and in the distribution of a variety of species [12,13]. Climate change is expected to widen the invasion niche of many fungal species [14], and it has already driven shifts in the composition of saprotrophic fungal populations [15]. Most fungi grow at 12–30°C, suffering a rapid decline in growth and viability at temperatures above 30°C [16]. Consequently, the high mammalian body temperatures are sufficient to inhibit the growth and replication of the majority of fungal strains [17–19]. However, a warmer climate could change the distribution of both heat-tolerant and susceptible species, by selecting for those that are more thermotolerant. This effect is expected not only to widen the geographic area of pathogenic (i.e., intrinsically heat-tolerant) species, but also to facilitate the close interaction of environmental fungi with human populations, leading to the emergence of novel fungal pathogens.

Virulence factors in environmental fungi: a reservoir of potential pathogenesis tools

The majority of fungi are facultative parasites, and the passage through a host is not an essential part of their life cycle. Most fungi capable of systemic human infection are environmental species whose opportunistic and fatal pathogenicity derives mostly from strategies developed to counteract environmental stresses [20]. Therefore, to survive in the dangerous environment of the human body – under high temperatures and constant attack by powerful immune defenses – fungi make use of virulence factors initially developed for survival in the rich ecological niche of the soil. In recent years, studies with amoeba, slime molds and worms have led to the proposal that interactions between fungi and other environmental microbes, including predators, select for characteristics that also favor survival in animal hosts [21–25]. Virulence factors allow fungi to interact with the complex and dangerous soil biota; thus, soil residence is associated with the maintenance of pathogenic fungal potential for mammalian host infection [26,27]. Interestingly, Rhame and co-workers (1995) showed that bacterial virulence factors are also versatile, with the same factors conferring pathogenicity to both plant and animal hosts [28].

Morphological changes (called fungal 'morphogenesis'), which include variations in cell size and shape, are employed by many fungal species to survive in the environment, and also within 'accidental' hosts. Morphogenesis is considered as a mechanism for fungal locomotion, is important for fungal dissemination in the environment, and may also represent a protection mechanism against amoeboid cell predation [29]. In addition, shape changes can be used as virulence factors in the establishment of fungal infections. Importantly, for most fungi, the ability for shape change is critical for human infectivity, because many pathogenic species enter the human host in the form of small, round airborne dispersal propagules, sporangiospores or conidia, which are produced from hyphal cells. Small fungal cells (1–5 μm in diameter) are ideal for dispersal and entry into hosts, and are also easily internalized by lung macrophages, which is important for pathogenesis by *Cryptococcus* spp. and *Histoplasma* spp. However, after morphogeneses, *Cryptococcus* spp. cells can reach up to 100 μm in diameter ('giant' cells), which happens in later stages of lung infection, while *Histoplasma* sp. form macroconidia of 8–16 μm in diameter [30–32]. Fungal hyphae can penetrate tissue barriers, while yeasts can disseminate more easily to distant sites [24,33]. Thus, 'dimorphic' fungal species such as *Histoplasma capsulatum*, *Paracoccidioides brasiliensis* or *Coccidioides immitis* grow in the hyphal form in the environment, for fungal dissemination and the absortion of nutrients, but shift to the yeast form in the human host. Temperature is a key host signal to trigger hyphae-to-yeast conversion during the establishment of fungal pathologies [34].

For their nutrition in the environment, fungi excrete an array of enzymes that digest complex molecules, for later absorption of their organic components (polypeptides, sugar or complex carbohydrates). In the host, the secretion of digestive enzymes degrades host tissues, generating nutritious products. Enzyme secretion also protects fungi against antimicrobial mechanisms [35,36]. Different examples of this phenomenon include the secretion of: superoxide dismutase, which protects against toxic free radicals, by detoxifying the superoxide anion [37]; phospholipases, which facilitates intracellular proliferation [38]; and urease, which favors intracellular growth and dissemination [39].

The ability to adapt rapidly to variations in the environment, via processes such as microevolution (i.e., rapid phenotypic changes driven by allelic fluctuations) [40] also provides a selective advantage to fungi during the colonization of human hosts. Rapid adaptation enables fungal pathogens to avoid recognition or destruction by the host immune system, resulting in latency and long-term pathogen persistence. In addition, rapid adaptation facilitates the emergence of resistance to antimicrobial agents, which can develop in sensitive species over time, due to improper antifungal use. Although, most acquired antifungal resistance develops in *Candida* species, resistance has also been described in other types of fungi, such as *Aspergillus* spp. [41–45].

Components of the cell wall that are exposed on the cell surface protect fungi from assaults by the environmental and the host, and some cell wall constituents are considered virulence factors [46]. The cell wall polysaccharides chitin and glucans were associated with an increase in virulence in *Blastomyces dermatitidis, P. brasiliensis* and *H. capsulatum*. These polyssaccharides protect fungal cells against humoral immune response induction and macrophage activation [47]. On the other hand, surface molecules that play a role in adhesion – such as Gp43 from *P. brasiliensis* [48] – are important for the colonization and subsequent invasion of host tissues.

Some fungal species are also capable of modifying the pre-existing external structure of the cell wall, or producing new structures outside the cell wall, such as the polysaccharide capsule that is the hallmark of the global fungal pathogen *Cryptococcus* spp. [49]. In the environment, the capsule protects the organism against stress conditions such as dehydration and phagocytosis by environmental amoeba [50]. In the host, it contributes to infection by protecting the cell against a variety of host immune defenses, interfering with phagocytosis and suppressing both cellular and humoral immunity [51,52].

Melanins are high-molecular-weight pigments produced by a wide variety of fungal species, including *Cryptococcus* spp., *Paracoccidioides* spp., *Histoplasma* spp., *Sporothrix* spp., *Aspergillus* spp. *and Alternaria* spp. [53]. Given its chemical properties (light absorption, hydrophybicity, insolubility and negative charge), melanin protects fungal cells in the environment from the action of hydrolytic enzymes, UV and gamma radiation, extreme temperatures and toxic compounds such as heavy metals. In the host, melanin affects phagocytosis by macrophages, reduces the release of proinflammatory cytokines by host cells, and protects fungi against antifungal drugs [53–58].

The growing challenge of emerging fungal pathogens

In the last 100 years, advances in regenerative medicine, stem cell research and chemotherapy contributed to a worldwide increase in life expectancy by more than 30 years. While these medical advances reverted effectively the lethality of several diseases, they also increased the immonusuppressed population, which is particularly susceptible to uncommon fungal infections. The development of virulence factors in the environment and of thermotolerance to mammalian temperatures – which was potentially accelerated by global warming – may also have contributed significantly to the rise in opportunistic fungal infections by environmental species. Here, we will highlight some of the key species of environmental fungi that have been increasingly associated with human infections, particularly in immunocompromised individuals.

Although, some species may represent truly novel pathogens, improved diagnosis has also identified as pathogenic some species that were previously thought to be restricted to the environment. New molecular methods for the detection and identification of fungal pathogens have been developed that significantly improved our ability to distinguish between previously identified and new pathogenic species, and led to the description of new opportunistic fungal pathogens in an effective and timely manner [59].

From 2008, *Candida auris* has emerged as a new multidrug-resistant *Candida* species associated with invasive infection and high rates of mortality. The first description of human pathogenesis by *C. auris* was in 2008, of a Japanese patient with otitis [60]. Since then, *C. auris* has been reported as an agent of candidemia in Japan, South Korea, India, Kuwait, South Africa, Pakistan and the UK and, more recently, in Venezuela, Colombia and the USA [61–64]. Multidrug resistance has been widely described in *C. auris*, with the majority of clinical strains reported as resistant to the widely used antifungal fluconazole, while some strains are resistant to azoles in general, to polyenes, and also to echinocandins [41,65–67]. These strains present all the major *Candida* virulence factors, including germination, adherence, biofilm formation, and phospholipase and proteinase production [68]. *C. auris* is a major public health concern due to its rapid spread as a nosocomial infection agent in healthcare settings, with potential to cause outbreaks with high mortality rates [69].

Cryptococcosis has also grown in the last decades as a disseminated fungal infection in immunocompromised patients. Although, *Cryptococcus neoformans* and *Cryptococcus gattii* are the main agents of human cryptococcosis, opportunistic infections by environmental species, such as *C. liquefaciens*, *C. albidus* and *C. laurentii*, have been

observed recently [70]. These infections are probably acquired from environmental reservoirs such as bird excrement, trees, food (cheese and fruit), soil and water. Factors that are likely to contribute to the increased incidence of non-*neoformans* and non-*gattii* human infections include improved laboratory detection, the higher incidence of immunocompromised individuals, and the selective pressure of global warming toward the development of tolerance to human host temperatures [70,71]. Studies on cryptococcal virulence factors are typically conducted in *C. neoformans*, since other species of the genus were presumed to have low pathogenic potential. Recently, Araujo and co-workers (2017) described striking similarities in composition and structure between PS molecules of *C. liquefaciens* and *C. neoformans*, and showed that PS molecules from these species have comparable behavior in several key biological activities [72,73]. The remarkable similarities in capsule ultrastructure and virulence between single isolates of *C. neoformans* and *C. liquefaciens* described in the study of Araujo and co-workers (2017), together with the first descriptions of human fungemia by *C. liquefaciens* in immunocompromised patients [70–74], suggest that this environmental cryptococcal species may represent a novel human pathogen. Importantly, *C. liquefaciens* infections are resistant to 5-fluorocytosine, indicating that that treatment of fungemia by this species may be challenging [70,71].

In general, aspergillosis is not a notifiable infection, and it often goes unreported by patients; therefore, it is difficult to determine the exact number of cases of this disease worldwide. However, it is clear that allergic bronchopulmonary aspergillosis is common worldwide, while invasive aspergillosis is considered as an uncommon form associated primarily with hospitalized immunocompromised patients [75,76]. Building works generate dust contamination and disperse large amounts of fungal spores in the environment. Invasive aspergillosis cases are, indeed, often associated with hospital renovation or building work, which can increase the amount of airborne *Aspergillus*, resulting in respiratory infections or surgical site infections in high-risk patients. Although, *Aspergillus* species are the most common causative pathogens in fungaemia outbreaks related to construction work, *Zygomycetes* and other fungi were occasionally reported [77,78]. Outbreaks of primary cutaneous and central nervous system aspergillosis associated with the use of contaminated medical devices have also been described [78].

Although, aspergillosis remains clearly the most frequent opportunistic mold infection in patients with immunodeficiency, new opportunistic pathogens have now emerged as the cause of life-threatening fungal infections worldwide. One important opportunistic fungal pathogen is *Fusarium* spp., a saprophytic soil fungus capable of causing disseminated infections in patients with hematologic malignancies [79–81]. Invasive fusarial infections are either airborne or inoculated through rupture of the skin barrier and, once established, are refractory to standard antifungal therapy [79–81].

Scedosporium spp. are soil fungi that have been implicated in numerous infections in immunocompromised and immunocompetent patients [82,83]. These mycoses are life threatening in susceptible patients and can be considered truly emerging diseases. *Scedosporium* infections are difficult to diagnose and treat. In particular, *S. apiospermum* and *S. prolificans* – emerging opportunistic pathogens associated with mycetoma and keratitis infections – are inherently resistant to most antifungal agents and, thus, considerably difficult to combat in the clinic [82–84].

An additional emerging pathogen that deserves mention is *Penicillium marneffei*, which is responsible for 'penicilliosis marneffei'. This disseminated and progressive disease represents the third most common opportunistic infection in HIV patients in certain parts of Southeast Asia (and is also endemic in Southeast Asia and Southern China) [85]. Infections caused by *Penicillium* species other than *P. marneffei* are rare, and the spectrum of disease produced by *Penicillium* spp. is similar to that of *Aspergillus* spp.; thus, diseases by these two fungal species are difficult to distinguish clinically. Pulmonary infection is the most common presentation of penicillosis marneffei; however, fungal transmission via trauma, surgery or prosthetic material is commonly implicated in nonpulmonary disease [86].

Future perspective

The frequency of invasive mycoses due to opportunistic fungal pathogens has increased significantly in the last two decades. Immunologically competent mammals display robust defenses against fungal diseases, but recent developments in the evolution of human societies has produced large cohorts of individuals susceptible to fungal infections. On the other hand, it is likely that environmental changes (including global warming) are selecting for fungi increasingly resistant to the harsher conditions encountered in mammalian hosts. Consequently, the number of fungal pathogens currently documented is extensive and ever increasing, and one can no longer ignore or discard environmental fungal species as nonpathogenic to humans. Also, improved diagnostic tools with increased microbial genotyping precision are likely to identify disease isolates that represent species not previously known as

pathogenic, but which had been classified incorrectly as a known pathogenic species. Growing resistance to fungal infection, due to excessive antifungal use in some countries, may also contribute to the emergence of novel fungal pathogens.

Executive summary

- Advances in regenerative medicine, transplants and chemotherapy contributed to extend life expectancy, while also increasing the number of immunocompromised hosts susceptible to unusual fungal infections.
- Environmental fungi subjected to mounting environmental pressure have developed new or more potent virulence factors.
- The emergence of saprophytic fungi as mycoses agents is a growing challenge, particularly when associated with drug resistance, highlighting the urgent need for novel antifungal agents.
- Advances in diagnostics have allowed the identification of new pathogenic fungal species previously indistinguishable from known pathogens.

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References

Papers of special note have been highlighted as: • of interest; •• of considerable interest

1. Martin GS, Mannino DM, Eaton S *et al.* The epidemiology of sepsis in the United States from 1979 through 2000. *New Engl. J. Med.* 348(16), 1546–1554 (2003).
2. Holzheimer RG, Dralle H. Management of mycoses in surgical patients – review of the literature. *Eur. J. Med. Res.* 7(5), 200–226 (2002).
3. Peres-Bota D, Rodriguez-Villalobos H, Dimopoulos G *et al.* Potential risk factors for infection with *Candida* spp. in critically ill patients. *Clin. Microbiol. Infect.* 10(6), 550–555 (2004).
4. Perlroth J, Choi B, Spellberg B. Nosocomial fungal infections: epidemiology, diagnosis, and treatment. *Med. Mycol.* 45(4), 321–346 (2007).
5. Meersseman W, van Wijngaerden E. Invasive aspergillosis in the ICU: an emerging disease. *Intensive Care Med.* 33(10), 1679–1681 (2007).
6. Casadevall A, Freij JB, Hann-Soden C, Taylor J. Continental drift and speciation of the *Cryptococcus neoformans* and *Cryptococcus gattii* species complexes. *mSphere* 2(2), e00103-17 (2017).

•• **Hypothesis is to stimulate thought and discussion about the mechanisms driving fungal speciation and the origins of fungal virulence.**

7. Sifuentes-Osornio J, Corzo-Leon DE, Ponce-de-Leon LA. Epidemiology of invasive fungal infections in Latin America. *Curr. Fungal Infect. Rep.* 6(1), 23–34 (2012).
8. Benedict K, Park BJ. Invasive fungal infections after natural disasters. *Emerg. Infect. Dis.* 20(3), 349–355 (2014).
9. Nakamura Y, Utsumi Y, Suzuki N *et al.* Multiple *Scedosporium apiospermum* abscesses in a woman survivor of a tsunami in northeastern Japan: a case report. *J. Med. Case Rep.* 5, 526 (2011).
10. Igusa R, Narumi S, Murakami K *et al. Escherichia coli* pneumonia in combination with fungal sinusitis and meningitis in a tsunami survivor after the Great East Japan Earthquake. *Tohoku J. Exp. Med.* 227(3), 179–184 (2012).
11. Schrag SJ, Wiener P. Emerging infectious disease: what are the relative roles of ecology and evolution? *Trends Ecol. Evol.* 10(8), 319–324 (1995).
12. Wookey PA, Aerts R, Bardgett RD *et al.* Ecosystem feedbacks and cascade processes: understanding their role in the responses of arctic and alpine ecosystems to environmental change. *Global Change Biology* 15(5), 1153–1172 (2009).
13. van der Putten WH. Climate change, aboveground–belowground interactions and species range shifts. *Ann. Rev. Ecol. Evolut. Syst.* 43, 365–383 (2012).
14. Greer A, Ng V, Fisman D. Climate change and infectious diseases in North America: the road ahead. *CMAJ* 178(6), 715–722 (2008).
15. Classen AT, Sundqvist MK, Henning JA *et al.* Direct and indirect effects of climate change on soil microbial and soil microbial–plant interactions: what lies ahead? *Ecosphere* 6(8), 130 (2015).

- **They explore how climatic change affects soil microbes and soil microbe–plant interactions.**

16. Robert VA, Casadevall A. Vertebrate endothermy restricts most fungi as potential pathogen. *J. Infect. Dis.* 200(10), 1623–1626 (2009).
17. Baxter M, Illston GM. Temperature relationships of fungi isolated at low temperatures from soils and other substrates. *Mycopathologia* 72, 21–25 (1980).
18. Marquez LM, Redman RS, Rodriguez RJ, Roossinck MJ. A virus in a fungus in a plant: three-way symbiosis required for thermal tolerance. *Science* 315, 513–515 (2007).
19. Garcia-Solache MA, Casadevall A. Global warming will bring new fungal diseases for mammals. *MBio* 1(1), e00061-10 (2010).

- **Hypothesis on how global warming will bring new fungal diseases for mammals.**

20. Brown AJ, Budge S, Kaloriti D *et al.* Stress adaptation in a pathogenic fungus. *J. Exp. Biol.* 217(Pt 1), 144–155 (2014).
21. Levitz SM. Does amoeboid reasoning explain the evolution and maintenance of virulence factors in *Cryptococcus neoformans*? *Proc. Natl Acad. Sci. USA* 98(26), 14760–14762 (2001).
22. Steenbergen JN, Casadevall A. The origin and maintenance of virulence for the human pathogenic fungus *Cryptococcus neoformans*. *Microbes Infect.* 5(7), 667–675 (2003).
23. Steenbergen JN, Nosanchuk JD, Malliaris SD, Casadevall A. *Cryptococcus neoformans* virulence is enhanced after growth in the genetically malleable host *Dictyostelium discoideum*. *Infect. Immun* 71(9), 4862–4872 (2003).
24. Steenbergen JN, Nosanchuk JD, Malliaris SD, Casadevall A. Interaction of *Blastomyces dermatitidis*, *Sporothrix schenckii*, and *Histoplasma capsulatum* with *Acanthamoeba castellanii*. *Infect. Immun.* 72(6), 3478–3488 (2004).
25. Steenbergen JN, Shuman HA, Casadevall A. *Cryptococcus neoformans* interactions with amoebae suggest an explanation for its virulence and intracellular pathogenic strategy in macrophages. *Proc. Natl Acad. Sci. USA* 98(26), 15245–15250 (2001).
26. McClelland EE, Bernhardt P, Casadevall A. Estimating the relative contributions of virulence factors for pathogenic microbes. *Infect. Immun.* 74(3), 1500–1504 (2006).
27. Casadevall A, Pirofski L-A. Accidental virulence, cryptic pathogenesis, martians, lost hosts, and the pathogenicity of environmental microbes. *Eukaryot. Cell* 6(12), 2169–2174 (2007).
28. Rahme LG, Stevens EJ, Wolfort SF, Shao J, Tompkins RG, Ausubel FM. Common virulence factors for bacterial pathogenicity in plants and animals. *Science.* 268(5219), 1899–1902 (1995).
29. Wang L, Lin X. Morphogenesis in fungal pathogenicity: shape, size, and surface. *PLoS Pathog.* 8(12), e1003027 (2012).
30. Seider K, Heyken A, Luttich A, Miramon P, Hube B. Interaction of pathogenic yeasts with phagocytes: survival, persistence and escape. *Curr. Opin. Microbiol.* 13, 392–400 (2010).
31. Okagaki LH, Strain AK, Nielsen JN *et al.* Cryptococcal cell morphology affects host cell interactions and pathogenicity. *PLoS Pathog.* 6, e1000953 (2010).
32. Zaragoza O, García-Rodas R, Nosanchuk JD *et al.* Fungal cell gigantism during mammalian infection. *PLoS Pathog.* 6, e1000945 (2010).
33. Neilson JB, Ivey MH, Bulmer GS. *Cryptococcus neoformans*: pseudohyphal forms surviving culture with *Acanthamoeba polyphaga*. *Infect. Immun.* 20(1), 262–266 (1978).
34. Sil A, Andrianopoulos A. Thermally dimorphic human fungal pathogens – polyphyletic pathogens with a convergent pathogenicity trait. *Cold Spring Harb. Perspect. Med.* 5(8), a019794 (2015).
35. Boelaert JR, de Locht M, Van Cutsem J *et al.* Mucormycosis during deferoxamine therapy is a siderophore-mediated infection. *In vitro* and *in vivo* animal studies. *J. Clin. Invest.* 91(5), 1979–1986 (1993).
36. Kronstad J, Saikia S, Nielson ED *et al.* Adaptation of *Cryptococcus neoformans* to mammalian hosts: integrated regulation of metabolism and virulence. *Eukaryot. Cell* 11(2), 109–118 (2012).
37. Cox GM, Harrison TS, McDade HC *et al.* Superoxide dismutase influences the virulence of *Cryptococcus neoformans* by affecting growth within macrophages. *Infect. Immun.* 71(1), 173–180 (2003).
38. Cox GM, McDade HC, Chen SC *et al.* Extracellular phospholipase activity is a virulence factor for *Cryptococcus neoformans*. *Mol. Microbiol.* 39(1), 166–175 (2001).
39. Rutherford JC. The emerging role of urease as a general microbial virulence factor. *PLoS Pathog.* 10(5), e1004062 (2014).
40. Boyce KJ, Wang Y, Verma S, Shakya VPS, Xue C, Idnurm A. Mismatch repair of DNA replication errors contributes to microevolution in the pathogenic fungus *Cryptococcus neoformans*. *MBio* 8(3), e00595-17 (2017).
41. Kumar D, Banerjee T, Pratap CB, Tilak R. Itraconazole-resistant *Candida auris* with phospholipase, proteinase and hemolysin activity from a case of vulvovaginitis. *J. Infect. Dev. Ctries* 9(4), 435–437 (2015).
42. Lortholary O, Desnos-Ollivier M, Sitbon K *et al.* Recent exposure to caspofungin or fluconazole influences the epidemiology of candidemia: a prospective multicenter study involving 2441 patients. *Antimicrob. Agents Chemother.* 71(9), 4862–4872 (2011).
43. Shah DN, Yau R, Lasco TM *et al.* Impact of prior inappropriate fluconazole dosing on isolation of fluconazole-nonsusceptible *Candida* species in hospitalized patients with candidemia. *Antimicrob. Agents Chemother.* 56, 3239–3243 (2012).