Fungi of entomopathogenic potential in *Chytridiomycota* and *Blastocladiomycota*, and in fungal allies of the *Oomycota* and *Microsporidia*

Agata Kaczmarek[1*] and Mieczysława I. Boguś[1,2]

Abstract

The relationship between entomopathogenic fungi and their insect hosts is a classic example of the co-evolutionary arms race between pathogen and target host. The present review describes the entomopathogenic potential of *Chytridiomycota* and *Blastocladiomycota* fungi, and two groups of fungal allies: *Oomycota* and *Microsporidia*. The *Oomycota* (water moulds) are considered as a model biological control agent of mosquito larvae. Due to their shared ecological and morphological similarities, they had long been considered a part of the fungal kingdom; however, phylogenetic studies have since placed this group within the *Straminipila*. The *Microsporidia* are parasites of economically-important insects, including grasshoppers, lady beetles, bumblebees, colorado potato beetles and honeybees. They have been found to display some fungal characteristics, and phylogenetic studies suggest that they are related to fungi, either as a basal branch or sister group. The *Blastocladiomycota* and *Chytridiomycota*, named the *lower fungi*, historically were described together; however, molecular phylogenetic and ultrastructural research has classified them in their own phylum. They are considered parasites of ants, and of the larval stages of black flies, mosquitoes and scale insects.

Keywords: Biological control, Host-specificity, Insect pathogens

INTRODUCTION

The interaction between host and parasite is characterised on the one hand by the parasites developing more effective strategies of host exploitation, and on the other, by the hosts mounting increasingly robust defences though Red Queen dynamics or coevolutionary arms races. Furthermore, depending on gene flow and differences in selection pressure between sites, both host and parasite may demonstrate local adaptation to their counterpart or develop more general resistance or offensive traits (Kaur et al. 2019).

Although chemical pesticides have been used for the control of insect pests for many decades, they are known to exert side effects on non-target organisms, contaminate groundwater and leave residues on food crops; they are also believed to support the development of insect resistance. Therefore, there is currently great interest in the development of alternative methods for integrated pest management (Kim et al. 2017; Mužinić and Želježić 2018; Neuwirthová et al. 2019). More recent studies have found entomopathogens to regulate many populations of arthropods (Lacey et al. 2001; Lacey et al. 2015; Lacey 2016), and that entomopathogenic fungi themselves may play a role in removing harmful substances and heavy metals from the environment (Litwin et al. 2020).

The success of using an entomopathogenic fungus as a myco-biocontrol agent depends on the selection of a

* Correspondence: aga_kaczm@twarda.pan.pl
[1]Witold Stefański Institute of Parasitology, Polish Academy of Sciences, Twarda 51/55, 00-818 Warsaw, Poland
Full list of author information is available at the end of the article

virulent and stable strain with specific efficacy for the target host. Hence, there has been considerable interest in the creation of new strains of entomopathogenic fungi, accompanied by the development of fermentation systems for optimal biomass production, and the design of delivery systems suitable for both the microorganism and common agricultural practices. One key step in the development of effective strains involves the cloning of genes that can enhance pathogenesis (St. Leger and Wang 2010; Jaronski and Mascarin 2017; Dobrowolski et al. 2018; Karabörklü et al. 2018).

The co-evolution of fungi and insects over hundreds of millions of years has resulted in the development of a wide range of complex and intricate interactions between them (Joop and Vilcinskas 2016). Entomopathogenic fungi have evolved to infect a wide range of insects in all developmental stages (viz. eggs, larvae, pupae, nymphs and imago) across a range of niches. Such adaptability requires considerable morphological diversity, resulting in a wide range of fungal species with morphological, phylogenetic and ecological diversity. However, although recent advances in the genome biology of entomopathogenic fungi indicate the genetic bases of their adaptation to insect hosts and host ranges, as well as the evolutionary relationships between insect and non-insect pathogens (Araújo and Hughes 2016; Wang and Wang 2017), the mechanisms of host specificity in pathogenic microbiology generally remain poorly understood.

EVOLUTION OF ENTOMOPATHOGENIC FUNGI

From an evolutionary point of view, entomopathogenic fungi do not constitute a monophyletic group. Instead, phylogenetic data suggests that entomopathogenic activity has arisen independently, and frequently, along the course of fungal evolution (Humber 2008; Araújo and Hughes 2016; Moonjely et al. 2016). Wang and Wang (Wang and Wang 2017) proposed that this evolutionary pattern is indicative of frequent cross-kingdom or cross-phylum host jumping during fungal pathogen speciation. Such phenomena may be explained by the *host-habitat hypothesis* or the *host-relatedness hypothesis*. The host-habitat hypothesis suggests that entomopathogens accidently switch between host organisms living in a common environment; this better explains the dynamic evolution of host diversification and adaptation in the Hypocreales (Nikoh and Fukatsu 2000; Spatafora et al. 2007; Blackwell 2010). In contrast, the host-relatedness hypothesis suggests that pathogens change hosts to new species closely related with the original host (Nikoh and Fukatsu 2000; Kepler et al. 2012).

Historically, the *Fungi* were divided into four phyla; however, following dramatic changes in higher-level taxonomy in the last 20 years, the number of fungal phyla

has tripled to 12, organized into six major groups: *Dikarya, Mucoromycota, Zoopagomycota, Blastocladiomycota, Chytridiomyceta* and *Opisthosporidia* (James et al. 2020). It has been estimated that about 750 species of entomopathogenic fungi exist, most of which are distributed in the phyla *Chytridiomycota, Blastocladiomycota, Zoopagomycota, Basidiomycota* and *Ascomycota*. Some authors have also traditionally included the *Microsporidia* and the ecologically-similar, but phylogenetically-distinct, *Oomycota* (water moulds), belonging to the *Straminipila*.

OOMYCOTA

The *Oomycota* are a genetically and morphologically-diverse clade that can form hyphae or exist as simple holocarpic thalli; the phylum contains at least 1500 species of fungus in 100 genera (Beakes and Thines 2017). The oomycetes are filamentous eukaryotic microorganisms belonging to the kingdom *Straminipila*; however, some authors placed them among the fungi, based on their ecological and morphological characteristics, viz. filamentous growth habit, nutrition by absorption, and reproduction via spores (Rossman and Palm 2006; Beakes et al. 2012; Thines 2018). Unlike the true fungi, the cell walls of the oomycetes are composed of cellulose derivatives that serve as structural components, rather than chitin (Hassett et al. 2019; Klinter et al. 2019). Sexual reproduction typically occurs between gametangia (antheridia and oogonia) on the same or different hyphae (Rocha et al. 2018; Spring et al. 2018); however, this is not always the case: *Periplasma isogametum*, for example, demonstrates a type of sexual reproduction involving morphological isogamy and physiological anisogamy (Martin and Warren 2020). Most oomycetes are also capable of asexual reproduction via their kidney bean-shaped biflagellate zoospores with apically or laterally-attached whiplash and tinsel flagella (heterokonts) (Walker and van West 2007). Interestingly, the mitochondria of the oomycetes possess tubular cristae, as opposed to the disc-like cristae of the fungi (Powell et al. 1985; Karlovsky and Fartmann 1992; Weber et al. 1998; Rossman and Palm 2006).

Pathogenic oomycetes are able to infect a broad range of algae, plants, protists, fungi, arthropods and vertebrates, including humans, and can cause losses in agriculture and aquaculture (Mendoza and Vilela 2013; van West and Beakes 2014; Kamoun et al. 2015). Twelve species of entomopathogenic *Oomycota* are known to exist within six genera: *Lagenidium* (*L. giganteum*), *Leptolegnia* (*L. caudata* and *L, chapmanii*), *Pythium* (*P. carolinianum, P. sierrensis,* and *P. flevoense*), *Crypticola* (*C. clavulifera* and *C. entomophaga*), *Couchia* (*C. amphora, C. linnophila*, and *C. circumplexa*), and *Aphanomyces* (*A. laevis*) (Araújo

and Hughes 2016; Mendoza et al. 2018). Most are facultative parasites of mosquito larvae belonging to the genera *Anopheles, Culex, Aedes, Ochlerotatus, Culiseta, Orthopodomyia, Uranotaenia, Psorophora,* and *Mansonia* (Frances et al. 1989; Woodring et al. 1995; Scholte et al. 2004; Patwardhan et al. 2005; Vyas et al. 2007; Singh and Prakash 2012; Vilela et al. 2018; Shen et al. 2019; Vilela et al. 2019; Shen et al. 2020a). Some examples of infected insects are listed in Table 1.

The most well-known and thoroughly-analysed entomopathogenic *Oomycota* is arguably *L. giganteum,* which was used as a biological control model of mosquito larvae (Vilela et al. 2019). Infection is initiated by free-floating zoospores formed in sporangia, which are released only in aqueous environments (Walker and van West 2007). The spores are responsible for recognising and binding to the host cuticle. After successful binding, they swell, germinate and penetrate the insect exoskeleton. The mycelia grow through the hemocoel, resulting in the death of the insect, while the fungus terminates with reproduction and the subsequent release of infectious zoospores. Successful infection requires the production of hydrolytic enzymes such as chitinases (Shen et al. 2020b), hexosaminidases (Dowd et al. 2007; Olivera et al. 2016) or glycoside hydrolase family 5 subfamily 27 (GH5_27), which play crucial roles in the cuticle-degrading process (Quiroz Velasquez et al. 2014). Other important enzymes produced by entomopathogenic oomycetes are trehalases, which not only take part in cuticle degradation, but may also hasten the infection process by depleting trehalose, the most abundant sugar source in insect hemolymph (McInnis and Domnas 1973).

It is possible that the ancestors of *L. giganteum* were plant pathogens, as sequence analyses have indicated the presence of genes characteristic of plant tissue infections, such as *crn* or *cbel*. However, other genes coding cuticle-degrading enzymes have been found, such as GH5_27 or GH20; these are characteristic of entomopathogens and are not observed in plant pathogens (Quiroz Velasquez et al. 2014; Olivera et al. 2016). This dichotomy suggests that fungal and oomycete entomopathogens not only share morphology and pathological strategies, but also evolutionary histories and ecological relationships (Leoro-Garzon et al. 2019; Shen et al. 2019).

L. giganteum was used as a control agent against mosquito larvae in the commercial product Laginex, which was registered and released in the USA in 1995 (Hallmon et al. 2000); however, due to several cases of life-threating mycoses in dogs recorded in the southern United States, the product was later deregistered and is no longer for sale (Mendoza and Vilela 2013; Vilela et al. 2015; Vilela et al.

2019). Recent data has shown the existence of two forms of *L. giganteum* (Vilela et al. 2015): the heat-tolerant taxon *L. giganteum* f. *caninum* pathogenic to mammals (Spies et al. 2016), and *L. giganteum* f. *giganteum,* which can infect mosquito larvae in nature. Both taxa share a common ancestor and can infect mosquito larvae; the only phenotypic difference between the two types is their tolerance for growth at different temperatures: the latter does not tolerate human body temperature (37°C) but develops well at 25°C and below (Vilela et al. 2015; Vilela et al. 2019).

Oomycetes have also been found to infect non-dipteran insects; for example *Crypticola entomophaga* has been observed to attack aquatic insects from the order *Trichoptera* (caddis flies; Dick 2003; Gani et al. 2019).

MICROSPORIDIA

Microsporidia are unicellular eukaryotes living as obligate intracellular parasites. All stages of their life-cycle associated with growing and replication can only take place inside host cells; in the environment, they can survive only as thick-walled spores (Timofeev et al. 2020). Their adaptation to a parasitic strategy has resulted in the development of a seemingly paradoxical mixture of characteristics: the cells lack mitochondria, and their metabolism does not employ electron transfer chains, oxidative phosphorylation, or the tricarboxylic acid (TCA) cycle. In addition, their genomes are poor in genes involved in resource-producing metabolic pathways, such as ATP synthesis, but rich in others that enhance transport mechanisms and allow resources to be hijacked from the host. Due to their highly-reduced morphology, ultrastructure, biochemistry and metabolism, as well as their considerably impoverished genome, microsporidia need to induce considerable disruption of host cell physiology to enable successful infection and development (Corradi 2015; Haag et al. 2019; Timofeev et al. 2020). One exception to this rule is *Microsporidium daphniae,* which has been found to possess a mitochondrial genome and the genes necessary for producing ATP from glucose (Haag et al. 2014).

Of all known parasites, those of *Microsporidia* are arguably the most host dependent (Corradi and Keeling 2009; Keeling 2009; Tamim El Jarkass and Reinke 2020), and this dependence may account for their development of a range of mechanisms to ensure intracellular survival. For example, they demonstrate reduced expression of immune-peptide or immune-related genes (Antúnez et al. 2009), reduced re-epithelization in infected ventriculi (Higes et al. 2007; García-Palencia et al. 2010), increased energetic stress (Mayack and Naug 2009; Martín-Hernández et al. 2011) and inhibited melanisation in the hemolymph of infected *Bombyx mori* larvae (Wu et al. 2016).

Table 1 Examples of insects infected by entomopathogenic fungi and fungal-like organisms

	Infected insect species	Literature data
Oomycota		
Aphanomyces laevis	Mosquito larvae	(Patwardhan et al. 2005)
Couchia spp.		(Wallace Martin 2000)
Crypticola spp.		(Frances 1991; Dick 2003; Mendoza et al. 2018)
Lagenidium giganteum		(Golkar et al. 1993; Vyas et al. 2007; Vilela et al. 2019)
Leptolegnia caudata spp.		(Bisht et al. 1996; Montalva et al. 2016; Gutierrez et al. 2017)
Pythium spp.		(Clark et al. 1966; Su et al. 2001; Su 2008; Shen et al. 2019)
Microsporidia		
Anncaliia algerae	*Drosophila melanogaster*	(Sokolova et al. 2020)
Antonospora locustae (Nosema locustae)	Grasshoppers	(Lange 2005; Zhou and Zhang 2009; Gerus et al. 2020)
Nosema adaliae	Two-spotted lady beetle, *Adalia bipunctata* (*Coleoptera: Coccinellidae*)	(Steele and Bjørnson 2014)
Nosema alticae	Flea beetle, *Altica hampei* (Coleoptera: Chrysomelidae)	(Yıldırım and Bekircan 2020)
Nosema bombi, Nosema ceranae, Tubulinosema pampeana	Bumblebees	(Brown 2017)
Nosema carpocapsae	Codling moth, *Cydia pomonella* L	(Zimmermann et al. 2013)
Nosema leptinotarsae	Colorado potato beetle, *Leptinotarsa decemlineata* Say. (Coleoptera: Chrysomelidae).	(Bekircan 2020)
Nosema maddoxi	Brown marmorated stink bug, *Halyomorpha halys* (Hemiptera: Pentatomidae)	(Preston et al. 2020b; Preston et al. 2020a)
Nosema pernyi	The Chinese oak silkworm *Antheraea pernyi* (Lepidoptera: Saturniidae)	(Liu et al. 2020)
Nosema pyrausta	European corn borer *Ostrinia nubilalis,* the Asian corn borer *Ostrinia furnacalis* and the adzuki bean borer *Ostrinia scapulalis*	(Grushevaya et al. 2018; Grushevaya et al. 2020)
TubuliNosema sp.	*Loxostege sticticalis L (*Lepidoptera: Crambidae*)*	(Malysh et al. 2018)
TubuliNosema suzukii	*Drosophila suzukii*	(Biganski et al. 2020)
Vairimorpha (Nosema) ceranae Vairimorpha (Nosema) apis	Honeybee *Apis mellifera*	(Forsgren and Fries 2010; Tokarev et al. 2018; Applegate and Petritz 2020; Chang et al. 2020; Ostroverkhova et al. 2020)
Chytridiomycota		
Myrmicinosporidium durum	Ants from Subfamilies: Myrmicinae, Formicinae and Dolichoderinae	(Sanchez-Peña et al. 1993; Gonçalves et al. 2012; Trigos-Peral et al. 2017)
Nephridiophaga maderae Nephridiophaga archimandrita Nephridiophaga lucihormetica Nephridiophaga blattellae Nephridiophaga blaberi	Madeira cockroach (*Leucophaea maderae*), *Archimandrita tessellate* *Lucihormetica verrucose* German cockroach *Blattella germanica* Death's head cockroach *Blaberus craniifer*	(Radek et al. 2017) (Radek et al. 2011) (Radek and Herth 1999) (Fabel et al. 2000)
Blastocladiomycota		
Coelomycidium simulii	Larval stage of black flies (Diptera): *Simulium asakoae, S. chamlongi, S. chiangmaiense, S. fenestratum, S. feuerborni, S. nakhonense, S. nodosum, S. quinquestriatum, S. tani,* and *S. japonicum*	(Levchenko et al. 1974; Kim 2011; Jitklang et al. 2012; Kim 2015; Adler and McCreadie 2019)
Coelomomyces africanus Coelomomyces angolensis Coelomomyces iliensis Coelomomyces indicus Coelomomyces irani Coelomomyces lairdi	Mosquito larvae from species of *Culex, Culiseta, Aedes, Anopheles, Psorophora,* and *Uranotaenia*	(Scholte et al. 2004; Balaraman et al. 2006; Rueda-Páramo et al. 2017; Dahmana and Mediannikov 2020; Gao et al. 2020)

Table 1 Examples of insects infected by entomopathogenic fungi and fungal-like organisms (Continued)

	Infected insect species	Literature data
Coelomomyces maclaeyae		
Coelomomyces numularius		
Coelomomyces opifex		
Coelomomyces pentangulatus		
Coelomomyces polynesiensis		
Coelomomyces punctatus		
Coelomomyces solomonis		
Coelomomyces santabrancae spp.		
Myiophagus cf. *ucrainicus*	Scale insect: Caribbean black scale, *Saissetia neglecta* (Hemiptera), Caliphornia red scale, *Aonidiella aurantii* (Hemiptera) *Chironomus atracinus* larvae	(Moore and Duncan 2017) (Czeczuga and Godlewska 2001)

The life-cycle of microsporidia is characterized by three phases: an infective or environmental phase (spores), a merogony (proliferative) phase and a sporogonic or spore-forming phase (Keeling and Fast 2002). Microsporidia spores are first ingested or inhaled by the host. The spores then germinate and produce a long and convoluted tubule extrusion apparatus (polar tubule); this tubule distinguishes them from other organisms and plays a crucial role in host cell invasion. The spore approaches the host cell, and its polar tubule is everted to enter the cell and inject its sporoplasm into the host cell cytoplasm. Following successful injection, the proliferative phase begins; this phase includes all cell growth and division from the sporoplasm until spore formation. It is known to include two possible processes: schizogony and merogony. Although very little is understood of schizogony in *Oomycota*, merogony is known to take place as the injected sporoplasm develops into meronts (the proliferative stage); these in turn multiply by binary fission or multiple fission, depending on the species, to form multinucleate plasmodial forms. Finally, these forms undergo sporogony, with the meronts developing into sporonts, which produce two or more sporoblasts; these in turn undergo metamorphosis into spores.

Interestingly, unlike other species, in which sporogony occurs in the presence of plasmalemmal thickening, the microsporidia undergo sporogony to produce diplokaryotic nuclei after meiosis. These *sporonts* can multiply by binary or multiple fission, acquire specialized organelles and become spores. Subsequently, the spores continue the cycle by spreading through the tissues of the host, infecting new cells (Lallo et al. 2016; Han and Weiss 2017; Han et al. 2020; Horta et al. 2020).

Traditionally, *Microsporidia* were classified within the phylum Apicomplexa as *sporozoan parasites*; however, phylogenetic studies suggest that microsporidia are related to fungi, either as a basal branch or sister group (Lee et al. 2008; Han and Weiss 2017). They have also been found to include chitin in the spore wall and use trehalose as a sugar reserve, and studies have noted the presence of closed mitosis, spindle pole bodies and meiosis (Han and Weiss 2017). Although they can infect a great number of domestic and wild animals, the most common hosts are arthropods and fish. Since their discovery in the 1850s as the causative agent of the silkworm disease pebrine, or *pepper disease*, which devastated the silk industry in Europe, these pathogens have demonstrated major economic significance in animal farming, such as nosemosis in beekeeping (*Nosema aApis* and *N. ceranae*) and microsporidiosis in aquaculture (*Loma salmonae* for salmonids and *Thelohania* spp. for shrimp); *Nosema bombycis* has also been found to infect the domesticated silkworm *Bombyx mori*. In addition, some microsporidia act as opportunistic pathogens of humans, especially in patients with AIDS (Weiss 2020).

About 93 genera of microsporidia have the ability to infect insects (Becnel and Andreadis 2014). They are considered as biological control agents for regulating the population of the beet webworm *Loxostege sticticalis*, responsible for serious damage to crops such as soybean, sugar beet, alfalfa and sunflower in northern Eurasia (Malysh et al. 2020). Interestingly, some authors consider microsporidia to have enabled the invasive success of the ladybird *Harmonia axyridis* (Vilcinskas et al. 2015; Verheggen et al. 2017), in which the microsporidia act as a symbiotic "biological weapon" against some predators, like *Coccinella septempunctata* and *Adalia bipunctata* (Ceryngier et al. 2018). Some examples of infected insects are listed in Table 1.

Also, *Antonospora locustae*, formerly known as *Nosema locustae* (syn. *Paranosema locustae*), is commonly used as a biological control agent for grasshoppers in the commercial products Nolo Bait and Semaspore (Lange 2005; Zhou and Zhang 2009; Solter et al. 2012). Like other microsporidial pathogens, *A. locustae* also needs to control the metabolic processes and molecular programmes of the host in order to proliferate. Successful infection was found to suppress the locust gut

microbiota, including the gut bacteria that produce aggregation pheromones, thus preventing host aggregation (Pan et al. 2018).

Most microsporidia species require two successive host generations to complete their life-cycle, and at least three genera, viz. *Amblyospora*, *Hyalinocysta* and *Parathelohania*, also require development in an intermediate copepod host (Becnel and Andreadis 2014). These genera are also highly host- and tissue-specific, with complex developmental sequences comprising unique stages and events (Becnel et al. 2005). They are mostly pathogens of mosquito larvae (Andreadis 1985; Andreadis 2007; Shen et al. 2020a); interestingly, microsporidian infection has also been found to be associated with a reduction in *Plasmodium falciparum* transmission in *Anopheles arabiensis* mosquitoes (Herren et al. 2020).

In contrast, some species of microsporidia exhibit simple life-cycles with a single spore type responsible for oral (horizontal) transmission; these affect only one generation of insects and are not usually host or tissue specific (Becnel et al. 2005). A good example of a genus with this simple life-cycle is *Nosema*, including the species *N. apis* and *N. ceranae*. These species are responsible for most microsporidian infections in bees and other species of *Hymenoptera*, resulting in ecological and economical losses in apiculture (Higes et al. 2006; Forsgren and Fries 2010; Grupe and Alisha Quandt 2020; Ostroverkhova et al. 2020; Paudel et al. 2020). Interestingly, phylogenetic studies have placed the *Nosema* species able to infect bees within a new genus, *Vairimorpha* (Tokarev et al. 2020).

N. apis is believed to have long been a parasite of the European honeybee *Apis mellifera*, whereas *N. ceranae* is presumed to have more recently undergone a host shift to *A. mellifera* from the Asian honeybee *A. ceranae* (Buczek et al. 2020; Ostroverkhova et al. 2020). The parasites exhibit a tissue tropism for honey bee ventriculus (Higes et al. 2020) and are believed to infect their ventricular epithelial cells during digestion; infected bees suffer impaired nutrient absorption, resulting in energy loss and death by starvation (Valizadeh et al. 2020).

Nosema pernyi, a microsporidium infecting the Chinese oak silkworm *Antheraea pernyi*, can enter the gut cell by polar tube infection in most situations. The spore in the polar tube germinates within the alkaline environment of the intestine, inducing destructive chronic disease (Wang et al. 2019; Han et al. 2020). It is also possible for the parasite to exploit the metabolism of the host cell to progress its own life-cycle. A recent study found that successful infection of the honeybee midgut by the microsporidian pathogen *N. ceranae* involves the inhibition of apoptosis (Higes et al. 2013; Kurze et al. 2018); in such cases, the parasite appears to inhibit apoptosis by regulating the genes involved in the

process, as well as in the cell cycle (Martín-Hernández et al. 2017), thus increasing oxidative stress and antioxidant enzyme generation in the gut, and inhibiting the genes involved in the homeostasis and renewal of intestinal tissues (Dussaubat et al. 2012; Liu et al. 2020).

Microsporidial infection can also alter host metabolism and induce both local and systemic innate immune responses (Pan et al. 2018). For example, *N. ceranae* infection is believed to suppress immune defence mechanisms in honey bees; studies have indicated that infection downregulates some immune-related genes, including *abaecin, apidecin, defensin, hymenoptaecin, glucose dehydrogenase (GLD)* and *vitellogenin (Vg)*, and upregulates Jun-related antigen *Jra* and *pi3k* (Chang et al. 2020). Additionally, transcription of the inhibitor of apoptosis protein (*iap-2*) gene was found to be upregulated in *Nosema*-tolerant honeybees (Kurze et al. 2015); it is possible that inhibition of apoptosis might help the parasites optimize their environment and extend the period during which they can grow and differentiate within host cells (Higes et al. 2013). It also appears that pheromone production in worker and queen honeybees to be modified during infection (Dussaubat et al. 2013).

CHYTRIDIOMYCOTA

The *Chytridiomycota* comprise zoosporic fungi phylogenetically related to the true fungi. This group comprises at least three major lineages of chytrids: (1) *Rozella* spp., the earliest diverging lineage in kingdom *Fungi*; (2) *Olpidium brassicae*, the only species classified in *Zygomycota*; and (3) the *core chytrid clade*, encompassing the remaining orders and families and most flagellated fungi, including those of *Chytridiales, Spizellomycetales* p.p., *Monoblepharidales* and *Neocallimastigales* (Barr 2001; James et al. 2006). Chytrids are unicellular, colonial or filamentous organisms with absorptive nutrition, and which reproduce through the production of motile zoospores, typically propelled by a single, posteriorly-directed flagellum (Barr 2001).

Infection begins with the attachment of a motile zoospore to the surface of a host cell, and the formation of a thickened wall around the zoospore. In a successful infection, the encysted zoospore will develop into a mature sporangium, following which, a germ tube typically forms, which enters the host cell via the girdle zone. The zoospores form within the host cell and are released to infect other cells by dehiscence. The attached zoospores completely depend on the host cell for nourishment and their development into mature sporangia (Ibelings et al. 2004).

Traditionally, *Chytridiomycota* have been regarded as aquatic fungi (Ibelings et al. 2004), but most species occur in soil as saprophytes growing on organic material (Digby et al. 2010). In addition, certain obligate

anaerobic species also occur in the digestive tract of herbivores, and these are probably very important in the digestion of cellulose and hemicellulose (Li and Heath 1993; Paul et al. 2018; Saye et al. 2021). A few halophytes have been found in estuaries (Powell et al. 2015). A significant number of species are known as parasites of algae, vascular plants, amphibians, rotifers, tardigrades, protists (Luttrell 1974; Barr 2001; Ibelings et al. 2004) and invertebrates such as nematodes (Betancourt-Román et al. 2016) and insects (Sinha et al. 2016). The first described parasites of vertebrates were isolated from frog skin (Longcore et al. 1999).

The chytrids include one entomopathogenic genus: the monotypic *Myrmicinosporidium* (*M. durum*). That species is an obligate endoparasite of various ant hosts with a global or wide-ranging specific distribution (Lapeva-Gjonova 2014; Trigos-Peral et al. 2017). Infected ants can be distinguished by their darker colour and larger abdomen size. Although the numbers of lentiform spores within an infected ant may initially be very low, they increase during infection, to the point where they can be found throughout the whole body, numbering more than one hundred in a single ant (Sanchez-Peña et al. 1993; Pereira 2004; Espadaler and Santamaria 2012; Gonçalves et al. 2012). Although the spores develop extensively in the ant haemocoel, an infected ant does not exhibit significant differences in life span or behaviour from uninfected ones (Sanchez-Peña et al. 1993); however, several studies have reported the early death of infected ants after hibernation (Espadaler and Santamaria 2012; Giehr and Heinze, 2015). Some examples infected of insects are listed in Table 1.

Recent studies have also placed the nephridiophagids in *Chytridiomycota* (Strassert et al. 2020). The systematic position of the nephridiophagids has been discussed intensively, and they were historically placed with the haplosporidians or microsporidians (Radek et al. 2017). A preliminary molecular analysis placed them within the *Fungi*, close to the *Zygomycota* (Wylezich C, Radek R 2004); however, a later phylogenetic analysis of SSU and LSU rRNA gene sequences placed them in the phylum *Chytridiomycota* (Strassert et al. 2020).

Nephridiophagids are unicellular, spore-forming parasites which infect the Malpighian tubules of insects, especially cockroaches (*Dictyoptera*) and beetles (*Coleoptera*), where they are mainly found in the lumen (Radek et al. 2011). The life-cycle of the nephridiophagids includes a merogony phase with vegetative multinucleate plasmodia that divide into oligonucleate and uninucleate cells. The sporogonial plasmodia form internal, 5–10 μm long, oval, flattened spores, generally with one nucleus, with the residual nuclei of the mother cell remaining in the cytoplasm between them (Radek et al. 2017).

BLASTOCLADIOMYCOTA

Historically, the fungi belonging to the phylum *Blastocladiomycota* were described together with the *Chytridiomycota*; however, recent molecular phylogenetic and ultrastructural research has classified them in their own phylum (James et al. 2006; Porter et al. 2011). Genome-scale trees suggest *Blastocladiomycota* may have diverged around the same time as *Chytridiomycota* (James et al. 2020).

The *Blastocladiomycota* contains only the order *Blastocladiales*; this group contains zoospore-producing true fungi, with well-developed hyphae, closed mitosis, cell walls with β-1–3-glucan, and a secretory vesicle complex known as the Spitzenkörper (James et al. 2020; Roberson 2020). Several of these were once model organisms (e.g., *Allomyces* and *Blastocladiella*; (Cantino et al. 1968; Burke et al. 1972), and obligate parasites of plants and animals (Longcore and Simmons 2012). They are divided into five families (Barr 2001): *Blastocladiaceae* which contains only saprobic species; *Catenariaceae* with both saprobes and pathogens; *Coelomomycetaceae* with pathogens of invertebrates; *Physodermataceae* with obligate parasites of plants; and *Sorochytriaceae* which contains a pathogen infecting tardigrades. Powell (2017) subsequently described a sixth family, *Paraphysodermataceae*, which includes parasites of algae.

The entomopathogenic *Blastocladiomycota* are found in the genera *Coelomomyces* and *Coelomycidium* of *Coelomomycetaceae*. These genera include approximately 70 species pathogenic to insects such as mosquitoes and flies (Powell 2017; Shen et al. 2020a). Some examples of insects infected by *Blastocladiomycota* are listed in Table 1.

Coelomycidium simulii is a widespread species pathogenic to black flies, being mostly found in the larvae, and rarely in pupae and adults. Infected larvae have large numbers of minute, spherical thalli throughout the body cavity. These thalli give rise to spores that are released into the water column after the death of the host. An intermediate host might be required to complete the life-cycle (McCreadie and Adler 1999; Boucias and Pendland 2012).

The species of *Coelomomyces* are aquatic fungi that act as obligate parasites. They are best known as pathogens of mosquito larvae; however, some might infect aquatic dipteran insects including those of the *Psychodidae*, *Chironomidae*, *Simuliidae* and *Tabanidae* (Scholte et al. 2004; Shen et al. 2020a). Although they are probably not host specific, they nevertheless have relatively restricted host ranges (Federici 1981). The life-cycle of *Coelomomyces* is complex and comprises obligatory development in an intermediate microcrustacean host (cyclopoid copepods, harpacticoid copepods, or ostracods) and two mosquito generations. Infection is also known to cause significant epizootics which can persist

in larval populations for several years, resulting in mortality rates greater than 50%, and often higher than 90% (Scholte et al. 2004). Moreover, the fungus might remain inside the insect, passing through the larval and pupal stages to mature in the ovaries of adult females, where the sporangia start to produce zoospores. After the first blood meal, these zoospore-filled sporangia are 'laid' by the female mosquito, ready to infect new larvae, instead of eggs (Araújo and Hughes 2016). Due to the relatively specific host range of the fungus, being generally restricted to mosquitoes, and the devastating effects of natural epizootics on larval populations, it has been proposed as a suitable biocontrol of mosquito populations. However, these advantages have to be weighed against its unpredictable infection rate, complicated life-cycle and problem with mass production (Scholte et al. 2004).

The fungi placed in *Myiophagus* were initially considered to belong to *Chytridiomycota* (Blackwell and Powell 2020); however, based on their morphological differences, they are now placed in *Blastocladiomycota* (Humber 2012; James et al. 2014; Powell 2017). Species of this genus infect leeches (Czeczuga et al. 2003), scale insects, mealybugs, beetle larvae, and dipteran pupae (Karling 1948) resulting in chytridiosis in subtropical regions (Tanada and Kaya 2012). As *Myiophagus* requires free water for dissemination (Cole 2012), infection takes place shortly after or during heavy rainfall. In such conditions, motile zoospores are released from resting sporangia and these swim through the water film that forms over "drip leaves" to find potential hosts; similar to other aquatic fungi, the zoospores are believed to be guided to the host by specific chemoreceptors (Luisa 2012).

CONCLUSION

Biological control is an effective and environmentally-acceptable alternative to chemical insect control methods. Entomopathogenic fungi and their metabolites are believed to represent potential alternatives to chemical pesticides. When used as biocontrol agents, entomopathogens might offer several advantages over conventional insecticides, such as high efficiency and selectivity, safety for beneficial organisms, reduction of residues in the environment, and increased biodiversity in human-managed ecosystems. However, relatively little is known of their effectiveness in field applications and the potential side effects of their use. It is also important to emphasise that due to the global distribution and variety of insects, many as yet unknown entomopathogenic fungi may exist; as such, there is a clear need for further comprehensive studies of the biology and ecology of entomopathogenic organisms and of the mechanisms underlying their action on insect hosts.

Author details
[1]Witold Stefański Institute of Parasitology, Polish Academy of Sciences, Twarda 51/55, 00-818 Warsaw, Poland. [2]Biomibo, Strzygłowska 15, 04-872 Warsaw, Poland.

References
Adler, P.H.; McCreadie, J.W. Black Flies (Simuliidae). In Medical and Veterinary Entomology, 3rd ed.; Mullen, G.R., Durden, L.A., Eds.; Elsevier Inc.: Amsterdam, The Netherlands, 2019; pp. 237–259

Andreadis TG (1985) Life cycle, epizootiology, and horizontal transmission of *Amblyospora* (Microspora: Amblyosporidae) in a univoltine mosquito, *Aedes stimulans*. Journal of Invertebrate Pathology 46:31–46. https://doi.org/10.1016/0022-2011(85)90127-2

Andreadis TG (2007) Microsporidian parasites of mosquitoes. Journal of the American Mosquito Control Association 23:3–29. https://doi.org/10.2987/8756-971x

Antúnez K, Martín-Hernández R, Prieto L, Meana A, Zunino P, Higes M (2009) Immune suppression in the honey bee (*Apis mellifera*) following infection by *Nosema ceranae* (microsporidia). Environmental Microbiology 11:2284–2290. https://doi.org/10.1111/j.1462-2920.2009.01953.x

Applegate JR, Petritz OA (2020) Common and emerging infectious diseases of honeybees (*Apis mellifera*). The Veterinary Clinics of North America. Exotic Animal Practice 23:285–297. https://doi.org/10.1016/j.cvex.2020.01.001

Araújo JPM, Hughes DP (2016) Diversity of entomopathogenic fungi. Which groups conquered the insect body? In: Lovett B, St. Leger RJ (eds) advances in genetics. Academic press, pp 1–39

Balaraman K, Vijayan V, Geetha I (2006) Biodiversity of mosquitocidal fungi and Actinomycetes. Biodivers Life to our mother earth :355–370

Barr DJS (2001) Chytridiomycota. In: McLaughlin DJ, McLaughlin EG, Lemke PA (eds) Systematics and evolution. Springer, Berlin Heidelberg, Berlin, Heidelberg, pp 93–112

Beakes GW, Glockling SL, Sekimoto S (2012) The evolutionary phylogeny of the oomycete 'fungi'. Protoplasma 249:3–19. https://doi.org/10.1007/s00709-011-0269-2

Beakes GW, Thines M (2017) Hyphochytriomycota and Oomycota. In: Archibald JM, Simpson AGB, Slamovits CH (eds) Handbook of the Protists, Second edn. Springer International Publishing, Cham, pp 435–505

Becnel JJ, Andreadis TG (2014) Microsporidia in insects. In: microsporidia: pathogens of opportunity: first edition. Pp 521–570

Becnel JJ, White SE, Shapiro AM (2005) Review of microsporidia-mosquito relationships: from the simple to the complex. Folia Parasitologica (Praha) 52: 41–50. https://doi.org/10.14411/fp.2005.006

Bekircan Ç (2020) Assignment of *Vairimorpha leptinotarsae comb. nov.* on the basis of molecular characterization of *Nosema leptinotarsae* Lipa, 1968 (microsporidia: *Nosema*tidae). Parasitology 147:1019–1025. https://doi.org/10.1017/S0031182020000669

Betancourt-Román CM, O'Neil CC, James TY (2016) Rethinking the role of invertebrate hosts in the life cycle of the amphibian chytridiomycosis pathogen. Parasitology 143(13):1723–1729. https://doi.org/10.1017/S0031182016001360

Biganski S, Wennmann JT, Vossbrinck CR, Kaur R, Jehle JA, Kleespies RG (2020) Molecular and morphological characterisation of a novel microsporidian species, *Tubulinosema suzukii*, infecting *Drosophila suzukii* (Diptera: Drosophilidae). Journal of Invertebrate Pathology 174. https://doi.org/10.1016/j.jip.2020.107440

Bisht GS, Joshi C, Khulbe RD (1996) Watermolds: potential biological control agents of malaria vector *anopheles culicifacies*. Current Science 70:393–395

Blackwell M (2010) Fungal evolution and taxonomy. BioControl 55(1):7–16. https://doi.org/10.1007/s10526-009-9243-8

Blackwell WH, Powell MJ (2020) A nomenclatural and systematic note on the genus Myiophagus (ex Chytridiomycota). Phytologia 102:5–8

Boucias DG, Pendland JC (2012) Principles of insect pathology Springer US

Brown MJF (2017) Microsporidia: an emerging threat to bumblebees? Trends in Parasitology 33:754–762. https://doi.org/10.1016/j.pt.2017.06.001

Buczek K, Trytek M, Deryło K, Borsuk G, Rybicka-Jasińska K, Gryko D, Cytryńska M, Tchórzewski M (2020) Bioactivity studies of porphyrinoids against microsporidia isolated from honeybees. Scientific Reports 10. https://doi.org/10.1038/s41598-020-68420-5

Burke DJ, Seale TW, McCarthy BJ (1972) Protein and ribonucleic acid synthesis during the diploid life cycle of *Allomyces arbuscula*. Journal of Bacteriology 110:1065–1072. https://doi.org/10.1128/jb.110.3.1065-1072.1972

Cantino EC, Truesdell LC, Shaw DS (1968) Life history of the motile spore of *Blastocladiella emersonii*: a study in cell differentiation. Journal of the Elisha Mitchell Scientific Society 84:125–146

Ceryngier P, Nedvěd O, Grez AA, Riddick EW, Roy HE, San Martin G, Steenberg T, Veselý P, Zaviezo T, Zúñiga-Reinoso Á, Haelewaters D (2018) Predators and parasitoids of the harlequin ladybird, *Harmonia axyridis*, in its native range and invaded areas. Biological Invasions 20:1009–1031. https://doi.org/10.1007/s10530-017-1608-9

Chang ZT, Ko CY, Yen MR, Chen YW, Nai YS (2020) Screening of differentially expressed microsporidia genes from *Nosema ceranae* infected honey bees by suppression subtractive hybridization. Insects. https://doi.org/10.3390/insects11030199

Clark TB, Kellen WR, Lindegren JE, Sanders RD (1966) *Pythium sp.* (Phycomycetes: Pythiales) pathogenic to mosquito larvae. Journal of Invertebrate Pathology 8(3):351–354. https://doi.org/10.1016/0022-2011(66)90049-8

Cole GT (2012) Biology of conidial fungi. Elsevier Science

Corradi N (2015) Microsporidia: eukaryotic intracellular parasites shaped by gene loss and horizontal gene transfers. Annual Review of Microbiology 69:167–183. https://doi.org/10.1146/annurev-micro-091014-104136

Corradi N, Keeling PJ (2009) Microsporidia: a journey through radical taxonomical revisions. Fungal Biology Reviews 23:1–8

Czeczuga B, Godlewska A (2001) Aquatic insects as vectors of aquatic zoosporic fungi parasitic on fishes. Acta Ichthyologica et Piscatoriat 31(2):87–104. https://doi.org/10.3750/AIP2001.31.2.07

Czeczuga B, Kiziewicz B, Godlewska A (2003) Zoosporic fungi growing on leeches (Hirudinea). Polish Journal of Environmental Studies 12:361–369

Dahmana H, Mediannikov O (2020) Mosquito-borne diseases emergence/resurgence and how to effectively control it biologically. Pathogens 9:310. https://doi.org/10.3390/pathogens9040310

Dick, M. W. (2001). *Straminipilous fungi: Systematics of the peronosporomycetes, including accounts of the marine straminipilous protists, the plasmodiophorids, and similar organisms*. Dordrecht: Kluwer Academic Publishers.

Digby AL, Gleason FH, McGee PA (2010) Some fungi in the Chytridiomycota can assimilate both inorganic and organic sources of nitrogen. Fungal Ecology 3: 261–266. https://doi.org/10.1016/j.funeco.2009.11.002

Dobrowolski JW, Bedla D, Czech T, Gambuś F, Górecka K, Kiszczak W, Kuźniar T, Mazur R, Nowak A, Śliwka M, Tursunov O, Wagner A, Wieczorek J, Zabochnicka-Światek M (2018) Integrated innovative biotechnology for optimization of environmental bioprocesses and a green economy. Optimization and Applicability of Bioprocesses, In, pp 27–71

Dowd PF, Johnson ET, Pinkerton TS (2007) Oral toxicity of β-N-acetyl hexosaminidase to insects. Journal of Agricultural and Food Chemistr 55: 3421–3428. https://doi.org/10.1021/jf063562w

Dussaubat C, Brunet JL, Higes M, Colbourne JK, Lopez J, Choi JH, Martín-Hernández R, Botías C, Cousin M, McDonnell C, Bonnet M, Belzunces LP, Moritz RFA, Le Conte Y, Alaux C (2012) Gut pathology and responses to the microsporidium *Nosema ceranae* in the honey bee *Apis mellifera*. PLoS One 7. https://doi.org/10.1371/journal.pone.0037017

Dussaubat C, Maisonnasse A, Crauser D, Beslay D, Costagliola G, Soubeyrand S, Kretzchmar A, Le Conte Y (2013) Flight behavior and pheromone changes associated to *Nosema ceranae* infection of honey bee workers (*Apis mellifera*) in field conditions. Journal of Invertebrate Pathology 113(1):42–51. https://doi.org/10.1016/j.jip.2013.01.002

Espadaler X, Santamaria S (2012) Ecto- and endoparasitic fungi on ants from the Holarctic region. Psyche (London). 2012:1–10. https://doi.org/10.1155/2012/168478

Fabel P, Radek R, Storch V (2000) A new spore-forming protist, *Nephridiophaga blaberi* sp. nov., in the Death's head cockroach *Blaberus craniifer*. European Journal of Protistology 36:387–395. https://doi.org/10.1016/S0932-4739(00)80044-9

Federici B (1981) Mosquito control by the fungi Culicinomyces, Lagenidium and Coelomomyces. Microbial control of pests and plant diseases. 1970-1980 v. 1981.:555–572

Forsgren E, Fries I (2010) Comparative virulence of *Nosema ceranae* and *Nosema apis* in individual European honey bees. Veterinary Parasitology 170:212–217. https://doi.org/10.1016/j.vetpar.2010.02.010

Frances SP (1991) Pathogenicity, host range and temperature tolerance of *Crypticola clavulifera* (oomycetes: Lagenidiales) in the laboratory. Journal of the American Mosquito Control Association 7:504–506

Frances SP, Sweeney AW, Humber RA (1989) *Crypticola clavulifera gen. Et sp. nov.* and *Lagenidium giganteum*: oomycetes pathogenic for dipterans infesting leaf axils in an Australian rain forest. Journal of Invertebrate Pathology 54: 103–111. https://doi.org/10.1016/0022-2011(89)90146-8

Gani M, Hassan T, Saini P, Gupta RK, Bali K (2019) Molecular phylogeny of Entomopathogens. In: Khan M, Ahmad W (eds) Microbes for sustainable insect Pest management. Sustainability in Plant and Crop Protection. Springer, Cham, pp 43–113. https://doi.org/10.1007/978-3-030-23045-6_3

Gao H, Cui C, Wang L, Jacobs-Lorena M, Wang S (2020) Mosquito microbiota and implications for disease control. Trends in Parasitology 36:98–111. https://doi.org/10.1016/j.pt.2019.12.001

García-Palencia P, Martín-Hernández R, González-Porto AV, Marin P, Meana A, Higes M (2010) Natural infection by *Nosema ceranae* causes similar lesions as in experimentally infected caged-worker honey bees (*Apis mellifera*). Journal of Apicultural Research 49(3):278–283. https://doi.org/10.3896/IBRA.1.49.3.08

Gerus A, Ignatieva A, Tokarev У (2020) Prevalence rates of microsporidia in locusts and grasshoppers in South-Western Russia. BIO Web of Conferences 21:00010. https://doi.org/10.1051/bioconf/20202100010

Giehr J, Heinze J, Schrempf A (2015) The Ant Cardiocondyla elegans as Host of the Enigmatic Endoparasitic Fungus *Myrmicinosporidium durum*", Psyche: A Journal of Entomology, 2015:4, Article ID 364967. https://doi.org/10.1155/2015/364967

Golkar L, Lebrun RA, Ohayon H, Gounon P, Papierok B, Brey PT (1993) Variation of larval susceptibility to *Lagenidium giganteum* in three mosquito species. Journal of Invertebrate Pathology 62:1–8. https://doi.org/10.1006/jipa.1993.1066

Gonçalves C, Patanita I, Espadaler X (2012) Substantial, and significant, expansion of ant hosts range for *Myrmicinosporidium hölldobler*, 1933 (fungi). Insectes Sociaux 59(3):395–399. https://doi.org/10.1007/s00040-012-0232-z

Grupe AC, Alisha Quandt C (2020) A growing pandemic: a review of *Nosema* parasites in globally distributed domesticated and native bees. PLoS Pathogens 16(6):16. https://doi.org/10.1371/journal.ppat.1008580

Grushevaya I, Ignatieva A, Tokarev Y (2020) Susceptibility of three species of the genus *Ostrinia* (Lepidoptera: Crambidae) to *Nosema* pyrausta (microsporidia: Nosematida). BIO Web of Conferences 21:00040. https://doi.org/10.1051/bioconf/20202100040

Grushevaya IV, Ignatieva AN, Malysh SM, Senderskiy IV, Zubarev IV, Kononchuk AG (2018) Spore dimorphism in *Nosema pyrausta* (microsporidia, Nosematidae): from morphological evidence to molecular genetic verification. Acta Protozoologica 57:49–52. https://doi.org/10.4467/16890027AP.18.004.8398

Gutierrez AC, Rueda Páramo ME, Falvo ML, López Lastra CC, García JJ (2017) *Leptolegnia chapmanii* (Straminipila: Peronosporomycetes) as a future biorational tool for the control of *Aedes aegypti* (L.). Acta Tropica 169:112–118. https://doi.org/10.1016/j.actatropica.2017.01.021

Haag KL, James TY, Pombert JF, Larsson R, Schaer TMM, Refardt D, Ebert D (2014) Evolution of a morphological novelty occurred before genome compaction in a lineage of extreme parasites. Proceedings of the National Academy of Sciences of the United States of America 111:15480–15485. https://doi.org/10.1073/pnas.1410442111

Haag KL, Pombert JF, Sun Y, De Albuquerque NRM, Batliner B, Fields P, Lopes TF, Ebert D (2019) Microsporidia with vertical transmission were likely shaped by nonadaptive processes. Genome Biology and Evolution 12:3599–3614. https://doi.org/10.1093/gbe/evz270

Hallmon CF, Schreiber ET, Vo T, Bloomquist MA (2000) Field trials of three concentrations of Laginex™ as biological larvicide compared to Vectobac™-12as as a biocontrol agent for *Culex quinquefasciatus*. Journal of the American Mosquito Control Association 16:5–8

Han B, Takvorian PM, Weiss LM (2020) Invasion of host cells by microsporidia. Frontiers in Microbiology. 11. https://doi.org/10.3389/fmicb.2020.00172

Han B, Weiss LM (2017) Microsporidia: obligate intracellular pathogens within the fungal kingdom. Microbiology Spectrum5. https://doi.org/10.1128/microbiolspec.funk-0018-2016

Hassett BT, Thines M, Buaya A, Ploch S, Gradinger R (2019) A glimpse into the biogeography, seasonality, and ecological functions of arctic marine Oomycota. IMA Fungus 10. https://doi.org/10.1186/s43008-019-0006-6

Herren, J.K., Mbaisi, L., Mararo, E. *et al.* A microsporidian impairs *Plasmodium falciparum* transmission in *Anopheles arabiensis* mosquitoes. Nat Commun11, 2187 (2020). https://doi.org/10.1038/s41467-020-16121-y

Higes M, García-Palencia P, Martín-Hernández R, Meana A (2007) Experimental infection of *Apis mellifera* honeybees with *Nosema ceranae* (microsporidia). Journal of Invertebrate Pathology 94(3):211–217. https://doi.org/10.1016/j.jip.2006.11.001

Higes M, García-Palencia P, Urbieta A, Nanetti A, Martín-Hernández R (2020) *Nosema Apis* and *Nosema ceranae* tissue tropism in worker honey bees (*Apis mellifera*). Veterinary Pathology 57:132–138. https://doi.org/10.1177/0300985819864302

Higes M, Juarranz Á, Dias-Almeida J, Lucena S, Botías C, Meana A, García-Palencia P, Martín-Hernández R (2013) Apoptosis in the pathogenesis of *Nosema ceranae* (microsporidia: Nosematidae) in honey bees (*Apis mellifera*). Environmental Microbiology Rep 5(4):530–536. https://doi.org/10.1111/1758-2229.12059

Higes M, Martín R, Meana A (2006) *Nosema ceranae*, a new microsporidian parasite in honeybees in Europe. Journal of Invertebrate Pathology 92:93–95. https://doi.org/10.1016/j.jip.2006.02.005

Horta MF, Andrade LO, Martins-Duarte ÉS, Castro-Gomes T (2020) Cell invasion by intracellular parasites - the many roads to infection. Journal of Cell Science, 133(4), https://doi.org/10.1242/jcs.232488

Humber RA (2008) Evolution of entomopathogenicity in fungi. Journal of Invertebrate Pathology 98:262–266. https://doi.org/10.1016/j.jip.2008.02.017

Humber RA (2012) Identification of entomopathogenic fungi. Manual of Techniques in Invertebrate Pathology, In

Ibelings BW, De Bruin A, Kagami M, Rijkeboer M, Brehm M, Van Donk E (2004) Host parasite interactions between freshwater phytoplankton and chytrid fungi (Chytridiomycota). Journal of Phycology 40(3):437–453. https://doi.org/10.1111/j.1529-8817.2004.03117.x

James TY, Letcher PM, Longcore JE, Mozley-Standridge SE, Porter D, Powell MJ, Griffith GW, Vilgalys R (2006) A molecular phylogeny of the flagellated fungi (Chytridiomycota) and description of a new phylum (Blastocladiomycota). Mycologia 98(6):860–871. https://doi.org/10.3852/mycologia.98.6.860

James TY, Porter TM, Martin WW (2014) 7 Blastocladiomycota. In: McLaughlin DJ, Spatafora JW (eds) Systematics and evolution: part a. Springer, Berlin Heidelberg, Berlin, Heidelberg, pp 177–207

James TY, Stajich JE, Hittinger CT, Rokas A (2020) Toward a fully resolved fungal tree of life. Annual Review of Microbiology 74(1):291–313. https://doi.org/10.1146/annurev-micro-022020-051835

Aronski, S., Mascarin, G.M. 2016. Mass production of fungal entomopathogens. In: Lacey, L.A., editor. Microbial Control of Insect and Mite Pests from Theory to Practice. Yakima, WA: IP Consulting International. p. 141-155. https://doi.org/10.1016/B978-0-12-803527-6.00009-3.

Jitklang S, Ahantarig A, Kuvangkadilok C, Baimai V, Adler PH (2012) Parasites of larval black flies (Diptera: Simuliidae) in Thailand. Songklanakarin Journal of Science and Technology 34:597–599

Joop G, Vilcinskas A. Coevolution of parasitic fungi and insect hosts. Zoology (Jena). 2016 Aug;119(4):350-8. https://doi.org/10.1016/j.zool.2016.06.005.

Kamoun S, Furzer O, Jones JDG, Judelson HS, Ali GS, Dalio RJD, Roy SG, Schena L, Zambounis A, Panabières F, Cahill D, Ruocco M, Figueiredo A, Chen XR, Hulvey J, Stam R, Lamour K, Gijzen M, Tyler BM, Grünwald NJ, Mukhtar MS, Tomé DFA, Tör M, Van Den Ackerveken G, Mcdowell J, Daayf F, Fry WE, Lindqvist-Kreuze H, Meijer HJG, Petre B, Ristaino J, Yoshida K, Birch PRJ, Govers F (2015) The top 10 oomycete pathogens in molecular plant pathology. Molecular Plant Pathology 16(4):413–434. https://doi.org/10.1111/mpp.12190

Karabörklü, S., Azizoglu, U. & Azizoglu, Z.B. Recombinant entomopathogenic agents: a review of biotechnological approaches to pest insect control. *World J Microbiol Biotechnol*34, 14 (2018). https://doi.org/10.1007/s11274-017-2397-0

Karling JS (1948) Chytridiosis of scale insects. American Journal of Botany 35:246. https://doi.org/10.2307/2437955

Karlovsky P, Fartmann B (1992) Genetic code and phylogenetic origin of oomycetous mitochondria. Journal of Molecular Evolution 34(3):254–258. https://doi.org/10.1007/BF00162974

Kaur R, Stoldt M, Jongepier E, Feldmeyer B, Menzel F, Bornberg-Bauer E, Foitzik S (2019) Ant behaviour and brain gene expression of defending hosts depend on the ecological success of the intruding social parasite. Philosophical Transactions of the Royal Society B: Biological Sciences 1;374(1769): 20180192https://doi.org/10.1098/rstb.2018.0192

Keeling P (2009) Five questions about microsporidia. PLoS Pathogens 5(9): e1000489. https://doi.org/10.1371/journal.ppat.1000489

Keeling PJ, Fast NM (2002) Microsporidia: biology and evolution of highly reduced intracellular parasites. Annual Review of Microbiology 56(1):93–116. https://doi.org/10.1146/annurev.micro.56.012302.160854

Kepler RM, Sung GH, Harada Y, Tanaka K, Tanaka E, Hosoya T, Bischoff JF, Spatafora JW (2012) Host jumping onto close relatives and across kingdoms by Tyrannicordyceps (Clavicipitaceae) gen. Nov. and Ustilaginoidea (Clavicipitaceae). American Journal of Botany 99:552–561. https://doi.org/10.3732/ajb.1100124

Kim KH, Kabir E, Jahan SA (2017) Exposure to pesticides and the associated human health effects. Science of the Total Environment 575:525–535. https://doi.org/10.1016/j.scitotenv.2016.09.009

Kim SK (2011) Redescription of *Simulium (Simulium) japonicum* (Diptera: Simuliiae) and its entomopathogenic fungal symbionts. Entomological Research 41(5): 208–210. https://doi.org/10.1111/j.1748-5967.2011.00336.x

Kim S-K (2015) Morphology and ecological notes on the larvae and pupae of *Simulium* (Simulium) from Korea. Animal Systematics, Evolution and Diversity 31(4):209–246. https://doi.org/10.5635/ased.2015.31.4.209

Klinter S, Bulone V, Arvestad L. Diversity and evolution of chitin synthases in oomycetes (Straminipila: Oomycota). Mol Phylogenet Evol. 2019 Oct;139: 106558. https://doi.org/10.1016/j.ympev.2019.106558

Kurze C, Le Conte Y, Dussaubat C, Erler S, Kryger P, Lewkowski O, et al. (2015) *Nosema* Tolerant Honeybees (*Apis mellifera*) Escape Parasitic Manipulation of Apoptosis. PLoS ONE 10(10):e0140174. https://doi.org/10.1371/journal.pone.0140174

Kurze C, Le Conte Y, Kryger P, Lewkowski O, Müller T, Moritz RFA (2018) Infection dynamics of *Nosema ceranae* in honey bee midgut and host cell apoptosis. Journal of Invertebrate Pathology 154:1–4. https://doi.org/10.1016/j.jip.2018.03.008

Lacey, L.A.. (2017). Entomopathogens Used as Microbial Control Agents. https://doi.org/10.1016/B978-0-12-803527-6.00001-9.

Lacey LA, Frutos R, Kaya HK, Vail P (2001) Insect pathogens as biological control agents: do they have a future? Biological Control 21:230–248. https://doi.org/10.1006/bcon.2001.0938

Lacey LA, Grzywacz D, Shapiro-Ilan DI, Frutos R, Brownbridge M, Goettel MS (2015) Insect pathogens as biological control agents: Back to the future. Journal of Invertebrate Pathology 132:1–41. https://doi.org/10.1016/j.jip.2015.07.009

Lallo MA, Vidoto Da Costa LF, Alvares-Saraiva AM, et al. Culture and propagation of microsporidia of veterinary interest, The Journal of Veterinary Medical Science 78(2):171-176. https://doi.org/10.1292/jvms.15>

Lange CE (2005) The host and geographical range of the grasshopper pathogen *Paranosema (Nosema) locustae* revisited. Journal of Orthoptera Research 14(2): 137–141. https://doi.org/10.1665/1082-6467(2005)14[137:THAGRO]2.0.CO;2

Lapeva-Gjonova A (2014) *Cataglyphis aenescens* - a newly discovered ant host of the fungal parasite *Myrmicinosporidium durum*. Bulgarian Journal of Agricultural Science 20:157–159

Lee SC, Corradi N, Byrnes EJ, Torres-Martinez S, Dietrich FS, Keeling PJ, Heitman J (2008) Microsporidia evolved from ancestral sexual fungi. Current Biology 18(21):1675–1679. https://doi.org/10.1016/j.cub.2008.09.030

Leoro-Garzon P, Gonedes AJ, Olivera IE (2019) Tartar a (2019) oomycete metabarcoding reveals the presence of *Lagenidium spp*. In phytotelmata. PeerJ. https://doi.org/10.7717/peerj.7903

Levchenko NG, Dubitskii AM, Vakker VG (1974) Enteropathogenic fungus *Coelomycidium simulii* (Phycomycetes, Chitridiales) in the larva of black flies of the genus *Odagmia* (Diptera, Simuliidae) in Kazakhstan. Meditsinskaya Parazitologiya i Parazitarnye Bolezni (Mosk) 43:110–112

Li J, Heath IB (1993) Chytridiomycetous gut fungi, oft overlooked contributors to herbivore digestion. Canadian Journal of Microbiology 39(11):1003–1013. https://doi.org/10.1139/m93-153

Litwin A, Nowak M, Różalska S (2020) Entomopathogenic fungi: unconventional applications. Reviews in Environmental Science and Bio/Technology 19(1): 23–42. https://doi.org/10.1007/s11157-020-09525-1

Liu W, Wang Y, Leng Z, Wang Q, Duan X, Luo Y, Jiang Y, Qin L (2020) Nitric oxide plays a crucial role in midgut immunity under microsporidian infection in *Antheraea pernyi*. Molecular Immunology 126:65–72. https://doi.org/10.1016/j.molimm.2020.07.018

Longcore JE, Pessier AP, Nichols DK (1999) *Batrachochytrium dendrobatidis gen. Et sp. nov.*, a chytrid pathogenic to amphibians. Mycologia 91(2):219–227. https://doi.org/10.2307/3761366

Longcore JE, Simmons DR. 2012. Blastocladiomycota (8 pp.). In: Encyclopedia of Life Sciences. John Wiley & Sons, Ltd: Chichester. www.els.net. http://dx.doi.org/10.1002/9780470015902.a0000349.pub3

Luisa BG (2012) Insect-fungus interactions. Academic Press

Luttrell ES (1974) Parasitism of fungi on vascular plants. Mycologia 66:1–15. https://doi.org/10.1080/00275514.1974.12019567

Malysh J, Tokarev Y, Malysh S, Xingfu J, Frolov A (2020) Biodiversity of beet webworm microsporidia in Eurasia. In, BIO Web of Conferences, p 00019

Malysh JM, Ignatieva AN, Artokhin KS, Frolov AN, Tokarev YS (2018) Natural infection of the beet webworm *Loxostege sticticalis L.* (Lepidoptera: Crambidae) with three microsporidia and host switching in *Nosema ceranae*. Parasitology Research 117:3039–3044. https://doi.org/10.1007/s00436-018-5987-3

Martin WW, Warren A (2020) *Periplasma, gen. Nov.*, a new oomycete lineage with isogamous sexual reproduction. Mycologia 1–14 . https://doi.org/10.1080/00275514.2020.1797385

Martín-Hernández R, Botías C, Barrios L, Martínez-Salvador A, Meana A, Mayack C, Higes M (2011) Comparison of the energetic stress associated with experimental *Nosema ceranae* and *Nosema apis* infection of honeybees (*Apis mellifera*). Parasitology Research 109(3):605–612. https://doi.org/10.1007/s00436-011-2292-9

Martín-Hernández R, Higes M, Sagastume S, Juarranz Á, Dias-Almeida J, Budge GE, Meana A, Boonham N (2017) Microsporidia infection impacts the host cell's cycle and reduces host cell apoptosis. PLoS One 12. https://doi.org/10.1371/journal.pone.0170183

Mayack C, Naug D (2009) Energetic stress in the honeybee *Apis mellifera* from *Nosema ceranae* infection. Journal of Invertebrate Pathology 100(3):185–188. https://doi.org/10.1016/j.jip.2008.12.001

McCreadie JW, Adler PH (1999) Parasites of larval black flies (Diptera: Simuliidae) and environmental factors associated with their distributions. Invertebrate Biology 118:310. https://doi.org/10.2307/3227000

McInnis T, Domnas A (1973) The properties of trehalase from the mosquito-parasitizing water mold, *Lagenidium sp*. Journal of Invertebrate Pathology 22: 313–320. https://doi.org/10.1016/0022-2011(73)90157-2

Mendoza L, Vilela R (2013) The mammalian pathogenic oomycetes. Current Fungal Infection Reports 7:198–208. https://doi.org/10.1007/s12281-013-0144-z

Mendoza L, Vilela R, Humber RA (2018) Taxonomic and phylogenetic analysis of the *Oomycota* mosquito larvae pathogen *Crypticola clavulifera*. Fungal Biology 122:847–855. https://doi.org/10.1016/j.funbio.2018.04.010

Montalva C, dos Santos K, Collier K, Rocha LFN, Fernandes ÉKK, Castrillo LA, Luz C, Humber RA (2016) First report of *Leptolegnia chapmanii* (Peronosporomycetes: Saprolegniales) affecting mosquitoes in Central Brazil. Journal of Invertebrate Pathology 136:109–116. https://doi.org/10.1016/j.jip.2016.03.012

Moonjely S, Barelli L, Bidochka MJ (2016) Insect pathogenic fungi as endophytes. Advanced Genetics 94:107–135. https://doi.org/10.1016/bs.adgen.2015.12.004

Moore SD, Duncan LW (2017) Microbial control of insect and mite pests of citrus. In: Lacey LA (ed) microbial control of insect and mite pests: from theory to practice. Academic press, pp 283–298.

Mužinić V, Želježić D (2018) Non-target toxicity of novel insecticides. Archives of Industrial Hygiene and Toxicology 69:86–102

Neuwirthová N, Trojan M, Svobodová M, Vašíčková J, Šimek Z, Hofman J, Bielská L (2019) Pesticide residues remaining in soils from previous growing season(s) - can they accumulate in non-target organisms and contaminate the food web? Science of the Total Environment 646:1056–1062. https://doi.org/10.1016/j.scitotenv.2018.07.357

Nikoh N, Fukatsu T (2000) Interkingdom host jumping underground: phylogenetic analysis of entomoparasitic fungi of the genus Cordyceps. Molecular Biology and Evolution 17(4):629–638. https://doi.org/10.1093/oxfordjournals.molbev.a026341

Olivera IE, Fins KC, Rodriguez SA, Abiff SK, Tartar JL, Tartar A (2016) Glycoside hydrolases family 20 (GH20) represent putative virulence factors that are shared by animal pathogenic oomycetes, but are absent in phytopathogens. BMC Microbiology 16(1):1–11. https://doi.org/10.1186/s12866-016-0856-7

Ostroverkhova NV, Konusova OL, Kucher AN, Kireeva TN, Rosseykina SA (2020) Prevalence of the microsporidian Nosema spp. in honey bee populations (*Apis mellifera*) in some ecological regions of North Asia. Veterinary. Sciences 7(3):111. https://doi.org/10.3390/vetsci7030111

Pan G, Bao J, Ma Z, Song Y, Han B, Ran M, Li C, Zhou Z (2018) Invertebrate host responses to microsporidia infections. Developmental & Comparative Immunology 83:104–113. https://doi.org/10.1016/j.dci.2018.02.004

Patwardhan A, Gandhe R, Ghole V, Mourya D (2005) Larvicidal activity of the fungus *Aphanomyces* (oomycetes: Saprolegniales) against *Culex quinquefasciatus*. Journal of Communicable Diseases 37:269–274

Paudel S, Sah LP, Devkota M, Poudyal V, Prasad PVV, Reyes MR (2020) Conservation agriculture and integrated pest management practices improve yield and income while reducing labor, pests, diseases and chemical pesticide use in smallholder vegetable farms in Nepal. Sustain 12. https://doi.org/10.3390/VETSCI7030111

Paul SS, Bu D, Xu J, Hyde KD, Yu Z (2018) A phylogenetic census of global diversity of gut anaerobic fungi and a new taxonomic framework. Fungal Diversity 89. https://doi.org/10.1007/s13225-018-0396-6

Pereira RM (2004) Occurrence of *Myrmicinosporidium durum* in red imported fire ant, *Solenopsis invicta*, and other new host ants in eastern United States. Journal of Invertebrate Pathology 86(1-2):38–44. https://doi.org/10.1016/j.jip.2004.03.005

Porter TM, Martin W, James TY, Longcore JE, Gleason FH, Adler PH, Letcher PM, Vilgalys R (2011) Molecular phylogeny of the Blastocladiomycota (fungi) based on nuclear ribosomal DNA. Fungal Biology 115(4-5):381–392. https://doi.org/10.1016/j.funbio.2011.02.004

Powell MJ (2017) Blastocladiomycota. In: handbook of the Protists: Second edition. Pp 1497–1521.

Powell MJ, Lehnen LP, Bortnick RN (1985) Microbody-like organelles as taxonomic markers among oomycetes. BioSystems 18:321–334. https://doi.org/10.1016/0303-2647(85)90032-2

Powell MJ, Letcher PM, Chambers JG, Roychoudhury S (2015) A new genus and family for the misclassified chytrid, Rhizophlyctis harderi. Mycologia 107:419–431. https://doi.org/10.3852/14-223

Preston CE, Agnello AM, Hajek AE (2020a) *Nosema maddoxi* (microsporidia: Nosematidae) in brown marmorated stink bug, *Halyomorpha halys* (Hemiptera: Pentatomidae), populations in the United States. Biological control 144. https://doi.org/10.1016/j.biocontrol.2020.104213

Preston CE, Agnello AM, Vermeylen F, Hajek AE (2020b) Impact of *Nosema maddoxi* on the survival, development, and female fecundity of *Halyomorpha halys*. Journal of Invertebrate Pathology 169:107303. https://doi.org/10.1016/j.jip.2019.107303

Quiroz Velasquez PF, Abiff SK, Fins KC, Conway QB, Salazar NC, Delgado AP, Dawes JK, Douma LG, Tartar A (2014) Transcriptome analysis of the entomopathogenic oomycete *Lagenidium giganteum* reveals putative virulence factors. Applied Environmental Microbiology 80:6427–6436. https://doi.org/10.1128/AEM.02060-14

Radek R, Herth W (1999) Ultrastructural investigation of the spore-forming protist *Nephridiophaga blattellae* in the Malpighian tubules of the German cockroach *Blattella germanica*. Parasitology Research 85:216–231. https://doi.org/10.1007/s004360050538

Radek R, Wellmanns D, Wolf A (2011) Two new species of Nephridiophaga (Zygomycota) in the Malpighian tubules of cockroaches. Parasitology Research 109:473–482. https://doi.org/10.1007/s00436-011-2278-7

Radek R, Wurzbacher C, Gisder S, Nilsson RH, Owerfeldt A, Genersch E, Kirk PM, Voigt K (2017) Morphologic and molecular data help adopting the insect-pathogenic nephridiophagids (Nephridiophagidae) among the early diverging fungal lineages, close to the Chytridiomycota. MycoKeys 25:31–50. https://doi.org/10.3897/mycokeys.25.12446

Roberson RW (2020) Subcellular structure and behaviour in fungal hyphae. Journal of Microscopy 280:75–85. https://doi.org/10.1111/jmi.12945

Rocha SCO, Lopez-Lastra CC, Marano AV, de Souza JI, Rueda-Páramo ME, Pires-Zottarelli CLA (2018) New phylogenetic insights into Saprolegniales (Oomycota, Straminipila) based upon studies of specimens isolated from Brazil and Argentina. Mycological Progress 17:691–700. https://doi.org/10.1007/s11557-018-1381-x

Rossman AY, Palm ME (2006) Why are Phytophthora and other Oomycota not true fungi? Outlooks on Pest Management 17:217–219

Rueda-Páramo ME, Montalva C, Arruda W, Fernandes KK, Luz C, Humber RA (2017) First report of *Coelomomyces santabrancae sp. nov.* (Blastocladiomycetes: Blastocladiales) infecting mosquito larvae (Diptera: Culicidae) in Central Brazil. Journal of Invertebrate Pathology 149:114–118. https://doi.org/10.1016/j.jip.2017.08.010

Sanchez-Peña SR, Buschinger A, Humber RA (1993) *Myrmicinosporidium durum*, an enigmatic fungal parasite of ants. Journal of Invertebrate Pathology 61: 90–96. https://doi.org/10.1006/jipa.1993.1016

Saye LMG, Navaratna TA, Chong JPJ, O'malley MA, Theodorou MK, Reilly M, Li J, Heath IB (2021) Chytridiomycetous gut fungi, oft overlooked contributors to herbivore digestion. Microorganisms 9:1003–1013. https://doi.org/10.1139/m93-153

Scholte E-J, Knols BGJ, Samson RA, Takken W (2004) Entomopathogenic fungi for mosquito control: a review. Journal of Insect Science 4:1–24. https://doi.org/10.1673/031.004.1901

Shen D, Nyawira KT, Xia A (2020a) New discoveries and applications of mosquito fungal pathogens. Current Opinion in Insect Science 40:111–116. https://doi.org/10.1016/j.cois.2020.05.003

Shen D, Tang Z, Wang C, Wang J, Dong Y, Chen Y, Wei Y, Cheng B, Zhang M, Grenville-Briggs LJ, Tyler BM, Dou D, Xia A (2019) Infection mechanisms and putative effector repertoire of the mosquito pathogenic oomycete *Pythium guiyangense* uncovered by genomic analysis. PLoS Genetics:15. https://doi.org/10.1371/journal.pgen.1008116

Shen D, Wang J, Dong Y, Zhang M, Tang Z, Xia Q, Nyawira KT, Jing M, Dou D, Xia A (2020b) The glycoside hydrolase 18 family chitinases are associated with development and virulence in the mosquito pathogen *Pythium guiyangense*. Fungal genetics and Biology135. https://doi.org/10.1016/j.fgb.2019.103290

Singh G, Prakash S (2012) Efficacy of the trichophyton ajelloi and *Lagenidium giganteum* metabolites against mosquitoes after flash chromatography. Parasitology Research 110:2053–2060. https://doi.org/10.1007/s00436-011-2734-4

Sinha KK, Choudhary AK, Kumari P (2016) Entomopathogenic fungi. In: Omkar (ed) ecofriendly Pest Management for Food Security. Academic press, pp 475–505.

Sokolova YY, Weidner E, DiMario PJ (2020) Development of *Anncaliia algerae* (microsporidia) in *Drosophila melanogaster*. Journal of Eukaryotic Microbiology 67:125–131. https://doi.org/10.1111/jeu.12762

Solter LF, Becnel JJ, Vávra J (2012) Research methods for entomopathogenic microsporidia and other protists. In: Second E (ed) Lacey LABT-M of T in IP. Manual of Techniques in Invertebrate Pathology. Academic Press, San Diego, pp 329–371

Spatafora JW, Sung GH, Sung JM, Hywel-Jones NL, White JF (2007) Phylogenetic evidence for an animal pathogen origin of ergot and the grass endophytes. Molecular Ecology 16(8):1701–1711. https://doi.org/10.1111/j.1365-294X.2007.03225.x

Spies CFJ, Grooters AM, Lévesque CA, Rintoul TL, Redhead SA, Glockling SL, Chen CY, de Cock AWAM (2016) Molecular phylogeny and taxonomy of *Lagenidium*-like oomycetes pathogenic to mammals. Fungal Biology 120: 931–947. https://doi.org/10.1016/j.funbio.2016.05.005

Spring O, Gomez-Zeledon J, Hadziabdic D, Trigiano RN, Thines M, Lebeda A (2018) Biological characteristics and assessment of virulence diversity in pathosystems of economically important biotrophic oomycetes. Critical Reviews in Plant Sciences 37:439–495. https://doi.org/10.1080/07352689.2018.1530848

St. Leger RJ, Wang C (2010) Genetic engineering of fungal biocontrol agents to achieve greater efficacy against insect pests. Applied Microbiology and Biotechnology 85:901–907. https://doi.org/10.1007/s00253-009-2306-z

Steele T, Bjørnson S (2014) *Nosema adaliae sp. nov.,* a new microsporidian pathogen from the two-spotted lady beetle, *Adalia bipunctata* L. (Coleoptera: Coccinellidae) and its relationship to microsporidia that infect other coccinellids. Journal of Invertebrate Pathology 115:108–115. https://doi.org/10.1016/j.jip.2013.09.008

Strassert, J.F.H., Wurzbacher, C., Hervé, V. et al. Long rDNA amplicon sequencing of insect-infecting nephridiophagids reveals their affiliation to the Chytridiomycota and a potential to switch between hosts. *Scientific Reports* 396 (2021). https://doi.org/10.1038/s41598-020-79842-6

Su X (2008) A report of the mosquito host range of *Pythium guiyangense* Su. Nature Precedings. https://doi.org/10.1038/npre.2008.1875.1

Su X, Zou F, Guo Q, Huang J, Chen T (2001) A report on a mosquito-killing fungus, *Pythium carolinianum*. Fungal Diversity 7:129–133

Tamim El Jarkass H, Reinke AW (2020) The ins and outs of host-microsporidia interactions during invasion, proliferation and exit. Cellular Microbiology 22. https://doi.org/10.1111/cmi.13247

Tanada Y, Kaya HK (2012) Insect pathology.

Thines M (2018) Oomycetes. Current Biology 28:R812–R813

Timofeev S, Tokarev Y, Dolgikh V (2020) Energy metabolism and its evolution in microsporidia and allied taxa. Parasitology Research 119(5):1433–1441. https://doi.org/10.1007/s00436-020-06657-9

Tokarev YS, Huang WF, Solter LF, Malysh JM, Becnel JJ, Vossbrinck CR (2020) A formal redefinition of the genera *Nosema* and *Vairimorpha* (microsporidia: Nosematidae) and reassignment of species based on molecular phylogenetics. Journal of invertebrate pathology 169. 169:107279. https://doi.org/10.1016/j.jip.2019.107279

Tokarev YS, Zinatullina ZY, Ignatieva AN, Zhigileva ON, Malysh JM, Sokolova YY (2018) Detection of two microsporidia pathogens of the European honey bee *Apis mellifera* (Insecta: Apidae) in Western Siberia. Acta Parasitologica 63: 728–732. https://doi.org/10.1515/ap-2018-0086

Trigos-Peral G, Rutkowski T, Wojtaszyn G, Espadaler X (2017) *Myrmicinosporidium durum* in Poland: a new location for this fungal ant endoparasite and updated world distribution. Acta Parasitologica. https://doi.org/10.1515/ap-2017-0106

Valizadeh P, Guzman-Novoa E, Goodwin PH (2020) Effect of immune inducers on *Nosema ceranae* multiplication and their impact on honey bee (*Apis mellifera* L.) survivorship and behaviors. Insects 11:572. https://doi.org/10.3390/insects11090572

van West P, Beakes GW (2014) Animal pathogenic oomycetes. Fungal Biology 118:525–526. https://doi.org/10.1016/j.funbio.2014.05.004

Verheggen FJ, Vogel H, Vilcinskas A (2017) Behavioral and immunological features promoting the invasive performance of the harlequin ladybird *Harmonia axyridis*. Frontiers in Ecology and Evolution 5

Vilcinskas A, Schmidtberg H, Estoup A, Tayeh A, Facon B, Vogel H (2015) Evolutionary ecology of microsporidia associated with the invasive ladybird *Harmonia axyridis*. Insect Sci. 22:313–324. https://doi.org/10.1111/1744-7917.12159

Vilela R, Humber RA, Taylor JW, Mendoza L (2019) Phylogenetic and physiological traits of oomycetes originally identified as *Lagenidium giganteum* from fly and mosquito larvae. Mycologia 111(3):408–422. https://doi.org/10.1080/00275514.2019.1589316

Vilela R, Montalva C, Luz C, Humber RA, Mendoza L (2018) *Pythium insidiosum* isolated from infected mosquito larvae in Central Brazil. Acta Tropica 185: 344–348. https://doi.org/10.1016/j.actatropica.2018.06.014

Vilela R, Taylor JW, Walker ED, Mendoza L (2015) *Lagenidium giganteum* pathogenicity in mammals. Emerging Infectious Diseases journal 21(2):290–297. https://doi.org/10.3201/eid2102.141091

Vyas N, Dua KK, Prakash S (2007) Efficacy of *Lagenidium giganteum* metabolites on mosquito larvae with reference to nontarget organisms. Parasitology ResearchParasitology Research 101:385–390. https://doi.org/10.1007/s00436-007-0496-9

Walker CA, van West P (2007) Zoospore development in the oomycetes. Fungal Biologyogy Reviews 21(1):10–18. https://doi.org/10.1016/j.fbr.2007.02.001

Wallace Martin W (2000) Two new species of Couchia parasitic in midge eggs. Mycologia 92:1149–1154. https://doi.org/10.1080/00275514.2000.12061262

Wang C, Wang S (2017) Insect pathogenic fungi: genomics, molecular interactions, and genetic improvements. Annual Review of Entomology 62: 73–90. https://doi.org/10.1146/annurev-ento-031616-035509

Wang Y, Ma Y, Wang D, Liu W, Chen J, Jiang Y, Yang R, Qin L (2019) Polar tube structure and three polar tube proteins identified from *Nosema pernyi*. Journal of Invertebrate Pathology:168. https://doi.org/10.1016/j.jip.2019.107272

Weber RWS, Wakley GE, Pitt D (1998) Histochemical and ultrastructural characterization of fungal mitochondria. Mycologist 12:174–179. https://doi.org/10.1016/S0269-915X(98)80076-1

Weiss LM (2020) Microsporidiosis. In: Ryan ET, hill DR, Solomon T, Aronson NE, Endy TP (eds) Hunter's tropical medicine and emerging infectious diseases (tenth edition), tenth edit. Content repository only!, London, pp 825–831.

Woodring JL, Kaya HK, Kerwin JL (1995) *Lagenidium giganteum* in *Culex tarsalis* larvae: production of infective propagules. Journal of Invertebrate Pathology 66:25–32. https://doi.org/10.1006/jipa.1995.1056

Wu K, Yang B, Huang W, Dobens L, Song H, Ling E (2016) Gut immunity in lepidopteran insects. Developmental & Comparative Immunology 64:65–74. https://doi.org/10.1016/j.dci.2016.02.010

Wylezich C, Radek RSM (2004) Phylogenetische analyse der 18S rRNA identifiziert den parasitischen Protisten Nephridiophaga blattellae (Nephridiophagidae) als Vertreter der Zygomycota (fungi). Denisia 13:435–442

Yıldırım H, Bekircan Ç (2020) Ultrastructural and molecular characterization of *Nosema alticae* sp. nov. (microsporidia: Nosematidae), pathogen of the flea beetle, *Altica hampei* Allard, 1867 (Coleoptera: Chrysomelidae). Journal of invertebrate pathology 170. 170:107302. https://doi.org/10.1016/j.jip.2019.107302

Zhou X, Zhang L (2009) Selection of *Antonospora locustae* (protozoa: Microsporidae) with higher virulence against *Locusta migratoria* manilensis (Orthoptera:Acrididae). Biocontrol Science and Technology 19:421–427. https://doi.org/10.1080/09583150902792570

Zimmermann G, Huger AM, Kleespies RG (2013) Occurrence and prevalence of insect pathogens in populations of the codling moth, *Cydia pomonella L.*: a long-term diagnostic survey. Insects 4:425–446. https://doi.org/10.3390/insects4030425

Publisher's Note