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181. © Viability of oomycete propagules following ingestion and excretion by fungus gnats, shore flies, and snails. Hyder, N., Coffey, M. D., and Stanghellini, M. E. *Plant Disease* 93(7):720-726. 2012.

Viability of Oomycete Propagules Following Ingestion and Excretion by Fungus Gnats, Shore Flies, and Snails

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ABSTRACT

Hyder, N., Coffey, M. D., and Stanghellini, M. E. 2009. Viability of oomycete propagules following ingestion and excretion by fungus gnats, shore flies, and snails. *Plant Dis.* 93:720-726.

Sporangia of *Phytophthora capsici* and *P. nicotianae*, as well as hyphal swellings of *Pythium splendens*, *P. sylvaticum*, and *P. ultimum*, were ingested by adult shore flies but none were viable after passing through the digestive tract. Oospores of *Pythium aphanidermatum* retained their viability following ingestion by adult shore flies. Larval stages of fungus gnats and shore flies ingested sporangia of *Phytophthora capsici*, *P. nicotianae*, and *P. ramorum*, but they were not viable upon excretion. In contrast, hyphal swellings of *Pythium splendens*, *P. sylvaticum*, and *P. ultimum*, chlamydozoospores of *Phytophthora ramorum*, and oospores of *Pythium aphanidermatum*, retained their viability after passage through the digestive tract of these larvae. Snails were capable of ingesting and excreting viable sporangia and chlamydozoospores of *P. ramorum*, which upon excretion infected detached leaves. Although the impact of larvae and snails in the rapid dissemination of pathogen propagules is unknown, this work does highlight the possibility that some often-ignored animal–fungus interactions should be considered in long-range dispersal of pathogen propagules via food webs.

Adult shore flies (*Scatella stagnalis* (Fallen)) and fungus gnats (*Bradysia imptiens* (Johannsen)) have been reported to function as aerial vectors of several plant pathogenic fungi, i.e., *Fusarium*, *Verticillium*, and *Thielaviopsis* (3,5,13,21). Although these fungi are soilborne, root-infecting pathogens, they also sporulate on aboveground portions of their hosts. The aboveground propagules of these fungi (hyphal swellings, microsclerotia, and/or chlamydozoospores) serve as a source of inoculum for insect acquisition, via internal and external contamination, and subsequent aerial dissemination. Aerial dissemination by insects can provide an additional means of spread for some soilborne pathogens and can greatly impact the management of such diseases under field and greenhouse production systems. However, with the exception of a single report on the aerial transmission of *Pythium aphanidermatum* by adult shore flies (8), there is no information regarding the role of these insects as vectors of other oomycetes, a major group of extremely destructive root-infecting pathogens. These pathogens produce diverse reproductive structures (i.e., oospores, hyphal swellings, sporangia, and chlamydozoospores) ranging in size from 20

to 60 μm on colonized roots. Their belowground location, however, could preclude acquisition by adult life stages of both shore flies and fungus gnats, which live strictly aboveground. However, some species of oomycetes also produce aboveground reproductive structures that could be vectored by these insects by ingestion and defecation or casual contact. For example, sporangia and chlamydozoospores of *Phytophthora ramorum* are produced on leaves of colonized hosts (2), and sporangia/oospores of *P. capsici* and *P. nicotianae* can be produced on aerial parts of colonized hosts (1,6,11). Additionally, larvae of shore flies and fungus gnats feed belowground and could acquire propagules of *Pythium* species in infected plant roots.

In addition to insects, snails also have been implicated as vectors of oomycetes (16,23). The role of snails in transmitting *Phytophthora citricola* among avocado trees has been documented by El-Hamalawi and Menge (4). Here, feces of snails collected from avocado groves contained viable oospores and hyphal fragments of *P. citricola*, and in greenhouse

experiments, snails exposed to infected avocado plants or infected avocado cuttings were able to transmit the disease to healthy avocado plants via feces or direct contact.

Phytophthora ramorum, the causal agent of Ramorum blight, thrives in moist conditions that also favor high snail populations, making it potentially possible for snails to ingest aboveground propagules of *P. ramorum* and spread the pathogen via infested feces.

The specific objectives of our research were to (i) assess the potential of fungus gnats and shore flies to ingest and excrete propagules of *Phytophthora capsici*, *P. nicotianae*, *P. ramorum*, *Pythium aphanidermatum*, *P. splendens*, *P. sylvaticum*, and *P. ultimum* and the ability of these excreted propagules to initiate infection, and (ii) evaluate ingestion, excretion, and transmission of *Phytophthora ramorum* by brown garden snails (European brown snail), *Helix (Cytomyphalus) aspersa* Müller.

MATERIALS AND METHODS

Oomycete pathogens. Three species of *Phytophthora* and four species of *Pythium* were selected for this study (Table 1). These oomycetes were specifically chosen because they are recognized as major pathogens of diverse greenhouse crops. Additionally, the various propagules produced by these oomycetes provided us the opportunity to assess the survivability of these propagules in the digestive tract of the insects investigated. Stock cultures of the oomycetes were maintained on 10% V8 juice agar unless otherwise specified.

Pathogen cultures. Two methods were employed to produce large quantities of oomycete propagules: cultivation in liquid culture and cultivation in/on infected host tissue. For cultivation of *Phytophthora nicotianae*, *Pythium aphanidermatum*, *P. splendens*, *P. sylvaticum*, and *P. ultimum*,

Table 1. Characteristics of Oomycetes selected for investigation

Oomycete	Propagule type	Average size (μ)	Source
<i>Phytophthora capsici</i>	Sporangia	60 × 36	Pepper
<i>P. nicotianae</i>	Sporangia	40 × 29	Citrus
<i>P. ramorum</i>	Sporangia	55 × 25	Recycled water
<i>P. ramorum</i>	Chlamydozoospores	46–60	Camellias
<i>Pythium aphanidermatum</i>	Oospores	20	Cucumbers
<i>P. splendens</i>	Hyphal swellings	43	Easter lily
<i>P. sylvaticum</i>	Hyphal swellings	32	Lettuce
<i>P. ultimum</i>	Hyphal swellings	25	Lettuce

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