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THE FIRE ANT (HYMENOPTERA: FORMICIDAE) PATHOGEN, *VAIRIMORPHA INVICTAE* (MICROSPORIDIA: BURENELLIDAE), NOT DETECTED IN FLORIDA

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Vairimorpha invictae Jouvenaz & Ellis (Microsporidia: Burenellidae) is a microsporidian pathogen of *Solenopsis* fire ants in South America. Because *Solenopsis invicta* Buren fire ants infected with *V. invictae* were associated with higher mortality, significantly slower colony growth, and reductions in field populations in Argentina, there is considerable interest in utilizing this pathogen as a classical biological control agent against *S. invicta* in the U.S. (Briano & Williams 2002; Oi et al. 2005; Briano 2005). *V. invictae* was formally described by Jouvenaz & Ellis in 1986, however, it was most likely observed much earlier, being the undescribed, dimorphic microsporidium reported from pathogen surveys conducted in Brazil in 1976 and 1979 (Jouvenaz et al., 1980; Briano & Williams 2002). Surveys for *V. invictae* in Argentina reported prevalence rates of 1, 2.3, and 10% after sampling 1836, 2528, and 2064 nests, respectively (Briano et al. 1995; 2006; Briano & Williams 2002).

In the U.S., there have been surveys for various fire ant pathogens since the 1970s. Limited surveys of red and black imported fire ants, *S. invicta and Solenopsis richteri* Forel, respectively, were conducted in northern Florida and in Mississippi in 1971 and 1972 (Broome 1974; Jouvenaz et al. 1977). More extensive surveys in the southeastern U.S. of the 2 imported fire ants (*S. invicta* and *S. richteri*) and the native fire ants *Solenopsis geminata* (F.) and *Solenopsis xyloni* McCook were documented by Jouvenaz *et al.* (1977). In these surveys, only ubiquitous, nonspecific organisms were detected, with the exception of a fungus that seemed to have minimal effects on *S. invicta* populations (Broome 1974; Jouvenaz et al. 1977, 1981). In 1989, the fungi *Metarhizium anisopliae* var. *anisopliae* (Johnston) Tulloch and a *Conidiobolus* species were isolated from *S. invicta* queens collected after a mating flight in Texas (Sánchez-Peña & Thorvilson 1992). Fire ants examined from studies conducted in Alabama, Florida, and Tennessee in 2000-2003 revealed infections of a Neogregarine protozoan, *Mattesia* sp., and the fungus *Myrmicinosporidium durum* Hölldobler (Pereira et al. 2002; Pereira 2004) in *S. invicta*. A microsporidian fire ant pathogen from South America, *Kneallhazia (=Thelohania) solenopsae* (Knell, Allen & Hazard), was first detected in the U.S. in 1996 (Williams et al. 1998, 2003). Interestingly, subsequent examination of archived *S. invicta* specimens revealed that *K. solenopsae* had been present in Texas since the 1980s (Snowden &Vinson 2006). Surveys for *K. solenopsae* have been conducted in Florida, Texas, Mississippi, and Louisiana, with no reports of *V. invictae* detection (Pereira et al. 2002; Streett et al. 2004; Mitchell et al. 2006; Milks et al. 2008). However, the focus of these surveys was toward *K. solenopsae*. To date, there have been no reports of *V. invictae* in the U.S. The occurrence of *K. solenopsae* in the U.S. permitted removal of the quarantine impediment which has facilitated its utilization against *S. invicta* (Oi & Valles 2009). Because of the aforementioned interest in *V. invictae* as a biological control agent, we specifically examined samples of *S. invicta* for *V. invictae* to ascertain its presence or absence in the U.S.

S. invicta samples were collected mainly in Florida, supplemented by limited sampling forays in Mississippi, Alabama, Georgia, and South Carolina. Sampling sites located in the Florida panhandle, Alabama, and Mississippi were usually at or near highway rest stops or along secondary and tertiary roads near exits of U.S. Interstate Highway 10. These samples were collected in August 2009. Sampling sites in peninsular Florida and Georgia were located along six roadside transects (1 near Macon, Georgia; 4 near Gainesville, Florida; 1 near Moore Haven, Florida) of about 3.5 miles with stops at roughly 0.5 mile intervals (8 sites in each transect). When possible, 3 representative fire ant nests were sampled at each of the 48 sites in spring 2011. Further sampling occurred in north-central Florida from 100 sites in 5 counties described by Porter (1992). For each of these sites an average of 3 (±1 SD) nest samples were examined. Additional samples (n = 270) were obtained from the Valles et al. (2010) study conducted in the vicinity of Gainesville, Florida (Alachua Co.). Remaining samples were collected at other sites in Florida. The South Carolina sampling was conducted during Sep 2003 at a West Ashley shopping mall on the outskirts of Charleston. This sampling was specifically conducted because one of 4 arbitrarily sampled nests at this location in 1997 revealed a possible observation of *V. invictae* spores in *S. invicta* workers (D.H.O. unpublished data).

Adult worker ants were collected by inserting a vial into a *S. invicta* nest for several minutes which allowed swarming ants to fall into the vial. Vial interiors were coated with talcum powder or fluon (Daikin-Polyflon PTFE D-210, Daikin America, Orangeburg, New York) to prevent ants from escaping. Ants were immediately preserved in 95% ethanol or chilled then frozen alive. Approximately 10-50 ants per nest were examined for *V. invictae* spores by phase-contrast microscopy or by PCR assay (Valles et al. 2004; Oi et al. 2005). For nests sampled along transects in peninsular Florida and Georgia and in north-central Florida, approximately 10 ants per nest were combined per sampling site and there were usually 3 nests per site. In addition to *V. invictae*, the presence of *K. solenopsae* was also documented, and percentages of polygyne nests at some sampling sites were estimated visually or by PCR assay (Oi et al. 2004).

V. invictae was not detected in any of the *S. invicta* nest samples (Table 1). A total of 1016 samples (i.e., colonies) were examined from 170 sites located in 21 counties among 5 states. The majority of the sampling sites (91%) and nest samples (83%) were from Florida. While social form (i.e. monogyny or polygyny) was not determined for all sites, polygyny was present at 64% of the 114 sites where it was assessed, with an average of 50.6% (±29.8% SD) of the nests being polygyne at the polygynous sites. Polygyny has been associated with higher incidences of pathogen infections in *S. invicta* (Valles et al. 2010). While it is conceivable that the roadside habitats that were predominantly sampled may not be conducive to *V. invictae* infections, another fire ant infecting microsporidium, *K. solenopsae*, was detected in 14.5% (147/1016) of the samples, which is within the 10-31% prevalence reported for *K. solenopsae* in surveys in Mississippi, Louisiana, and Texas (Streett et al. 2004; Mitchell et al. 2006, Milks et al. 2008). Thus, our methods, which were similar to procedures used to detect both microsporidia in South America (Valles et al. 2004; Briano et al. 1995), should have been sufficient to detect *V. invictae* in the Florida survey. Furthermore, all PCR reactions included positive controls to verify the proper function of the test. In our search for *V. invictae*, there is no evidence of this microsporidium being established in *S. invicta* populations in the U.S. As a result, importation and establishment of *V. invictae* in U.S. populations of *S. invicta* remains an important research goal that would add another natural enemy of this invasive ant (Porter 1998).

Summary

Surveys were conducted to search specifically for the microsporidian pathogen *Vairimorpha invictae* in red imported fire ants (*Solenopsis invicta*), in the U.S. *V. invictae* was not detected in any of the 1,016 nest samples collected in 21 counties located in 5 states. A majority of samples (83%) originated from Florida. Despite the absence of *V. invictae*, another microsporidum, *K. solenopsae*,

Table 1. Number of *Solenopsis invicta* sites and nest samples per county with *Varimorpha invictae* and/or *Kneallhazia solenopsae*.

State (County)	No. Sites	No. Nest Samples	No. Samples with *V. invictae*	No. Samples with *K. solenopsae*
Alabama (Baldwin)	1	23	0	0
Alabama (Mobile)	1	24	0	0
Georgia (Crawford)	8	17	0	0
Florida (Alachua)	51	428	0	89[1]
Florida (Bradford)	2	8	0	0
Florida (Citrus)	3	5	0	0
Florida (Columbia)	3	9	0	2
Florida (Escambia)	1	21	0	0
Florida (Gilchrist)	5	12	0	1
Florida (Glades)	8	25	0	1
Florida (Holmes)	1	19	0	0
Florida (Lake)	1	3	0	0
Florida (Levy)	14	43	0	7[1]
Florida (Marion)	42	148	0	41
Florida (Putnam)	9	25	0	0
Florida (Santa Rosa)	3	63	0	1
Florida (Sumter)	2	7	0	0
Florida (Union)	9	23	0	0
Mississippi (Harrison)	2	36	0	2
Mississippi (Jackson)	3	64	0	3
South Carolina (Charleston)	1	13	0	0
Totals:	170	1016	0	147

[1]Based partly on 260 (Alachua Co.) and 10 (Levy Co.) samples that were from the same collections used in Valles et al. (2010) in which 22.5% of the samples were infected with *Kneallhazia solenopsae*.

was detected in 14.5% of the samples. Based on our sampling, *V. invictae* is currently not established in the U.S. *V. invictae* represents an additional natural enemy that could be imported from South America for the classical biological control of *S. invicta*.

References Cited

Briano, J. A. 2005. Long-term studies of the red imported fire ant, *Solenopsis invicta*, infected with the microsporidia *Vairimorpha invictae* and *Thelohania solenopsae* in Argentina. Environ. Entomol. 34: 124-132.

Briano, J. A., and Williams, D. F. 2002. Natural occurrence and laboratory studies of the fire ant pathogen *Vairimorpha invictae* (Microsporida: Burenellidae) in Argentina. Environ. Entomol. 31: 887-894.

Briano, J. A., Jouvenaz, D. P., Wojcik, D. P., Cordo, H. A., and Patterson, R. S. 1995. Protozoan and fungal diseases in *Solenopsis richteri* and *S. quinquecuspis* (Hymenoptera: Formicidae), in Buenos Aires province, Argentina. Florida Entomol. 78: 531-537.

Briano, J. A., Calcaterra, L. A., Vander Meer, R., Valles, S. M., and Livore, J. P. 2006. New survey for the fire ant microsporidia *Vairimorpha invictae* and *Thelohania solenopsae* in southern South America, with observations on their field persistence and prevalence of dual infections. Environ. Entomol. 35: 1358-1365.

Broome, J. R. 1974. Microbial control of the imported fire ant *Solenopsis richteri*. Ph.D. dissertation. Mississippi State University, Mississippi State, MS.

Jouvenaz, D. P., and Ellis, E. A. 1986. *Vairimorpha invictae* n. sp. (Microspora: Microsporida), a parasite of the red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae). J. Protozool. 33: 457-461.

Jouvenaz, D. P., Allen, G. E., Banks, W. A., and Wojcik, D. P. 1977. A survey for pathogens of fire ants, *Solenopsis* spp. in the southeastern United States. Florida Entomol. 60: 275-279.

Jouvenaz, D. P., Banks, W. A., and Atwood, J. D. 1980. Incidence of pathogens in fire ants, *Solenopsis* spp., in Brazil. Florida Entomol. 63: 345-346.

Jouvenaz, D. P., Lofgren, C. S., and Banks, W. A. 1981. Biological control of imported fire ants; a review of current knowledge. Bull. Entomol. Soc. Am. 27: 203-208.

Milks, M. L., Fuxa, J. R., and Richter, A. R. 2008. Prevalence and impact of the microsporidium *Thelohania solenopsae* (Microsporidia) on wild populations of red imported fire ants, *Solenopsis invicta,* in Louisiana. J. Invertebr. Pathol. 97: 91-102.

Mitchell, F. L., Snowden, K., Fuxa, J. R., and Vinson, S. B. 2006. Distribution of *Thelohania solenopsae* (Microsporida: Thelohaniidae) infecting red imported fire ant (Hymenoptera: Formicidae) in Texas. Southwest Entomol. 31: 297-306.

Oi, D. H., and Valles, S. M. 2009. Fire ant control with entomopathogens in the USA, pp. 237-257 *In* A. E. Hajek, T. R. Glare and M. O'Callaghan [eds.], Use of microbes for control and eradication of invasive arthropods. Springer, New York, NY. 366 pp.

Oi, D. H., Valles, S. M., and Pereira, R. M. 2004. Prevalence of *Thelohania solenopsae* (Microsporidia: Thelohaniidae) infection in monogyne and polygyne red imported fire ants (Hymenoptera: Formicidae). Environ. Entomol. 33: 340-345.

Oi, D. H., Briano, J. A., Valles, S. M., and Williams, D. F. 2005. Transmission of *Vairimorpha invictae* (Microsporidia: Burenellidae) infections between red imported fire ant (Hymenoptera: Formicidae) colonies. J. Invertebr. Pathol. 88: 108-115.

Pereira, R. M. 2004. Occurrence of *Myrmicinosporidium durum* in *Solenopsis invicta* and other new host ants in eastern United States. J. Invertebr. Pathol. 86: 38-44.

Pereira, R. M., Williams, D. F., Becnel, J. J., and Oi, D. H. 2002. Yellow head disease caused by a newly discovered *Mattesia* sp. in populations of the red imported fire ant, *Solenopsis invicta*. J. Invertebr. Pathol. 81: 45-48.

Porter, S. D. 1992. Frequency and distribution of polygyne fire ants (Hymenoptera: Formicidae) in Florida. Florida Entomol. 75: 248-257.

Porter, S. D. 1998. Biology and behavior of *Pseudacteon* decapitating flies (Diptera: Phoridae) that parasitize *Solenopsis* fire ants (Hymenoptera: Formicidae). Florida Entomol. 81: 292-309.

Sánchez-Peña, S. R., and Thorvilson, H. G. 1992. Two fungi infecting red imported fire ant founding queens from Texas. Southwestern Entomol. 17: 181-182.

Snowden, K., and Vinson, S. B. 2006. Development of *Thelohania solenopsae* as an effective biological control agent for the red imported fire ant, *Solenopsis invicta*. Texas Imported Fire Ant Research and Management Project Progress Report Year 1-VII-2006. http://fireant.tamu.edu/research/projects/pdf/06snowdenvinson.pdf).

Streett, D. A., Freeland Jr., T. B., and Pranschke, A. M. 2004. Distribution of *Thelohania solenopsae* in red imported fire ant populations in Mississippi, pp 150-152 *In* Proc. 2004 Imported Fire Ant Conf., Baton Rouge, Louisiana 21-23-III-2004.

Valles, S. M., Oi, D. H., Briano, J. A., and Williams, D. F. 2004. Simultaneous detection of *Vairimorpha invictae* and *Thelohania solenopsae* in fire ants by PCR. Florida Entomol. 87: 85-87.

Valles, S. M., Oi, D. H., and Porter, S. D. 2010. Seasonal variation and the co-occurrence of four pathogens and a group of parasites among monogyne and polygyne fire ant colonies. Biol. Control 54: 342-348.

Williams, D. F., Knue, G. J., and Becnel, J. J. 1998. Discovery of *Thelohania solenopsae* from the red imported fire ant, *Solenopsis invicta*, in the United States. J. Invertebr. Pathol. 71:175-176.

Williams, D. F., Oi, D. H., Porter, S. D., Pereira, R. M., and Briano, J. A. 2003. Biological control of imported fire ants (Hymenoptera: Formicidae). American Entomol. 49: 150-163.