

# Occurrence of *Myrmicinosporidium durum* in red imported fire ant, *Solenopsis invicta*, and other new host ants in eastern United States

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## Abstract

*Myrmicinosporidium durum*, a parasitic fungus in several ant species, is reported from seven new hosts collected in the eastern United States, including *Solenopsis invicta*, *Solenopsis carolinensis*, *Paratrechina vividula*, *Pheidole tysoni*, *Pheidole bicarinata*, *Pyramica membranifera*, and *Pogonomyrmex badius*. Spores can be found in most ant body parts, are dark brown when mature, and clear to light brown while immature. Ants infected with mature spores appear darker than normal. Spores from different hosts were 47–57 µm in diameter. Prevalence in host populations varied between 2 and 67% of the ants, and 3–100% of the colonies. Infection was most common in *S. carolinensis* with prevalence rates between 22 and 67%. Prevalence rates for *S. invicta* individuals were lower than for other ants, however, prevalence rates within the infected colonies were as high as 31%. Observations of disease were recorded mostly from late summer and fall. Possible implications of this new disease in red imported fire ants are discussed. Published by Elsevier Inc.

**Keywords:** *Myrmicinosporidium durum*; Fungus; Formicidae; *Solenopsis invicta*; *Solenopsis carolinensis*; *Paratrechina vividula*; *Pheidole tysoni*; *Pheidole bicarinata*; *Pyramica membranifera*; *Pogonomyrmex badius*

## 1. Introduction

*Myrmicinosporidium durum* is a parasitic fungus found infecting several ant species classified in several different subfamilies (Sánchez-Peña et al., 1993). Infected ants are usually filled with dark, thick-walled spores that can be seen through the insect cuticle and which usually cause the ant to appear darker in color than normal. This fungus has been reported from several localities in Europe (Buschinger and Winter, 1983; Espadaler, 1982; Hölldobler, 1933; Sánchez-Peña et al., 1993), but reports from the Americans are limited to a single publication (Sánchez-Peña et al., 1993) and only from specimens collected in pitfall traps. Insects collected in these traps fall into a preserving liquid that prevents isolation of the fungus for other studies.

The exact nature of this fungus, its phylogenetic relationships, and the effects on the host ants remain un-

certain. Insects infected with late stages of the disease, i.e., thick-walled spores with few, or no vestiges of mycelium, have been collected in pitfall traps, which only collect active insects. Therefore, effects of the fungus may be minor in some infected ants at least until very late stages of fungal development. Also, no estimates of prevalence rates have been published for this fungus in the different ant populations that it infects.

This report includes seven new hosts for *M. durum*, including the red imported fire ant, *Solenopsis invicta* Buren, all collected in the eastern United States. The red imported fire ant causes close to 6 billion dollars in annual economic impact in the southeastern United States (Pereira et al., 2002) and has been the target of renewed biological control efforts in recent years (Williams et al., 2003). Previous surveys in the United States failed to detect *M. durum* in this ant (Jouvenaz, 1983, 1986, 1990; Jouvenaz et al., 1977, 1980). Estimated prevalence rates for the disease were obtained for red imported fire ants and other ant populations, both within infected nests and within localities.

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## 2. Materials and methods

### 2.1. Ant collections

Pitfall traps, each consisting of a 25-ml plastic tube 1/3-filled with propylene glycol anti-freeze fluid, were used for general ant collection. These traps were placed in several locations, usually in association with experiments in which an assessment of the ant and other crawling arthropod fauna was intended. Pitfall traps were sunk upright into the soil until the tube top was level with the soil surface. Typically, the traps were left at least overnight, but could be left in the field for several days before being collected. Once the traps were collected and returned to the laboratory, the arthropods were separated from the anti-freeze liquid, preserved in 70% ethanol, and classified into taxonomic categories. During the separation and classification of ants, any individuals that showed signs of pathogens were separated and examined further. A battery-operated vacuum cleaner was also used for general ant collection. This device was used to aspirate material from several 3-m<sup>2</sup> areas from a lawn in Gainesville, FL (N 29.637°; W 82.360°). The collected material was placed in Berlese funnels and insects were collected in 70% ethanol before being classified as described above.

At the Ames Plantation near Grand Junction, TN, (N 35.082°; W 89.210°) a total of 13 pitfall traps were placed at 6-m intervals in the north–south and east–west directions from the center of plots where *Solenopsis richteri* (black imported fire ant) colonies were inoculated with the microsporidium *Thelohania solenopsae*. In Florida and Alabama locations, pitfall traps were generally used in the spring and the fall. These sites were of two types: roadsides and pastures. On roadsides, 15 pitfall traps were placed at approximately 8-m intervals in 1–3 lines along the road, depending on space availability. In pasture sites, 4–8 pitfall traps were arranged in one or two circles 4–8 m from the plot center, or trap placement was similar to that on the roadsides.

*Paratrechina vividula* and *Pheidole tysoni* were also collected in 50-mm plastic Petri dishes containing a cotton pad soaked with a sugar solution. These baited traps were placed at 6-m intervals in a 7 × 7 grid surrounding the center of plots at the Ames Plantation mentioned above. Baited traps were left open for 20–30 min, after which the dishes were closed, returned to the laboratory, and the ants were freeze-killed. If different ants were present at the same trap, they were separated, identified, and preserved in 70% ethanol.

*Pogonomyrmex badius* was also collected in Madison Co., FL (N 30.522°; W 83.289°), using a battery-operated vacuum cleaner by aspirating ants as they exited the colony galleries in the soil. To obtain larger number of individuals, nests were excavated carefully to expose the

galleries as the ants were aspirated. Collected ants were maintained alive in the laboratory in trays, the walls of which were coated with Fluon (Asahi Glass Fluoropolymers, Chadds Ford, PA) to prevent escape. Ants were fed sunflower seeds and given access to water and nesting harborage.

*Solenopsis carolinensis* was also collected in north Union Co., FL (N 30.126°; W 82.204°), using baited traps consisting of 74-ml plastic vials containing a piece of Sandies pecan shortbread (Keebler, Elmhurst, IL) and capped with a lid punctured with small (0.5–1 mm) holes. These traps were buried in soil to a depth of 10–15 cm, and collected after 1–3 days. Collected ants were maintained alive in the laboratory in Fluon-coated trays as described above. Ants were fed pecan shortbread and given access to water and nesting harborage.

*Solenopsis invicta* was also collected in a farm near Marianna, FL (N 30.679°; W 85.254°), using plastic tubes of various sizes that were coated internally with Fluon to prevent escape. Tubes were inserted directly into the fire ant nests and collected after 1–10 min when sufficient number of ants had crawled into the tubes. Ants collected in this manner were typically freeze-killed and examined in laboratory, but occasionally ants were examined while still alive. Colonies of the red imported fire ants were collected by shoveling nest soil into Fluon- or talc-coated 20-l plastic buckets. Ants were flooded out of the soil as described by Banks et al. (1981), and maintained alive in the laboratory in Fluon-coated trays containing nesting harborage, water tubes, 10% sugar solution tubes, cooked egg yolks, and freeze-killed crickets.

### 2.2. Pathogen observations and fungal measurements

Ant specimens were examined with a dissecting microscope, or with a light or phase microscope for signs of *M. durum* infection including darker coloration and the presence of round spores in different regions of the body. Other characteristics of the ant host and the fungal pathogen were noted during these observations and compared across the ant species.

In those species with several infected ants, mature spores from an ant with typical infection were measured. Two measurements, for the longest axis and a broadest dimension perpendicular to that axis, were obtained using microscope-mounted micrometer and averaged for each spore. Also, a spore shape ratio was obtained by dividing the shorter diameter by the longer diameter. This ratio is equal to 1 for round spores, but less than 1 for spores that are more oval in shape. Mean measurements for the average diameter and spore shape ratio were compared across ant species using ANOVA followed by mean comparisons using Fisher's protected least significant difference at the 5% level.

### 2.3. Prevalence rate estimates

Prevalence rates were estimated at two levels. When distinct collection samples or colonies were obtained for an ant species, the colony prevalence rate was determined by dividing number of infected samples by the total number of samples obtained at a location. On several occasions, the ant samples obtained were also used to estimate the prevalence rate within an ant population. Except for *S. invicta*, the ants from a single species collected in a single pitfall trap were considered to belong to the same field colony, because spacing of the traps in the field was expected to exceed the foraging territory of most ants. Therefore, ants collected in a single pitfall trap were used to estimate the within population prevalence rate. *P. vividula* collections from pitfall traps and baited Petri dish traps from each of the 13 positions for the central row and column in the 7 × 7 grid were combined before prevalence rates were calculated. Population prevalence rates were calculated considering all the ants collected at a specific location/time combination. However, for *P. badius* and *S. invicta*,

distinct colonies were sampled or collected in the field, so the colony prevalence rates were obtained directly from individual colonies.

## 3. Results

### 3.1. Pathogen observations and fungal measurements

The general aspect of the infected individuals is similar in each of the different ant species (Fig. 1). Spores can be found in most parts of the ant body, although smaller host species do not have spores in their antennae, legs, and other regions where the development of the large spores is restricted by the body part diameter. Mature spores are dark brown while immature spores vary from a clear to light brown. A mixture of spores of different stages of development can be found in different hosts. Except for specimens observed very carefully, the presence of immature spores could be mistaken as oil droplets or other lipids in the ant body. Thus, the disease can be recognized by the darker than normal color

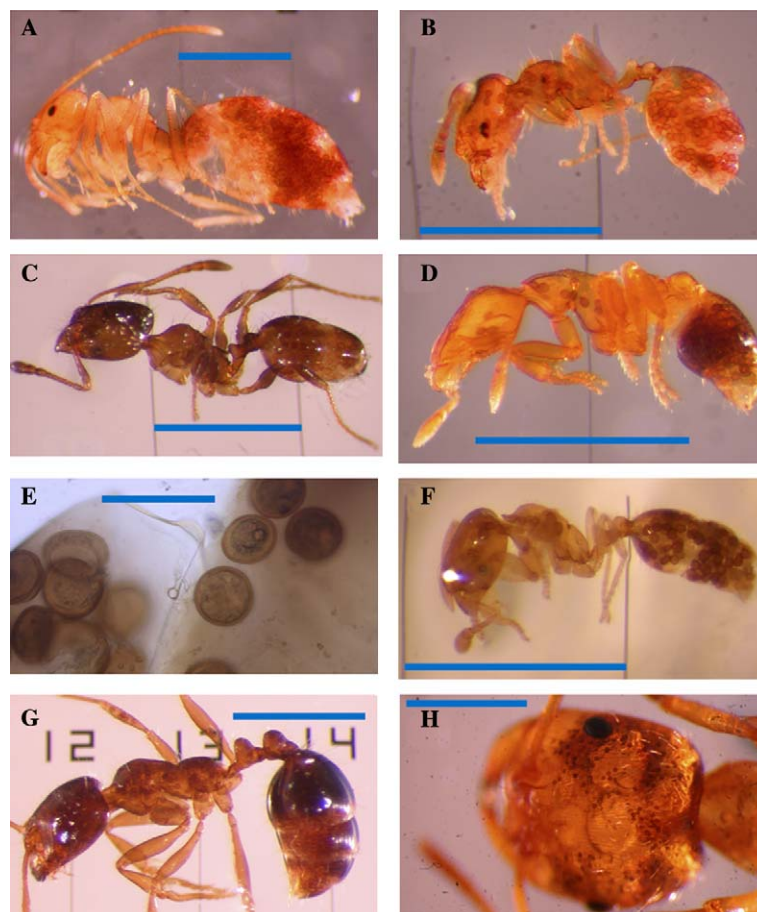


Fig. 1. New ant hosts of *Myrmicinosporidium durum*: (A) *Paratrechina vividula*; (B) *Pheidole tysoni*; (C) *Pheidole bicarinata*; (D) *Pyramica membranifera*; (E) *Myrmicinosporidium durum* spores seen inside abdomen of *P. bicarinata* ant in (C); (F) *Solenopsis carolinensis*; (G) *Solenopsis invicta*; and (H) *Pogonomyrmex badius* head. Note. dark and light colored round spores in different body parts. Blue bars are 1 mm long except for (E) where bar is 100 μm long.

Table 1  
Spore dimensions for *Myrmicinosporidium durum* from four different ant hosts

	Spore diameter ( $\mu\text{m}$ ) <sup>a</sup> (Mean $\pm$ SEM)	Spore shape ratio <sup>b</sup> (Mean $\pm$ SEM)
<i>Solenopsis invicta</i>	54.8 $\pm$ 6.79 a	0.97 $\pm$ 0.0004 ab
<i>Solenopsis carolinensis</i>	55.9 $\pm$ 1.87 a	0.93 $\pm$ 0.0004 b
<i>Pogonomyrmex badius</i>	47.0 $\pm$ 0.24 b	0.98 $\pm$ 0.0004 a
<i>Paratrechina vividula</i>	57.4 $\pm$ 3.23 a	0.97 $\pm$ 0.0001 a

Means followed by the same letter are not significantly different at  $P > 0.05$ .

<sup>a</sup>  $P$  value = 2.4E-08;  $F = 22.32$ ,  $df = 3$ ; 36.

<sup>b</sup>  $P$  value = 0.0054,  $F = 4.99$ ,  $df = 3$ ; 36.

of the ants carrying mature spores. This is especially true for those ant species that are normally very light in color such as *S. carolinensis*, *P. membranifera*, and *P. tysoni*. However, infections in dark colored ants such as *P. badius* and *S. invicta* can also be recognized by an unusually darker color, especially of the head.

With the exception of *P. badius*, no significant differences were observed in spore size found in any of the other three ant species (Table 1). The spores in *P. badius* were approximately 16% smaller (significant at the 5% confidence level) than those in other hosts. The spores observed in *S. carolinensis* were slightly more

oval than those in the other three ant hosts observed, as indicated by the spore shape ratio of 0.93 compared to ratios between 0.97 and 0.98 for the other ant species (Table 1).

The USDA-ARS laboratory in Gainesville, FL (CMAVE—Center for Medical, Agricultural, and Veterinary Entomology) and a farm in Houston Co., AL (N 31.156°; W 85.212°) are the only locations where more than one ant species has been collected with *M. durum* infection (Table 2). Infected *S. invicta*, *S. carolinensis*, and *Pheidole bicarinata* were collected at the Alabama farm, whereas *S. invicta*, *S. carolinensis*, and *P. membranifera* were collected from a grassy area (approximately 30  $\times$  100 m) just north of CMAVE.

### 3.2. Prevalence rate estimates

For samples in which the disease was detected, prevalence rates based on number of ants collected varied between 2.4 and 66.7% for all samples, and between 2.4 and 45.4% for samples of more than 20 ants (Table 2 and Fig. 2). Most prevalence rates based on number of colonies with infected insects were obtained from smaller samples but vary from 3.2 to 100%.

Table 2  
*Myrmicinosporidium durum* prevalence rates in different ant species, in eastern United States

Species	Locality <sup>a</sup>	Date	# Ants	(% Prevalence)	# Trap or colony <sup>b</sup>	(% Prevalence)
<i>Paratrechina vividula</i>	Ames Plantation, TN	Aug. 2000	501	(4.6)	3 C <sup>c</sup>	(66.7)
<i>Pheidole tysoni</i>	Ames Plantation, TN <sup>d</sup>	Aug. 2000	23	(8.7)	3	(33.3)
<i>Pheidole bicarinata</i>	Houston Co., AL	Oct. 2002	102	(6.9)	19	(15.8)
<i>Pogonomyrmex badius</i>	Madison Co., FL	Oct. 2001	6	(16.7)	1	(100)
	Madison Co., FL	Aug. 2003	109	(23.9)	3 C	(100)
<i>Pyramica membranifera</i>	CMAVE, Gainesville, FL	Oct.–Nov. 2003	Only 1 infected ant collected		11	(9.1)
<i>Solenopsis carolinensis</i>	Newberry, FL	Apr. 2000	3	(66.7)	2	(50.0)
	Union Co., FL	Oct. 2001	55	(45.4)	13	(61.5)
	Union Co., FL	Oct. 2002	102	(30.4)	17	(47.1)
	Houston Co., AL	Oct. 2002	151	(22.5)	20	(25.0)
	CMAVE, Gainesville, FL	Nov. 2003	—	—	8	(25.0)
<i>Solenopsis invicta</i>	Houston Co., AL	Oct. 2002	41	(2.4)	—	—
	Marianna, FL	Sep. 2003	266	(31.2)	22 C	(13.6)
	Marianna, FL	Oct. 2003	92	(4.3)	30 C	(10.0)
	Marianna, FL	Nov. 2003	Dead 97 Live 1581	(36.1) <sup>e</sup> (25.9)	27 C	(3.7)
	CMAVE, Gainesville, FL	Nov. 2003	5	(20.0)	62	(3.2)

<sup>a</sup> Latitude/longitude coordinates for sample localities are: CMAVE, Gainesville, FL (N 29.637°; W 82.360°), Newberry, FL (N 29.651°; W 82.656°), Madison Co., FL (N 30.522°; W 83.289°), Union Co., FL (N 30.126°; W 82.204°), Marianna, FL (N 30.679°; W 85.254°), Ames Plantation, TN, (N 35.082°; W 89.210° for *P. vividula* and N 35.099°; W 89.216° for *P. tysoni*), and Houston Co., AL (N 31.156°; W 85.212°).

<sup>b</sup> Numbers represent number of traps with the ant species in the collection, unless followed by “C,” which indicates that ant colonies were sampled.

<sup>c</sup> *Paratrechina vividula* population distribution in the sampling area suggests existence of three colonies (west end, northeast, and southeast populations) as seen in Fig. 2.

<sup>d</sup> The *Pheidole tysoni* collections were from a pasture 1.9 km NNW from the area with *P. vividula* described in Fig. 2.

<sup>e</sup> Separate prevalence rates estimates were obtained for all live and dead ants collected from one colony.

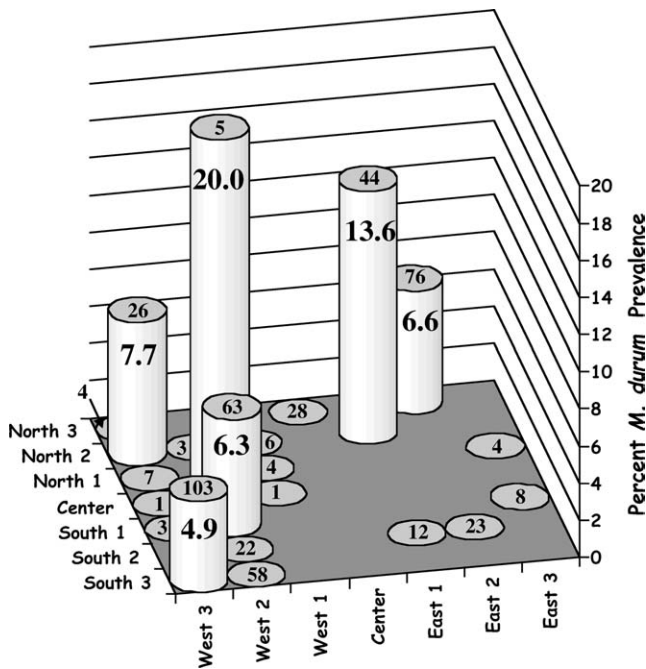


Fig. 2. Percent prevalence of *Myrmicinosporidium durum* in *Paratrechina vividula* at the Ames Plantation, Grand Junction, TN, (N 35.082°; W 89.210°) on samples collected 3–5 August 2000. A zero-height bar indicates samples that contained only uninfected *P. vividula*. Small numbers on top of bars indicate total *P. vividula* ants in the sample, and larger numbers on the side of the bar are the percent *M. durum* prevalence in the sample.

One of only three *P. membranifera* ants collected was infected with *M. durum*. The only infected sample obtained for *P. tysoni* contained 23 ants from which two were infected with *M. durum*. These infected ants came from the Ames Plantation near Grand Junction, TN, from where the *P. vividula* ants (Fig. 2) were obtained. Another *Pheidole* species infected with *M. durum*, *P. bicarinata*, was found in Alabama. Prevalence rates, based on number of *P. bicarinata* collected, were 1 and 13% in the two samples obtained.

*Myrmicinosporidium durum* prevalence seems to be quite common in *S. carolinensis* populations in northern Florida and southern Alabama, with estimated prevalence rates between 22.5 and 66.7% (Table 2) at sites where the disease was detected. Of the traps containing *S. carolinensis*, 40% contained infected ants.

All three *P. badius* colonies collected within 190 m along a dirt farm road in Madison Co., FL, were infected with *M. durum*. Estimates from two of these colonies indicated that between 20 and 32% of the ants were infected with *M. durum* for an average of 23.9% disease prevalence (Table 2). In this ant species, young adult workers with almost no melanized integument can be observed carrying *M. durum* spores. Older infected workers can be easily recognized in the colony by their very dark brown to black color, compared to a red-brown coloration of uninfected ants. At this stage, the

abdominal contents of the infected ants seem to be a liquid mass of spores.

*Solenopsis invicta* samples collected in September 2003 consisted of small groups of ants collected from 10 nests within 500-m<sup>2</sup> plots. Collections from October and November 2003 consisted of all fire ant colonies found in the same plots or adjacent areas not exceeding 1000 m<sup>2</sup>. Prevalence rates for *S. invicta* populations in terms of infected colonies seem to be lower than that of other ants observed. However, prevalence rates within the infected colonies were as high as 31% (Table 2). For a single small colony for which prevalence rate was obtained for all living and dead ants present after the colony was removed from soil and established in rearing trays, *M. durum* prevalence was slightly higher in cadavers (36.1%) than in living ants (25.9%).

*Myrmicinosporidium durum*-infected *P. vividula* were collected in the Ames Plantation in August 2000 from a population of ants that was also sampled in May 1999, May and October 2000, and April 2001 without any other collection of diseased ants. A single *M. durum*-infected *P. vividula* was collected in September 1997, in the Ames Plantation but in a field 6.4 km away (N 35.139°; W 89.225°). The spatial distribution of infected and uninfected *P. vividula* samples within a 40 × 40 m area is represented in Fig. 2. Ant distribution in this area suggests that infected ants came from at least two colonies (west end, and northeast populations) of at least three colonies present in the area. Only the colony in the southeast quadrant of the area did not show any infected ant. The two infected colonies had prevalence rates of 3.9 (12/306) and 7.4% (11/148), respectively for the west end and northeast populations, and an overall prevalence rate of 4.6% (Table 2).

#### 4. Discussion

This is the first report of *M. durum* in seven new ant hosts: *P. vividula*, *P. tysoni*, *P. bicarinata*, *P. membranifera*, *S. carolinensis*, *S. invicta*, and *P. badius*. The genera *Paratrechina* and *Pyramica* were not previously described as hosts of this fungus. These are also the first reports of this fungus occurring in eastern United States. The only other record of *M. durum* in the United States is from *Pogonomyrmex barbatus* from Texas (Sánchez-Peña et al., 1993). These authors also list 14 other hosts from five ant genera as *M. durum* hosts.

The *M. durum* spores observed in the different hosts reported here are within the sizes described previously (Buschinger and Winter, 1983; Espadaler, 1982; Sánchez-Peña et al., 1993). The smaller size of the spores in *P. badius* may be an indication that this host is infected with different strain of *M. durum* or even a different species of similar fungus. However, the smaller spore size may also indicate some degree of suboptimal

nutritional suitability of the host for *M. durum*. The *M. durum* spores from the European ant *Leptothorax unifasciatus* seem pronouncedly oval in published photographs (Buschinger and Winter, 1983), as reported here for infections in *S. carolinensis*. The shape of the spores in *S. carolinensis* may have been affected by the small host size. Despite superficial similarities among the fungi found in the different hosts, and the reports in the literature indicating that *M. durum* infects many diverse ant species (Sánchez-Peña et al., 1993), definite classification of the fungi affecting different ant populations will probably require comparisons of genetic materials. With the large number of *M. durum*-infected ants available, and advances in molecular biology and genetic techniques, it may now be possible to elucidate the nature of this fungus. Studies have been initiated with this purpose.

Most of the collections containing *M. durum*-infected ants were not intended to serve as a disease survey. Discovery of the diseased ants was due to the close examination of many ant samples from diverse localities in Florida, Alabama, and Tennessee. Although *M. durum* was not observed in many other ant samples from southeastern United States, it is possible that this ant disease is much more widely distributed than has been reported in the literature (Sánchez-Peña et al., 1993). Researchers that routinely examine large samples of ants may be able to identify diseased individuals from other locations and ant species.

The area behind the USDA-ARS laboratory in Gainesville, FL, where *M. durum*-infected *S. invicta*, *S. carolinensis*, and *P. membranifera* were collected, has had its ant populations closely scrutinized through the years. It is also an area where residues of rejected *S. invicta* colonies have been disposed. Whether the occurrence of three *M. durum*-infected species within this small area, as well as in a pasture in Houston, AL, indicates some level of transmission of the disease among the observed species is not known. However, *M. durum* has only now been identified from *S. invicta* after many years of intensive research on this ant in numerous locations throughout the southeastern United States. It is possible that this fungus has been acquired by *S. invicta* from native ants. This suggests that other organisms may evolve into biological control agents of *S. invicta*. Such host switching is apparently a common occurrence among other parasites (Strong et al., 1977).

Estimation of prevalence rates was difficult because most of the *M. durum*-infected ants were not collected from a known nest, but from traps. Some of these estimates were based on very small samples, whereas others are from larger samples that allow greater confidence in the results. More samples are needed before reliable prevalence rates can be obtained for some of the species reported here. For instance, within the Ames Plantation, *M. durum*-infected ants were collected from three loca-

tions along a 6.4 km land span including pastures, cultivated fields, and forested areas. A thorough examination of ants from the Ames Plantation, TN, may provide a more precise estimation of *M. durum* prevalence rates in *P. tysoni* and *P. vividula* populations. Such examination may also reveal other ant species infected with this fungus at that location.

The relatively high prevalence of *M. durum* in *S. carolinensis* populations in northern Florida and southern Alabama suggests that this ant species may serve as a reservoir of the disease in the region, and as a possible inoculum source for other ant populations. *S. carolinensis* is known as a thief ant that usually nests in or near other ants and robs food and brood from their nests (Hölldobler and Wilson, 1990). This behavior may provide opportunity for disease transmission between *S. carolinensis* and other ants.

*Pogonomyrmex badius* colonies collected in Florida contained *M. durum*-infected young adult workers. This suggests that the infection is carried over from the pupal stage and that the disease progresses in the adult ants as they age. Whether this phenomenon is common to the other infected species is not known. However, development of *M. durum* in host ants may cause higher mortality rates, at least under stress conditions as observed with *S. invicta* workers during the transfer from the field to the laboratory. Stressed *M. durum*-infected *P. badius* also died at a faster rate in the laboratory.

Except for an observation in April 2000 [*S. carolinensis* from Newberry, FL (N 29.651°; W 82.656°)], all the disease observations reported here were made in late summer and fall. Previous reports of this disease also point to a predominant occurrence during this time of the year (Espadaler, 1982). This author also suggested that winter mortality could affect *M. durum*-infected ants more strongly than uninfected ants.

Whether the recent increase in detection of diseases in fire ant populations (Pereira et al., 2002; Valles and Pereira, 2003a; Williams et al., 1998, and the present publication) is an indication of a trend toward increased disease susceptibility of the North American *S. invicta* population is not known at this time. It has been suggested that recent trends toward the predominance of polygynous colonies in this ant species may be associated with higher prevalence of the microsporidium *T. solenopsae* in fire ant populations (Oi et al., 2004). This occurs because of a lower aggressiveness among ants from different mounts and an increased contact and exchange of individuals among neighboring nests.

Discovery of yet another disease of red imported fire ants in a population that has been under close observation by several researchers over a long period of time may suggest an increase in parasitism, perhaps derived from other ants (*P. membranifera*, *P. bicarinata*, and *S. carolinensis*). After >70 years of unchecked expansion in the United States, the imported fire ant population may

have acquired diseases from the native ants. Whether this hypothesis is correct, and whether a newly acquired, or increasing disease susceptibility can slow the fire ant expansion, or decrease its dominance in the infested areas, needs further investigation. Also, stress factors, such as parasitoid biocontrol agents (Graham et al., 2003; Porter et al., 2004), new chemical pesticides, weather conditions, or the interactions of some of these factors (Valles and Pereira, 2003b), may be important in determining increased susceptibility to diseases. Because parasite populations increase with an increase in the host range area (Strong et al., 1977), the large infestation area may explain an increase in disease incidence on *S. invicta* populations. Exposures to increasing number of parasites, may offer hopes for containment of the fire ant population in North America.

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