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Observations on questing activity of adult Gulf Coast ticks, *Amblyomma maculatum* Koch (Acari: Ixodidae), in Mississippi, U.S.A.

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Abstract

During August of 2008 and 2009, observations were made on the questing heights, behaviors, and spatial distribution of adult Gulf Coast ticks, *Amblyomma maculatum*, in a plot near Gautier, Jackson County, Mississippi, U.S.A. Ticks were not evenly distributed in the plot, being found mostly on torpedograss and/or wiregrass along and in a small dirt field road. Adult ticks were visually observed questing on three different plants: torpedograss, *Panicum repens*, wiregrass, *Aristida stricta*, and Johnsongrass, *Sorghum halepense*, all but the last of which have small-diameter stems and thin, pointed leaves. Ticks were located at or near the tips of the plants (2-tail binomial probability, $p = 0.0074$). Observed questing heights ranged from 20–75 cm, with an average of 36 cm. Nine of 15 ticks (60%) seen questing were oriented head upward, while 6 (40%) were head-down. Limited mark-release-recapture observations were made in the study site, using ticks collected from the field road. Of 27 ticks marked and released, 15 were recaptured in three samples spanning a 24-d period. Of these, 5 had moved closer to the dirt road where they were originally captured and 2 farther away.

Key words: Gulf Coast tick, *Amblyomma maculatum*, questing behavior, Mississippi

Introduction

The Gulf Coast tick (GCT), *Amblyomma maculatum* Koch (Acari: Ixodidae), is a large and aggressive tick species occurring from parts of South America northward into the United States, where it commonly may be found along the southern Atlantic and Gulf coasts from North Carolina to Texas (Bishopp & Hixson 1936; Cooley & Kohls 1944; Bishopp & Trembley 1945). There are also occasional reports of the species from inland and northern states (Goddard & Norment 1983; Snoddy & Cooney 1984), and a well-established population occurs in Oklahoma and Kansas (Semtner & Hair 1973). Recently, the GCT has emerged as an arthropod of increasing medical and veterinary importance, being known to transmit *Rickettsia parkeri* to humans, causing a condition sometimes called "American boutonneuse fever" (Goddard 2004; Paddock *et al*. 2004), the protozoan *Hepatozoon americanum* to dogs (Ewing & Panciera 2003), and potentially the agent of heartwater, *Ehrlichia ruminantium*, to ruminants (Uilenberg *et al*. 1982).

GCT thrive in coastal uplands and tall-grass prairies, where the immature stages feed on a variety of birds and small rodents and adults feed on large mammals, such as cattle and deer (Bishopp & Hixson 1936; Cooley & Kohls 1944; Bishopp & Trembley 1945). High populations of GCT have been reported from coastal Mississippi, with *R. parkeri* infection rates as high as 40% (Paddock *et al*. 2010). Ticks use at least two strategies for locating potential targets for blood meals, namely ambush and hunter strategies (Sonenshine 2005). Ambush strategy involves "questing" by climbing onto the tips of vegetation to wait for passing hosts, while hunter strategy (*e.g.*, some of the large,

aggressive *Hyalomma* ticks of the Middle East) involves running rapidly across the ground to find and attack hosts. Most hard ticks in the U.S. utilize the ambush strategy, although some species, like the lone star tick, *Amblyomma americanum* (Linnaeus), may use a combination ambush/hunter strategy (Sonenshine 2005). Little is known about the questing behavior of the GCT and, specifically, what plants they quest on, heights of questing, and their ambush or hunter strategies for finding hosts.

Methods and materials

Adult GCT were collected with a 1-m^2 drag cloth in a .25 ha plot within the Sandhill Crane National Wildlife Refuge (SCNWR), near Gautier, Jackson County, Mississippi, during August (peak of their activity) of 2008 and 2009. SCNWR was chosen because of its known high populations of GCT. The site, approximately 40 m long and 62.5 m wide, is a pine savannah containing mostly wiregrass, *Aristida stricta,* gallberry, *Ilex glabra,* and invasive torpedograss, *Panicum repens,* along and in a dirt field road (Figure 1). Twenty to 30-year-old slash pines, *Pinus elliottii,* lined the edges of the road. Other vegetation sparsely scattered within the plot or along the edge of the road included common ragweed, *Ambrosia artemisifolia,* longleaf pine, *Pinus palustris,* and Johnsongrass, *Sorghum halepense.*

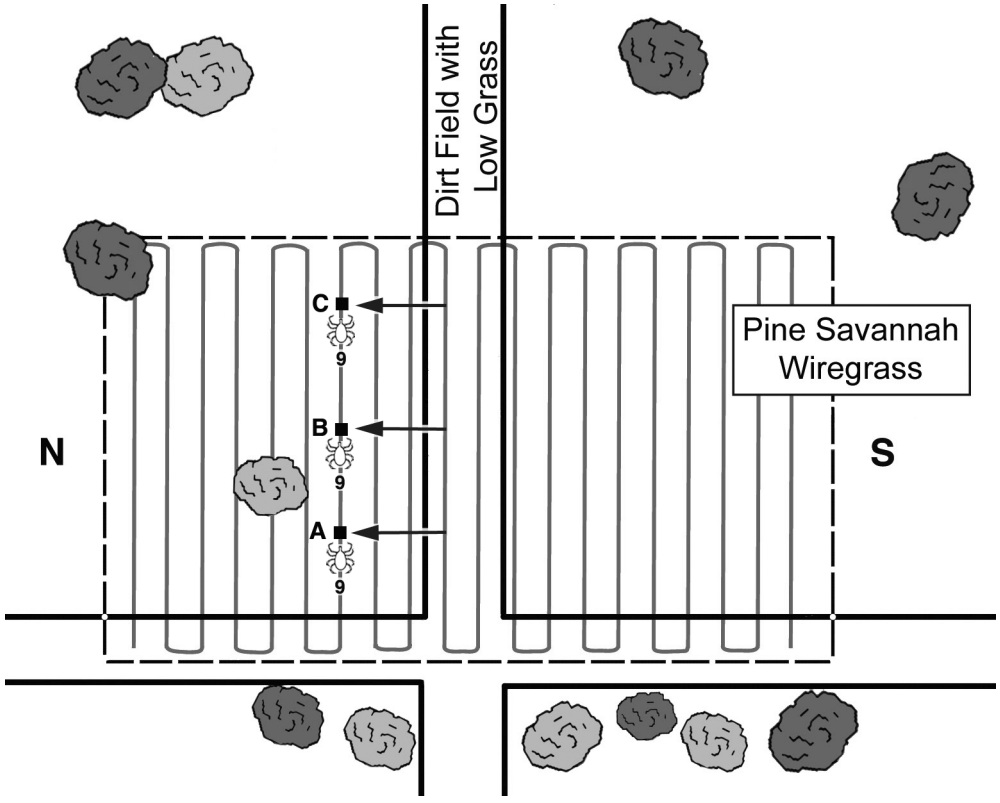

FIGURE 1. Plot within Sandhill Crane National Wildlife Refuge where questing and mark-release-recapture observations were made. Study site with release sites "A," "B," and "C" of Gulf Coast ticks and the path of drag cloth sampling (lines) for released ticks.

Spatial distribution (clustering). Once in both 2008 (Aug 20) and 2009 (Aug 18), ticks were carefully collected in the plot between 9:00 and 10:00 am, checking the cloth every 5 m, and locations of ticks were marked on a map. Nine swaths through the plot, approximately 4.5 m apart, were randomly chosen and sampled by drag cloth. Notes were made concerning plant(s) on which ticks were questing, and heights questing, if observed. When ticks were seen questing, the entire plant was carefully examined from the ground up for additional questing specimens.

Questing heights. During the collecting events described above, careful efforts were made to observe GCT questing on vegetation and to measure questing height (Figure 2). In addition, notes were taken as to other nearby vegetation.

FIGURE 2. Measuring questing heights of adult Gulf Coast ticks on vegetation.

Mark-release-recapture. On Aug 20, 2009, three areas (subplots) along the dirt road were selected within the site and 9 ticks collected from each one. These 27 ticks were marked dorsally with a single dot of Testor's® model paint (Hobby Lobby, U.S.A.), as reported previously (Goddard 1993), and 9 were released at each of three sites (A, B, C) 7.5 m from the road (Figure 1). Five GCT, collected outside the study site, were similarly marked with the same paint, kept in the lab in a humidity chamber, and observed for 4 weeks with no apparent ill effects. Efforts to recapture the ticks were made at 3 intervals roughly one week apart (range 4–10 days) to determine movement patterns, if any. Upon post-release sampling, the plot was systematically sampled with the drag cloth, using twenty swaths approximately 2.5 m apart. At each sampling event, dragging was initiated on the south edge of the plot and ended at the north edge; any marked ticks were released back into the plots at the place of capture. In addition, the collector's (JG) pants and clothing were examined for any ticks crawling on them.

Data analysis. The average distance moved (distance south positive) by marked ticks from the release line at each of the three sampling dates was regressed against sampling date using the number of ticks recaptured as weights. For b slope of regression, H_0: $b \leq 0$ and H_A: $b > 0$ (*i.e.*, a one-tail test).

Results and discussion

Thirty-five adult GCT were collected at the site during August 2008 and 23 were collected during August 2009. Ticks were not evenly distributed in the plot, being found almost entirely on torpedograss and/or wiregrass along and in the dirt field road. During the mark-release-recapture study only one specimen was collected out in the savannah, which consisted almost entirely of wiregrass. Other ticks, such as the American dog tick, *Dermacentor variabilis* (Say), have been previously shown to prefer trails and road edges (Smith *et al*. 1946; Newhouse 1983). However, some of this clustering phenomenon may be a collecting artifact. Edge habitat may be more easily sampled than tall savannah grass. Earlier studies have shown that *D. variabilis* may be easier to collect along trails than in other habitats due to limited contact of the cloth with questing ticks in high vegetation (Carroll *et al*. 1991).

Questing heights. Adult GCT were seen questing on three different plants, *A. stricta*, *P. repens*, and *S. halepense* (Table 1), all but the last of which have small-diameter stems and thin, pointed leaves. Only ticks that were visible were included in the questing height analysis, and it is possible that some were questing at lower levels and not seen. However, of those observed, specimens seemed preferentially located at or near the tips of the plants (2-tail binomial probability, $p = 0.0074$). Questing heights ranged from 20 to 75 cm, with an average of 36 cm. Nine of 15 ticks (60%) seen questing were oriented head upward, while 6 (40%) were head-down.

Mark-release-recapture. In the recapture experiment, 20 GCT were collected during the first post-release sampling period (10 days), and 7 of them (35.0%) had been marked with model paint (Table 2). At the second sampling period, two weeks post-release, 13 GCT were collected, of which 6 had been marked (46.2%). At the third sampling period, 24 days post-release, 8 GCT were collected and 2 of these had been marked (25%). No ticks were seen crawling on the collector's clothing during any of the 3 sampling events. Overall, of marked ticks recaptured, 5 had moved closer to the dirt road where they were originally captured and 2 moved further away (Table 2). Interestingly, 2 specimens had moved all the way back to the road from which they were first captured. The average location of recaptures moved toward the dirt road ($t = 3.23$, $\gamma = 1$, $p = 0.096$). These data are limited by low numbers, but movement toward roadsides has been reported in *D. variabilis* (Smith *et al*. 1946), so it is not unreasonable that such behavior could occur in other ixodid ticks. Guanine and uric acid have been identified as assembly pheromones in hard ticks and are

deposited by the ticks as they move around in their environments, causing an "arrest" of movement and, thus, assembly at certain areas such as roadsides (Sonenshine 2004; Yoder *et al*. 2008).

TABLE 1. Direct observation of adult GCT questing, Sandhill Crane NWR, Gautier, MS, 2009.

Date	Number ticks observed	Plant	Height	Where on plant	Orientation	Higher grasses/ weeds nearby
8/18	1	*Panicum repens*	37 cm	Tip	Head up	Yes
8/18	1	*Panicum repens*	34 cm	Tip	Head up	Yes
8/18	1	*Panicum repens*	39 cm	Tip	Head up	Yes
8/20	1	*Panicum repens*	20 cm	Tip	Head up	No
8/20	1	*Panicum repens*	20 cm	Tip	Head up	No
8/20	1	*Sorghum halepense*	75 cm	Tip	Head up	No
8/20	2*	*Panicum repens*	45 and 40 cm	1 near tip and one 8 cm from tip	1 head up and 1 head down	No
8/20	1	*Aristida stricta*	30 cm	Tip	Head down	Yes
8/20	1	*Aristida stricta*	30 cm	Tip	Head down	Yes
8/30	1	*Panicum repens*	33 cm	Tip	Head up	Yes
9/3	1	*Panicum repens*	30 cm	Near tip	Head down	Yes
9/3	2*	*Panicum repens*	40 and 32 cm	1 near tip and one 10 cm from tip	Both head down	Yes
9/13	1	*Aristida stricta*	40	Near tip	Head up	Yes
Total	**15**	**Avg Height Questing 36 cm**				

*On same plant

TABLE 2. Mark-release-recapture results, showing movement by adult ticks. Ticks were originally collected from vegetation in the dirt road.

First sampling, 10 d post-release			
Release site	**# originally released**	**# recaptured**	**Where recaptured**
A	9	1	Release point
B	9	2 2	Release point 2.5m north (away from road)
C	9	1 1	Release point 2.5m southeast (toward road)
Second sampling, 14 d post-release			
A	9	0	--
B	9	2 2 1	Release point 2.5m south (toward road) In dirt road (original capture area)
C	9	1	Release point
Third sampling, 24 d post-release			
A	9	0	--
B	9	1 1	Release point In dirt road (original capture area)
C	9	0	--

Further investigations are needed to elucidate GCT questing behaviors, vegetation associations, and spatial distribution (clustering) in nature. Understanding these factors may lead to development of better avoidance and personal protection measures, as well as GCT management and control techniques.

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