Survival and Control of the Boll Weevil, *Anthonomus grandis*, Around Overwintering Habitat in the United States

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ABSTRACT

A field and a laboratory experiment were performed to evaluate factors that influence the overwinter survival of boll weevil, Anthonomus grandis Boheman, populations. Spring surveys of boll weevil densities were conducted as indicators of overwintering survival in 1994, 1995, 1996 and 1997 in four Arkansas counties. Approximately 1000 traps were placed adjacent to defined overwintering habitats near cotton fields. Traps near forested habitats consistently contained the highest average boll weevil catches among habitat types, while grassy field borders generally had the lowest mean trap captures. Trap captures near tree-line and brushy field border habitats were moderate. Larger and more significant differences between habitat types occurred during springs following colder winters. In the laboratory experiment, diapause-conditioned boll weevils were subjected to freezing temperatures within containers submerged in a cold circulation bath and held for one to eight hours. Results showed that temperature, duration of exposure, moisture and substrate were significant factors in boll weevil mortality. Mortality increased with temperature reduction and exposure time. The presence of dry substrate significantly improved weevil survival over those in empty containers at -10.0 and -12.5°C and over those in moist substrate at -5.0 to -12.5°C. Over 70% of weevils were able to survive temperatures of -2.5°C for eight hours, in either moist or dry substrate, while high (>75%) mortality occurred at -10°C or colder temperatures in moist substrate, even for short (1 hour) exposures. These results indicate that temperature and litter type within overwintering habitat microsites are important indicators of boll weevil survival.

Introduction

The need for improved control and the boll weevil eradication program has stimulated efforts to predict boll weevil, *Anthonomus grandis* Boheman, winter survival patterns. Overwintering survival is important because it largely determines the magnitude of early cotton field populations (Parajulee *et al.*, 1996; Rummel and Carrol, 1993; Fuchs and England, 1989) especially in cotton production areas where boll weevil winter mortality is significant. These predictions could help focus strategic planning efforts for boll weevil control. The formation of new strategies for boll weevil control can supplement or improve the cultural, mechanical and chemical practices already being used to control this insect (El-Kissy and Myers, 1996).

Boll weevils spend the winter as diapausing adults in natural or man-made habitats near cotton fields (Brazzel and Newsom, 1959), preferably within deciduous litter layer (Beckman, 1957; Fye *et al.*, 1958; Rummel and Adkisson, 1970) and emerge in the spring. Spring captures from Grandlure-baited traps are strong indicators of emerging boll weevil populations (Carroll and Rummel, 1985).

Climatic factors, such as the severity of winter freezes, are important indicators of boll weevil winter survival and thus spring infestations (Pfrimmer and Merkl, 1981; Gaines, 1943; Bondy and Rainwater, 1942). Many investigators have also found relationships between weevil survival and exposure to sub-freezing temperatures in laboratory tests (Sorenson and House, 1995; Slosser *et al.*, 1996; Sorenson *et al.*, 1996; Sorenson and George, 1996). The presence of moisture also influences boll weevil winter survival in freezing temperatures. In the relatively arid climate of the Texas rolling plains, greater winter rainfall is associated with increased survivorship (Price *et al.*, 1985; Stone *et al.*, 1990; Parajulee *et al.*, 1996) apparently due to reduced freeze-drying affects. Dry, cold winter weather has also been highly lethal to Mississippi weevil populations (Pfrimmer and Merkl, 1981). On the other hand, Taft and Hopkins (1966) reported that weevil mortality in South Carolina was highest under excessively moist conditions and in southeast Missouri, over-winter survival was low in wet, poorly drained areas (Sorenson and George, 1996).

The main objective of this study was to determine the relationship between sub-freezing temperatures, exposure time and presence of moisture in leaf litter substrate and boll weevil survival. A second objective was to evaluate various habitat types as to their potential to provide protection from winter conditions to boll weevil. It was hypothesized that cold, wet conditions would increased the mortality rate of overwintering boll weevils and that favorable

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overwintering habitats could, therefore, provide some protection for increased survivorship.

Methods

Field Population Survey. A survey of overwintering boll weevils was conducted in four Arkansas counties (Craighead, Crittenden, Lonoke and Mississippi) during the springs of 1994, 1995, 1996 and 1997. Approximately 1000 boll weevil phermone traps were placed adjacent to defined overwintering habitats near cotton fields. Survey areas included approximately 28 square kilometers in each county. Defined habitat types included: (1) forest (2) tree line - large trees; (3) tree line - small trees; (4) field border - brush; (5) field border - grass. Traps were monitored by counting and discarding trapped weevils every two weeks from March to June. Occasionally, a trap would be down due to severe weather or farm machinery: this data was discarded. Trap data from Crittenden country in years 1996 and 1997 were not included in the analysis due to an initiation of a boll weevil control program in this area. Differences in spring trap capture associated with overwintering habitat type (blocked by census date) were tested using the General Linear Regression Method (GLM). Habitat type means were separated using Fisher's protected least significant difference (LSD) ($\propto$ = 0.05) (SAS Institute, 1990).

Laboratory Survival Study. Adult boll weevils were collected from pheromone traps near cotton fields or were allowed to emerge from cotton squares placed in plastic ventilated cages in the laboratory. All collections were made in September and early October 1997, in Lonoke County, Arkansas. Collected and newly emerged weevils were induced into a diapause state using techniques described by Slosser *et al.* (1996).

Boll weevil mortality patterns in sub-freezing temperatures (0, -2.5, -5.0, -7.5, -10.0, –12.5 and -15$_o$C) were examined for three experimental substrate types and four durations of exposure. Weevils (n=20) were placed inside a 29.6 ml clear plastic cup with a paper lid. Cups were immersed into a circulating cold bath in a container attached to the tops of the cold baths (Forma Scientific Model 2067 CH/P, Forma Scientific, Marietta, OH) to obtain temperatures tested. A solution of equal parts ethylene glycol antifreeze and water was used as the cooling solution in the cold baths. Temperatures within the cups were verified using a thermocouple attached to an electronic data recorder (Stowaway$_{TM}$ XTI, Onset Computer Corporation, Pocasset, MA). Substrate types included moist leaves, dry leaves and no leaves. Leaf fragments were placed in plastic cups for the moist and dry substrate treatments. Leaf fragments were collected from partially decomposed leaves (2-5 cm2 fragments) beneath a nearby oak stand. Duration of exposure were 1, 2, 4 and 8 consecutive hours. Treatments at each temperature were replicated four times. Boll weevil survival was evaluated 16 to 24 hours (overnight) after cups were removed from the cold bath. Only individuals that were able to stand and walk were considered as having survived exposure to freezing temperature. Boll weevil mortality data were tested using a GLM test. Duration of exposure and interaction of main effects treatment means were separated using Fisher's protected LSD ($\propto$ = 0.05) (SAS, Institute 1990).

Results and Discussion

Field Population Survey. Mean boll weevil trap catches were greatest in 1995, followed by those of 1994. Years 1996 and 1997 had relatively low weevil trap catches. These differences were probably due to the severity of winter temperatures preceding the spring surveys: the winters of 1996 and 1997 were relatively cold and severe, while the winter of 1995 was relatively warm. Boll weevil overwinter survival is generally high when winter temperatures are mild.

There was a significant spatial correlation between spring trap catches and overwintering boll weevil habitat types. Traps associated with forested habitats had the greatest number of boll weevils, significantly greater than those of other habitat types. The smallest trap catches were generally associated with grassy field borders, while tree lines and brushy field borders had moderate boll weevil densities (Table 1).

Forested habitats provided the most favorable habitat for overwintering boll weevils (as seen by greater spring trap catches near these areas), probably because of the relatively thick litter layer beneath the deciduous stand of trees. Boll weevils overwinter within the litter layer, where temperatures can be 10 to 20$_o$C warmer than sub-freezing ambient temperatures. In contrast to leaf litter, light grass cover does not provide much insulation from freezing air (ambient and within-litter temperatures in a forested, tree-line and grassy areas measured with thermocouples attached to dataloggers, unpublished data). Although tree-line habitats also have a layer of leaf litter, these linear habitats generally have less total area (thus fewer favorable microsites) than that of a forest. This shape difference may explain why spring weevil densities were lower near tree-line than near forest habitats.

Laboratory Survival Study. Temperature, substrate type and length of exposure were all significant factors in boll weevil survival. Interactions between substrate and exposure, as well as among temperature, substrate and exposure time were also significant elements in mortality. Weevil mortality increased with temperature reduction and increased exposure time and was greater in moist substrate than in dry substrate at all sub-zero temperatures (Table 2).

At the warmest (0.0 and -2.5$_o$C) temperatures tested, exposure time and substrate type were not significant factors of weevil mortality. Most (>70%) weevils were able to survive freezing temperatures of –2.5$_o$C or higher for up to eight hours duration, even when exposed within moist substrate and at -5.0oC in dry or no litter. Slosser *et al.* (1996) reported similar results, with over 90% of diapausing boll weevils surviving an eight-hour exposure to temperatures of -5oC or warmer. Although our mortality rates were about 5 to 20% higher than those of Slosser *et al.* (1996) were, these differences were probably due to the temperature measurement since our test measured temperatures directly in the cups and not on the machine monitor. In these tests, temperature probes indicated a 1.5 to 2.5 degree difference between the temperatures inside the cups and the cooling solution. No weevils survived a one-hour exposure at the coldest temperature (-15.0oC) tested, regard of the presence or dryness of leaves in the cups (Table 2). Sorenson and House (1995) reported greater survival at this temperature in a similar study, with over 20% survival following a 1.5-hour exposure and three hours required for complete mortality.

Boll weevil mortality levels in empty cups were not significantly different from those in dry substrate, for the temperature exposures ranging from 0 to -7.5oC. However, at colder temperatures (–10 and –12.5oC), weevils in dry substrate had significantly greater survival than those exposed in empty cups. These comparisons indicate that the dry substrate increased boll weevil survival at intermediate sub-freezing temperatures, probably due to a conductivity effect. This difference is important because several authors have reported boll weevil mortality estimates based upon laboratory results from cold exposures within empty containers (Slosser *et al.*, 1996; Sorenson *et al.*, 1996; Sorenson and George, 1996; Sorenson and House, 1995). However, in the field, weevils overwinter under a cover of plant litter (Bondy and Rainwater, 1942). These results indicate that laboratory techniques used to measure weevil survival under field conditions at sub-freezing temperatures may result in extra-conservative estimates of weevil survival.

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Table 1. Mean spring boll weevils trap captures from defined habitat types in Arkansas for four years. Mean trap catches within columns sharing the same letter were statistically indistinguishable (Fisher's protected LSD, α=0.05).

Habitat Type	Year (Survey Period - March through June)				
	1994	1995	1996	1997	All Four Years
Forest	20.95 a	46.33 a	9.33 a	8.26 a	24.80 a
Tree line - large trees	8.50 c	31.30 b	6.02 b	4.77 b	14.39 b
Tree line - small trees	10.43 b	30.00 bc	3.13 cd	3.66 c	13.66 bc
Field border - brush	8.22 c	26.28 cd	3.49 c	4.14 bc	12.71 c
Field border - grass	5.84 d	21.82 d	1.92 d	3.67 c	9.73 d

Table 2. Mean boll weevil mortality (%) following exposure to sub-freezing temperatures in no, dry leaf litter and moist leaf litter substrates for four exposure times. Substrate type means (for all four exposures) sharing the same letter were statistically indistinguishable (Fisher's protected LSD, α=0.05). Exposure time means (for all three substrate types) sharing the same letter were statistically indistinguishable (α=0.05).

Exposure	Leaf Substrate Type			
(Hours)	None	Dry	Moist	All Types
	Temperature = 0.0°C			
1	3.8	12.4	2.6	6.3a
2	7.2	6.4	2.6	5.4a
4	12.5	9.1	1.3	7.6a
8	11.8	1.3	3.8	5.6a
All	8.8a	7.3ab	2.6b	
	Temperature = -2.5°C			
1	18.5	24.5	5.0	16.0a
2	16.8	20.3	6.6	14.6a
4	23.9	7.4	7.5	12.9a
8	13.7	9.9	26.6	16.7a
All	18.2a	15.5a	11.4a	
	Temperature = -5.0°C			
1	15.2	16.8	27.2	19.8a
2	14.5	5.2	29.5	16.4a
4	15.0	9.7	70.5	31.7b
8	20.6	14.6	77.0	37.4b
All	16.3a	11.6a	51.1b	

Exposure	Leaf Substrate Type			
(Hours)	None	Dry	Moist	All Types
	Temperature = -7.5°C			
1	15.8	21.5	23.5	20.2a
2	19.8	18.8	78.8	39.1b
4	18.2	15.8	93.7	42.5b
8	36.5	22.2	100.0	52.9c
All	22.6a	19.6a	74.0b	
	Temperature = -10.0°C			
1	30.2	25.8	57.1	37.7a
2	36.2	28.7	100.0	54.9b
4	95.6	73.9	100.0	89.8c
8	100.0	93.0	100.0	97.7c
All	65.5a	55.4b	89.3c	
	Temperature = -12.5°C			
1	70.0	37.9	97.6	68.5a
2	100.0	100.0	100.0	100.0b
4	100.0	100.0	100.0	100.0b
8	100.0	100.0	100.0	100.0b
All	92.5 a	84.5 b	99.4c	
	Temperature = -15.0°C			
1	100.0	100.0	100.0	