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PERFORMANCE OF STERILE *CACTOBLASTIS CACTORUM* (LEPIDOPTERA: PYRALIDAE) FEMALES IN LURING MALES TO TRAPS

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ABSTRACT

Cactoblastis cactorum (Berg) (Lepidoptera: Pyralidae) is renown for its control of invasive cacti (*Opuntia* spp.). Its accidental arrival in Florida and its rapidly expanding range along the Gulf coast pose an imminent threat to native *Opuntia* spp., especially in the southwestern U.S. and Mexico. Adequate survey techniques are crucial in order to delineate the rate of spread of this invasive species. Virgin female-baited sticky traps have been effective in detecting *C. cactorum* adult males in areas where visual surveys failed to detect larval damage. However, the use of fertile females in traps placed beyond the currently infested area is discouraged because an escaped fertile female might establish a breeding population and expand the infested area. In this study we compare the attractiveness and the longevity of fertile and irradiated (sterile) females deployed as bait in traps. Traps baited with females sterilized with gamma radiation were as effective as traps baited with unirradiated (fertile) females in detecting populations of feral *C. cactorum* male moths.

Key Words: invasive species, *Opuntia*, cactus moth, survey, SIT

RESUMEN

Cactoblastis cactorum (Berg) (Lepidoptera: Pyralidae) es un insecto bien conocido por su efectividad como agente de control biológico de especies invasoras de cactus (*Opuntia* spp.). La llegada accidental de *C. cactorum* al estado de Florida y su rápida expansión a lo largo de la costa del Golfo de Méjico, representan una amenaza real para las especies nativas de *Opuntia* spp., especialmente en areas del suroeste de los Estados Unidos y Méjico. El desarrollo de técnicas adecuadas de detección es de suma importancia para poder delinear la distribución y expansión de esta especie. Trampas que utilizan hembras vírgenes como cebo atractivo han sido efectivas en la detección de machos de *C. cactorum* en areas donde no se ha detectado la presencia de esta especie por daño en plantas. Sin embargo, el uso de hembras vírgenes fértiles como cebo en trampas colocadas fuera del area de infestación no es recomendable debido a que si las hembras se escapan podrian establecer un nuevo foco de infestación. En este estudio, la atractividad y longevidad de hembras fértiles como cebo en trampas se comparó con la atractividad y longevidad de hembras irradiadas (estériles). Las trampas con hembras estériles resultaron igualmente eficaces en su habilidad de detección y captura de machos silvestres de *C. cactorum*.

Translation provided by author.

The cactus moth, *Cactoblastis cactorum* (Berg) (Lepidoptera: Pyralidae) is renown for its control of invasive cacti (*Opuntia* spp.) in Australia (Dodd 1940), and it has been cited as one of the most successful examples of biological control of weeds (Sweetman 1936). However, the accidental arrival of *C. cactorum* in Florida (Habeck & Bennett 1990; Dickel 1991), first detected in 1989, has raised concerns about its potential impact on native *Opuntia* in the southern United States and Mexico (Johnson & Stiling 1998; Zimmermann et al. 2001). Pemberton (1995) estimated that infestations of *C. cactorum* should be able to survive as far north as Charleston, South Carolina, San Antonio, Texas, and the lower altitude areas of New Mexico, Arizona and California. Current distributional information published in Hight et al. (2002) suggests that the range of *C. cactorum* is expanding by 50-75 km per year. Although specific interactions cannot be predicted at this time, establishment of *C. cactorum* in the southwestern U.S. and Mexico could have devastating effects on the landscape and biodiversity of native desert ecosystems, and on the forage and vegetable *Opuntia*

industries in these areas (Soberón et al. 2001; Zimmermann et al. 2001).

No satisfactory method of control has been identified for *C. cactorum* (Habeck & Bennett 1990; Stiling 2002). Because many of the *Opuntia* species in the U.S. are associated with sensitive ecological areas, widespread use of pesticides is not recommended (Leibee & Osborne 2001). The use of insect pathogens does not appear to hold promise, as most Lepidopteran viruses and parasitic nematodes have non-selective modes of action and could negatively impact other native Lepidoptera present in the area of infestation. In its native habitat in South America, a number of natural enemies have been found attacking *C. cactorum*, including members of the families Braconidae, Ichneumonidae, Chalcididae and Tachinidae (Habeck & Bennett 1990; Pemberton & Cordo 2001). However, most of the species are generalist parasitoids and their host range and potential non-target effects would have to be carefully scrutinized before their release would be approved in the U.S. (Pemberton & Cordo 2001). Our research is focusing on the potential application of the Sterile Insect Technique (SIT) and the phenomenon of inherited or F_1 sterility to help study and manage the spread of *C. cactorum* (Carpenter et al. 2001a, b). SIT/F_1 sterility is a species-specific pest control tactic that could be used to eradicate new or localized infestations, protect environmentally sensitive areas, or establish a barrier to prevent further geographic range expansion.

The ability to quickly detect new infestations, accurately delimit the size of an infestation (i.e., the leading edge of an expanding population), and assess population trends are of critical importance to the successful application of any strategy using SIT/F_1 sterility. Unfortunately, although females produce a pheromone that attracts males, no synthetic pheromone has been identified yet for *C. cactorum*, which makes continuous insect monitoring especially difficult. Hight et al. (2002) reported on the use of sticky traps baited with virgin female *C. cactorum* to corroborate field damage and better understand the current distribution of the species in Florida and Georgia. However, trapping beyond the leading edge of the currently infested area using fertile females is not recommended. Here, we compare the ability of laboratory-reared, virgin fertile females to attract male *C. cactorum* into sticky traps with that of females treated with a sterilizing dose of 200 Gy. The results are discussed in the context of developing an SIT program for *C. cactorum* management in the U.S. and elsewhere.

Materials and Methods

Test Insects

Cactoblastis cactorum used in these experiments came from a laboratory colony maintained at the USDA-ARS Crop Protection and Management Research Unit in Tifton, Georgia. Larvae were reared on cladodes of *Opuntia ficus-indica* (L.) Miller inside plastic boxes that were kept at 26°C ± 1°C, a photoperiod of 14:10 (L:D), and 70% relative humidity as described by Carpenter et al. (2001b). As larvae matured, cocoons were collected every 2-3 days from the containers. Pupae were then extracted from the cocoons and sorted by gender.

Three "types" of adult virgin females were tested in the field for their ability to attract *C. cactorum* males to sticky traps: untreated control females (Ua♀), females that were treated with a reproductively sterilizing dose of radiation as pupae (Tp♀), and females that were irradiated and sterilized as adults (Ta♀). Untreated control females were obtained by placing pupae in a screen cage (30.5 by 30.5 by 30.5 cm) and allowing them to emerge at the above mentioned conditions. Sterile females that were treated as pupae were obtained by holding pupae in a 473 ml plastic container until pharate adults had formed inside the pupal skins. Mature pupae that were within 12 h of emerging were placed in individual 30 ml plastic cups and irradiated with 200 Gy of gamma radiation using a Cobalt^{60} Gammacell 220 irradiator (J.L. Shepherd & Associates, San Fernando, CA; dose rate of 15.47 Gy/min). Treated pupae were then held at the conditions described above and allowed to emerge. Sterile females that were treated as adults were obtained by placing pupae in a screen cage as above (for control females) and allowing them to emerge. Fully eclosed females were removed every 24-36 h, placed individually in 30 ml plastic cups, and irradiated with 200 Gy as described for pupae. For all groups, only adult females that were less than 48 h old were used in the trapping experiments.

Traps and Sites

Females, either untreated (Ua♀), treated as pupae (Tp♀), or treated as adults (Ta♀), were placed individually inside modified plastic film (35 mm photographic) canisters and used to bait Pherocon 1-C wing traps. The film canisters had two 2 by 2 cm screened windows cut into them and were provisioned with a small square of *O. ficus-indica*. A cotton wick was fitted through a hole cut in the top of the canisters to provide the females with moisture. Velcro® glued to the bottom of the canisters allowed the canisters to be attached to the inside tops of the wing traps, which also were fitted with Velcro® (Fig. 1). Canisters with females were transported to the field in a small cooler.

Experiments were conducted in the proximity of a salt marsh estuary on the southern banks of the Brunswick River in Glynn County, Georgia, west of U.S. Highway 17. A large area was chosen

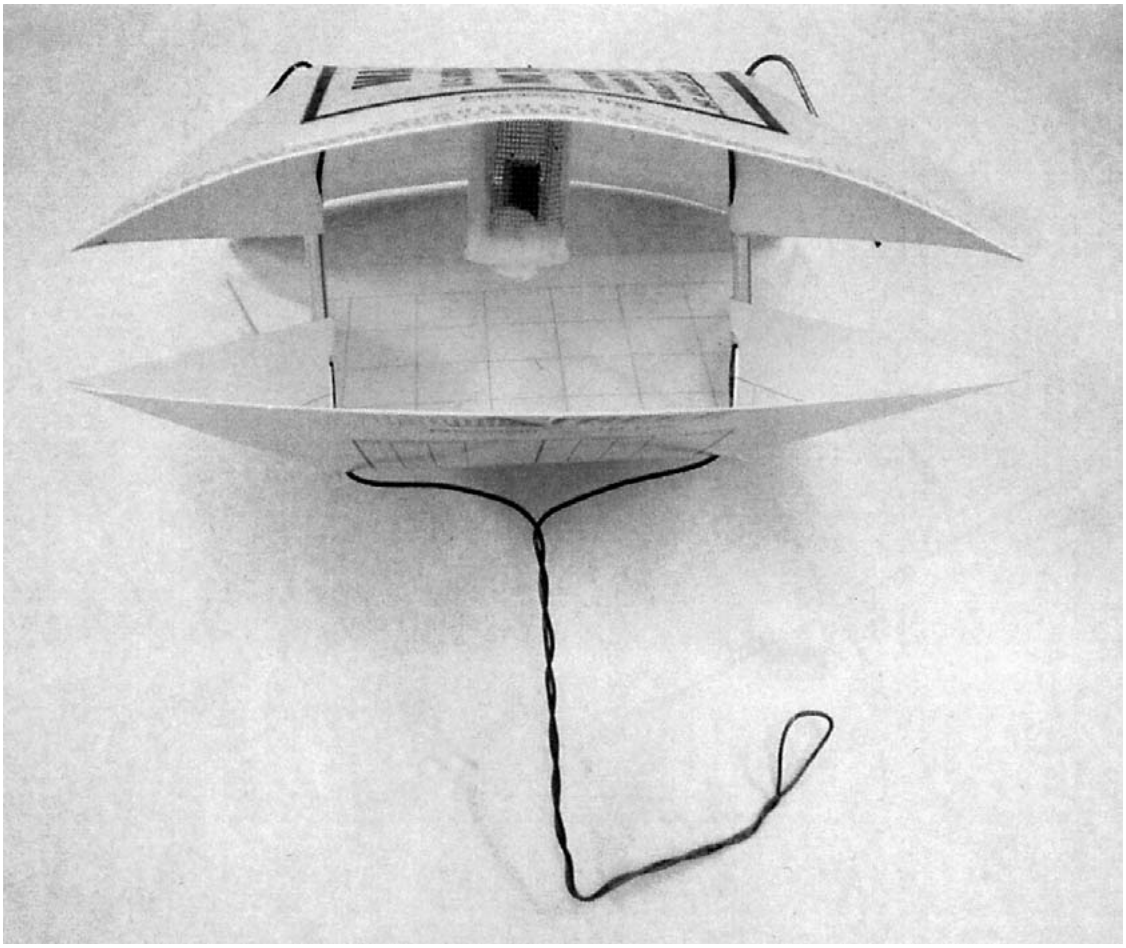

Fig. 1. Photograph of Pherocon 1-C sticky trap.

within the estuary with naturally occurring patches of *O. stricta* (Haworth) Haworth plants. Ten patches with cactus plants between 0.5-1.5 m in height were selected for the experiments. Two or three hollow metal stakes were placed in the ground at a height of approximately 0.75 m within each patch on which to attach the traps. All of the cactus patches were separated from one another by at least 10 m. The trap stakes within a patch were separated by no less than 4 m.

Overall Trap Performance

Ten Pherocon 1-C traps baited with Ua♀'s and 10 baited with Ta♀'s were deployed on 11 April 2003 and serviced every three days until 1 May 2003. Two traps, one with each type of female, were placed in each of the 10 cactus patches. During each trap servicing, the number of *C. cactorum* males captured in each trap was recorded, the traps were re-baited with canisters containing fresh Ua♀ and Ta♀, and the placement of the two treatments within each patch was alternated. The traps were serviced a total of six times.

Daily Trap Captures and Female Field Longevity

Ten untreated females (Ua♀), 10 females that had been treated with 200 Gy as mature pupae (Tp♀) and 10 females that had been treated with 200 Gy as newly emerged adults (Ta♀) were used to bait 30 Pherocon 1-C traps. Three traps, one of each type (Ua♀, Tp♀, Ta♀), were affixed to the metal stakes at random within each of the 10 cactus patches. Traps were first deployed on 19 July 2003. Cactus patches were visited every 24 h and each trap was examined to determine whether the female was alive and how many male *C. cactorum* had been captured. Traps that captured males received a new sticky bottom. Traps were then rotated clock-wise among the three trap positions within each cactus patch. Daily observations, trap servicing, and trap rotation continued until all females died.

Statistical Analysis

Trap capture data for both experiments were not normally distributed. Since $\log_{10}$ and arcsine transformations did not normalize the data, the GLM-RANK procedure was used to test for treatment effects in the daily trap capture and field longevity study, and the GLM-TTEST procedure for unequal variances was used to test for treatment effects on trap capture in the overall trap performance study (SAS 1989). The mean longevity of females used as bait in traps was analyzed using GLM and the Waller-Duncan K-ratio *t* test (SAS 1989).

Results and Discussion

In the overall trap performance trial, where traps were checked for captures every three days, we found no significant difference ($P \geq 0.2353$) between the mean (±S.D.) number of males captured in traps baited with untreated females (Ua♀ = 3.20 ± 4.24) and those baited with sterile females (Ta♀ = 3.25 ± 4.33). In the daily trap capture study, no significant differences ($P \geq 0.1039$) were seen in the overall mean (±S.D.) number of male *C. cactorum* captured in Pherocon 1-C traps baited with Ua♀ (10.7 ± 7.3), Tp♀ (7.2 ± 5.5), or Ta♀ (7.5 ± 6.7). The trend in cumulative trap captures for the traps baited with the three female types also was similar (Fig. 2). In addition, the mean (±S.D.) longevity in days for control and treated females used to bait the traps was not significantly different ($P > 0.6504$). Untreated females lived an average of 6.70 ± 0.72 d. Treated females irradiated as mature pupae or as adults lived an average of 7.56 ± 0.80 d and 7.60 ± 0.81 d, respectively.

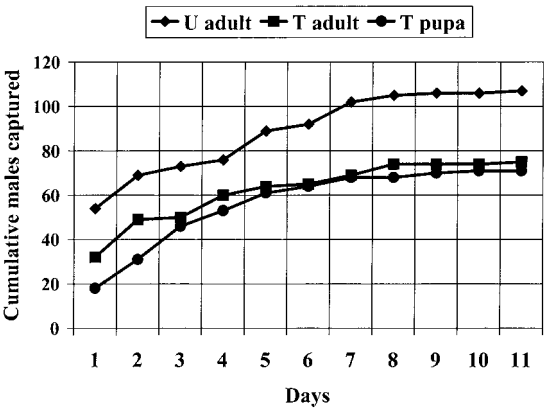

Fig. 2. Cumulative total *C. cactorum* males captured in sticky traps (n = 10 per female type) baited with *C. cactorum* females that were untreated (U adult), treated with a reproductively sterilizing dose of radiation (200 Gy) as pupae (T pupa), and treated with a reproductively sterilizing dose of radiation (200 Gy) as adults (T adult). All females were caged in the trap for the length of their life.

Hight et al. (2002) reported finding infestations of *C. cactorum* along the coast as far north as Folly Island near Charleston, South Carolina and as far west as St. George Island, Franklin County, Florida. Several previously unreported inland infestations were also identified in Orange and Osceola Counties halfway "up" the Florida peninsula. *Cactoblastis cactorum* infestations were discovered by looking for damaged cladodes exhibiting mucilage from larval entry holes or "whitened" cladodes that had fallen to the ground (Stiling 2002). Surveys are often visually based on large *Opuntia* species that are common in yards, such as *O. ficus-indica* and *O. stricta*. However, searching for damage on smaller *Opuntia* species that are common in natural areas and roadsides is difficult because they are often hidden in native vegetation. Virgin female-baited sticky traps were able to detect the presence of *C. cactorum* at a beach site with numerous low growing *O. pusilla* (Haworth) Haworth but where no larval damage was yet evident. During 2003, several additional surveys for plant damage by *C. cactorum* were conducted along the west coast of Florida (St. Joe Peninsula, Mexico Beach, Panama City, and Pensacola). Based upon sightings of larval damage in *O. ficus-indica* and *O. stricta*, the current westward limit of *C. cactorum* infestation is at Pensacola Beach on the west end of Santa Rosa Island in Escambia County (Hight et al. 2003).

The results suggest that the use of females sterilized with radiation will be just as effective as unirradiated (fertile) females in detecting populations of feral *C. cactorum* male moths. Reliable use of traps baited with irradiated females will allow for more widespread monitoring and surveying of areas beyond the current known limit of *C. cactorum* distribution without fear of establishing a new breeding population if the traps, once deployed, are vandalized or destroyed by people or wildlife. Based on our data for mean daily trap captures (Fig. 3) and female longevity, traps baited with irradiated females could be serviced once per week because females continued to attract males into the traps until they died. Although the number of males captured per living female was greatest on day 1, this does not necessarily mean that 1-d-old females are more attractive than older females. The male capture on day 1 could have been influenced by the presence of more males on that day. Additional trapping studies are ongoing to investigate the attractiveness of females at different ages, and the effectiveness of different trap densities, trap types, and trap heights.

The efficiency of the virgin female-baited traps relative to the absolute number of *C. cactorum* males present in a given area has not yet been determined. However, experiments to address this

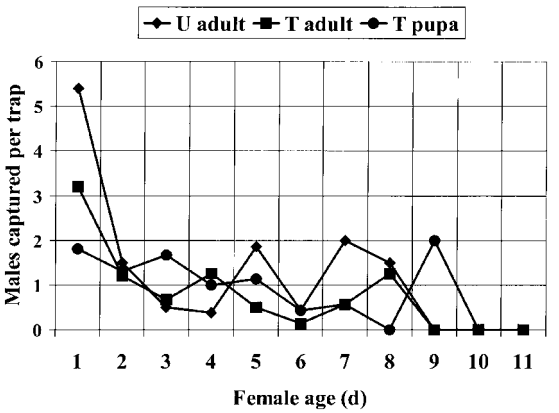

Fig. 3. Effect of female age and female treatment on the ability of *C. cactorum* females to lure male *C. cactorum* males into sticky traps (n = 10 per female type). Females were untreated (U adult), treated with a reproductively sterilizing dose of radiation (200 Gy) as pupae (T pupa), and treated with a reproductively sterilizing dose of radiation (200 Gy) as adults (T adult).

question are planned for late 2003 and 2004 in South Africa and Florida. Release-recapture studies with marked male *C. cactorum* and estimates of population density for feral *C. cactorum* will be used to determine sticky trap efficiency.

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