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Authors: Paraiso, Oulimathe, Smith, Trevor Randall, Hight, Stephen D., and Davis, Bobbie Jo

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## REARING A NATIVE CACTUS MOTH, *MELITARA PRODENTIALIS* (LEPIDOPTERA: PYRALIDAE), ON ARTIFICIAL DIET AND *OPUNTIA* CLADODES: PRELIMINARY COMPARISONS

OULIMATHE PARAISO<sup>1,\*</sup>, TREVOR RANDALL SMITH<sup>1</sup>, STEPHEN D. HIGHT<sup>2</sup> AND BOBBIE JO DAVIS<sup>1</sup>

<sup>1</sup>Methods Development & Biological Control, Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Gainesville, FL 32608, USA

<sup>2</sup>USDA-ARS, Center for Medical Agricultural and Veterinary Entomology, Tallahassee, FL 32308, USA

\*Corresponding author; E-mail: oulimathe.paraíso@freshfromflorida.com

Prickly pear cactus (*Opuntia* spp.; Caryophyllales: Cactaceae) has been cultivated worldwide because of the plants' value as an ornamental and agricultural commodity, and its ability to adapt to various climatic conditions (Hanselka & Paschal 1989). In addition to being used as fodder for livestock in dry seasons and times of drought, prickly pear cactus provides shelter and food for a wide variety of wildlife species in desert areas (Hanselka & Paschal 1989). *Opuntia* spp., which have been important cultural and agricultural plants since pre-European contact with Mexico, currently generate over 300,000 tons of fruit and vegetables, and are produced on more than 70,000 ha (Pimienta-Barrios 1990; Flores-Valdez et al. 1995; Soberón et al. 2001). *Cactoblastis cactorum* (Berg) (Lepidoptera: Pyralidae), commonly known as the Argentine cactus moth, originates from South America and feeds on most species of *Opuntia* (Mafokoane et al. 2007; Tate et al. 2009; Jezorek et al. 2010; Varone et al. 2012). Cactus moth larvae feed within cactus pads, also referred to as cladodes, which are then quickly taken over by bacterial rot and fungi, and which kill the plant (Mann 1970). Since its introduction in the U.S., *C. cactorum* has been gaining considerable attention due to its potential negative ecological and economic impacts in the southern portions of the country. This invasive moth has spread from southern Florida as far north as South Carolina and west to Louisiana over the last 20 years (Hight et al. 2002; Hight & Carpenter 2009). In 2006 and 2007, *C. cactorum* was detected in Mexico but was eradicated by using a combination of control techniques including host plant removal/sanitation and the sterile insect technique (SIT) (Carpenter et al. 2008; NAPPO 2009). The pathway of moth introduction into Mexico is still unclear; however, considering the close proximity of populations of this pest in the Caribbean, there is a high probability that it will again find its way into this country.

A classical biological control program against *C. cactorum* was recently initiated in the U.S. In 2013, *Apanteles opuntiarum* Bertha & Mar-

tínez (Hymenoptera: Braconidae), a co-evolved natural enemy of *C. cactorum*, was imported from Argentina into a quarantine facility of the Florida Dept. of Agric. and Consumer Serv., Div. of Plant Ind. in Gainesville, Florida. Field studies in Argentina identified this gregarious koinobiont larval parasitoid as being potentially highly host specific (Martínez et al. 2012). Host specificity experiments of this parasitoid on native cactus-feeding moths from the southeastern and southwestern U.S. are currently taking place in the Florida quarantine facility.

North America has 10 species of native *Opuntia* feeding pyralids represented by 2 genera (*Melitara* and *Ozamia*) (Neunzig 1997). Seven species of *Melitara* are known in the U.S. including *M. prodentialis* Walker in the eastern states, *M. dentata* (Grote) in the western states, and *M. doddalis* Dyar in the southwestern states (Habeck & Bennett 1990; Neunzig 1997; Simonsen et al. 2009). *Melitara prodentialis* often share the same feeding niches (even the same cladode) as the non-native *C. cactorum* (Paraiso et al. 2011, 2013). In field conditions, feeding impact on *Opuntia* cacti by native cactus moths differs greatly from that of non-native cactus moths (Baker & Stiling 2009). In laboratory conditions, rearing native cactus moths on fresh cactus cladodes is difficult because *Melitara* larvae have difficulty surviving in healthy prickly pear cladodes (Carlton & Kring 1994). Native larvae are more fastidious than their non-native counterparts and can be easily overwhelmed with mucilage produced by host plants (pers. obsv.). *Melitara* colonies are prone to pathogen contamination. Availability of fresh pads and larval stress caused when transferring individuals from old pads to fresh pads are additional limiting factors. High mortality rates have been attributed to larvae drowning in rotting pads or their inability to feed on fresh cladodes (pers. obsv.). The use of artificial diet to improve testing methodologies for native cactus moths may be a feasible alternative to rearing the insects on *Opuntia* cladodes. Artificial diets for rearing native cactus moths have been previously tested without suc-

cess (Carlton & Kring 1994). However, a successful artificial diet was developed and used for mass rearing *C. cactorum* for a SIT program in the southeastern U.S. (Marti & Carpenter 2008; Marti et al. 2008; Carpenter & Hight 2012). Healthy and uniformly developed hosts are necessary to conduct host specificity testing with *A. opuntarium*. In the present study, the *C. cactorum* artificial diet was tested for mass rearing *Melitara* spp. Comparisons were made for several biological parameters of the native cactus moth, *M. prodenialis*, reared on artificial versus natural cladode diet.

Eggsticks of *M. prodenialis* were obtained from field collections around Jacksonville, Florida and along coastal Mississippi. The moths were reared either on artificial diet or on *Opuntia* cladodes following procedures of Marti & Carpenter (2008) (Fig. 1). The artificial diet was comprised of ground white pinto beans, sugar, Brewer's yeast, vitamins and minerals (Carpenter & Hight 2012). The artificial diet was cut into 5 × 6.4 cm blocks and then covered with thin wax to prevent early desiccation and to simulate the cuticle of cactus cladodes (Marti & Carpenter 2008; Marti et al. 2008). *Opuntia* spp. cladodes were obtained from native stands and cactus plantations in northern Florida. Only mature (at least 2 yr old), non-woody, dark green cladodes were selected from various *Opuntia* plants [*O. ficus-indica* (L.) Miller, *O. stricta* Hawarth]. *Melitara* eggsticks were set up in 20 containers (replications) on the artificial diet and 10 containers on the natural diet. An average of 22.3 eggs on artificial diet and 23.6 eggs on *Opuntia* cladodes were used for rearing purposes. Rearing containers were incubated in growth chambers (T 28–29° C, RH 60 ± 5%, 14:10 h L:D) until larval development was complete. Each container was checked every 2 days to remove frass produced by larval feeding and provide fresh food in excess as needed. The following biological parameters were recorded: hatching rate (number of hatched eggsticks per day), pupation rate (number of collected pupae per day), adult emergence rate (number of emerged adult per day), larval development time, pupal development time and adult longevity. Estimates of central tendencies were reported as mean ± standard error of mean. Significance among means was analyzed using a two-sample t test ( $P = 0.05$ ). Female ratios and pupal deformities were analyzed as proportions. The Statistix Statistical Software Version 8.0 (Tallahassee, FL) was used to perform the statistical analyses.

Larvae completed their development on the artificial diet suggesting that the diet provided a sufficient level of nutrition for development of the moths under laboratory conditions (Fig. 2). Comparisons between biological parameters

measured for *M. prodenialis* reared on artificial diet verses those reared on *Opuntia* cladodes are presented in Table 1. Larvae reared on artificial diet resulted in 52% of the larvae completing their pupation while on natural diet 46% of the larvae completed their pupation. The pupation rate and emergence rate were higher from larvae reared on artificial diet as opposed to natural diet. Egg development time and both female and male pupal lengths were not significantly different between natural and artificial diets. Larval and pupal duration on *Opuntia* cladodes were both significantly longer than on artificial diet; however, adult longevity was not affected by the type of diet. A significantly lower number of larvae completed their life cycle when reared on *Opuntia* cladodes as opposed to artificial diet. Larvae developing on cladodes took twice as long to reach the pupal stage as larvae on the diet. Consequently, total development period was approximately 98 days on *Opuntia* cladodes versus 65 days on artificial diet. This represented a significant reduction in time and labor when rearing these moths on artificial diet. Larvae reared on cladodes frequently needed to be physically transferred from old, rotten or dried up cladodes to fresh ones. Otherwise, larvae would crawl out of old cladodes and die from starvation on the side of containers, failing to penetrate new cladodes. In contrast, very few larvae reared on artificial diet died from starvation. Larvae found cracks in the wax surrounding the diet blocks and resumed feeding on new diet blocks without assistance. However, a higher male ratio and an increase in the percentage of deformed larviform pupae were recorded on the artificial diet.

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## SUMMARY

This study compared several biological parameters of native cactus moth, *Melitara prodenialis*, reared on an artificial versus the natural diet of *Opuntia* spp. cladodes. Results suggest that the current artificial diet developed for mass rearing *C. cactorum* can provide nutritional value for the rearing of *Melitara* spp. native cactus moths. Overall, rearing *M. prodenialis* on the artificial diet was more successful than on *Opuntia* cladodes and required less time and labor. Mass rearing of *M. prodenialis* using artificial diet should improve once sub-

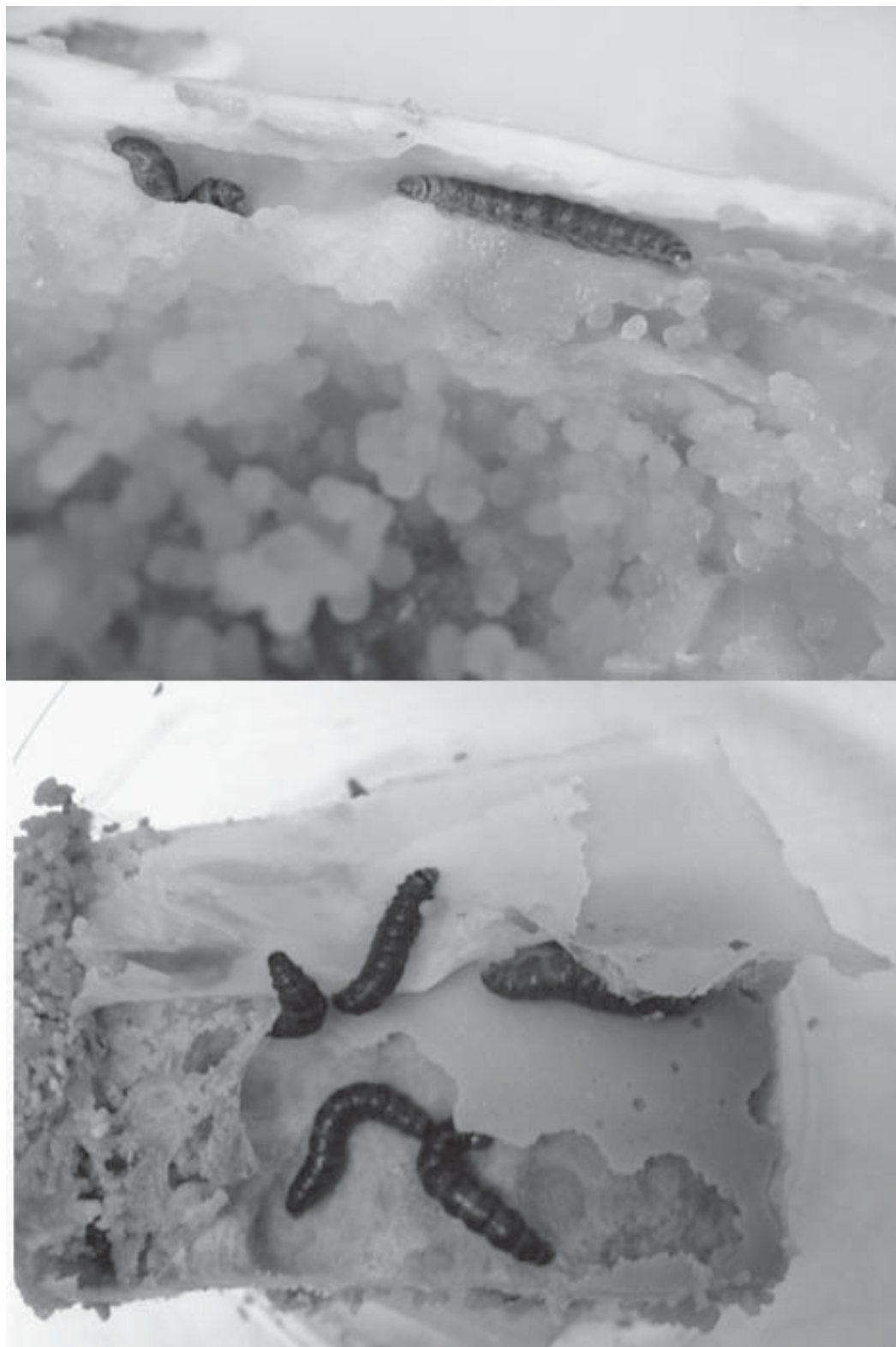


Fig. 1. Late instar larvae of a native cactus moth, *Melitara prodenialis*, feeding on A) an *Opuntia* cladode and B) an artificial wax-covered diet cake.



Fig. 2. Pre-pupae and pupae of *Melitara prodenialis* reared on artificial diet.

sequent generations become adapted to laboratory rearing conditions.

Key Words: *Apanteles opuntiarum*, *Cactoblastis cactorum*, cladodes, developmental abnormalities, mass rearing, nutrient deficiencies, *Opuntia*

RESUMEN

Este estudio comparó varios parámetros biológicos de la palomilla nativa del cactus (nopal), *Melitara prodenialis*, criadas sobre una dieta artificial versus una dieta natural de cladodios de *Opuntia* spp. Los resultados sugieren que la

dieta artificial actual desarrollada para la cría masiva de *Cactoblastis cactorum* puede proveer valor nutricional para la cría de las polillas *Melitara* spp. nativas del cactus. En general, la cría de *M. prodenialis* sobre la dieta artificial tuvo más éxito que las criadas sobre cladodios de *Opuntia* y requirió menos tiempo y trabajo. La cría masiva de *M. prodenialis* usando dieta artificial debería mejorar una vez que las generaciones subsiguientes se adapten a las condiciones de cría en el laboratorio.

Palabras clave: *Apanteles opuntiarum*, *Cactoblastis cactorum*, cladodios, anomalías del desarrollo, cría en masa, deficiencias de nutrientes, *Opuntia*

TABLE 1. COMPARISON OF BIOLOGICAL PARAMETERS OF *MELITARA PRODENIALIS* REARED ON ARTIFICIAL DIET AND ON *OPUNTIA* CLADODES.

Parameters	Mean ± S.E.	
	Type of Diet	
	Artificial	<i>Opuntia</i> cladodes
Hatching rate (# hatched eggstick/day)	2.1 ± 1.45 A	2.5 ± 2.17 B
Pupation rate (#pupae/day)	5.65 ± 1.79 A	1.00 ± 0.49 B
Emergence rate (#adult/day)	3.47 ± 1.00 A	1.22 ± 0.64 B
Egg stage duration (days)	13.00 ± 0.43 A	12.21 ± 0.87 A
Larval stage duration (days)	35.94 ± 1.03 A	71.62 ± 2.77 B
Pupal stage duration (days)	15.26 ± 1.68 A	19.80 ± 1.31 B
Total development duration (days)	64.78 ± 1.05 A	98.42 ± 1.64 B
Adult longevity (days)	6.00 ± 0.15 A	6.80 ± 0.84 A
Female ratio	0.48 ± 0.05 A	0.63 ± 0.18 B
Female pupal length (mm)	13.89 ± 0.29 A	14.87 ± 0.62 A
Male pupal length (mm)	13.90 ± 0.32 A	12.94 ± 0.33 A
Pupal length (mm)	13.80 ± 0.32 A	14.14 ± 0.85 A
Deformed pupae proportion	0.41 ± 0.10 A	0.14 ± 0.03 B

Means within a row followed by the same letter are not statistically different with two-sample t test (*P* < 0.05).



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