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Authors: Andress, Earl, War, Mamadou, and Shelly, Todd

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EFFECT OF PRE-RELEASE STORAGE ON THE FLIGHT ABILITY OF STERILE MEDITERRANEAN FRUIT FLIES (DIPTERA: TEPHRITIDAE)

EARL ANDRESS[1], MAMADOU WAR[2] AND TODD SHELLY[3,*]
[1]USDA-APHIS, 3802 Constitution Avenue, Los Alamitos, CA 90720, USA

[2]CDFA, 3802 Constitution Avenue, Los Alamitos, CA 90720, USA

[3]USDA-APHIS, 41-650 Ahiki Street, Waimanalo, HI 96795, USA

*Corresponding author; E-mail: todd.e.shelly@aphis.usda.gov

The California Department of Food and Agriculture and the US Department of Agriculture operate a preventative release program (PRP) in which sterile adult males of the Mediterranean Fruit Fly, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae), are released by aircraft over a large area of southern California. Sterile fly pupae are received from 2 mass-rearing facilities, Guatemala (GT) and Hawaii (HW), respectively, which produce the same strain (*tsl* Vienna-7). After emergence, the adults are held at a facility in Los Alamitos, California. When 2 d old, the sterile flies are "knocked down" by chilling (at 4 °C) to allow their transfer to and storage within the aircraft used for the releases. The interval between the start of chilling and release is typically 2-3 h.

Andress et al. (2012) measured the flight ability (following the standard protocol, FAO/IAEA/USDA 2003) of sterile GT and HW males after chilling. As chill time increased, flight ability decreased at the same rate for GT and HW flies. However, for any given chill regimen, the HW flies displayed greater flight ability than the GT flies. Standard quality control tests detected no significant difference between GT and HW flies in their post-shipping (but pre-chilling) flight ability, indicating that potential differences in shipping did not account for the difference observed in post-chilling flight. While these results suggest a differential effect of chilling on HW and GT flies, that earlier study did not assess the effects of holding per se (i.e., independent of chilling) on flight performance. Specifically, flight ability was not measured after confinement in the emergence towers but before chilling, making it impossible to distinguish potential effects of holding conditions (food availability, fly density, etc.) versus chilling per se. Thus, the lower flight performance recorded for GT flies may have originated, not from the chilling, but from holding conditions that had a greater negative impact on GT flies than HW flies.

To address this shortcoming, the present study supplies data on flight ability for HW and GT males i) upon emergence (post-shipping but without adult storage and subsequent chilling; lowest stress), ii) held in 'relaxed' conditions of low density and high food but chilled for the standard period of 2 h, iii) held in towers but not subject to long-term chilling, or iv) after the routine procedure of storage in the towers and pre-release knockdown (highest stress). All 4 sets of measurements were made on GT and HW flies that arrived in Los Alamitos on the same day for 7 different days in August, 2012. Flies from the 2 facilities were subject to equal durations of hypoxia before irradiation (2-4 h) and during shipment (24-25 h).

Shipping, handling, and rearing procedures followed Andress et al. (2012). Flies used in the 4 treatments noted above were collected as follows for individual daily shipments.

Post-shipping, no storage, no chilling (NS-NC). Flight tests were conducted on 5 groups of 100 pupae for GT and HW flies, respectively, as part of routine quality control. As this standard estimate is a composite measure that includes emergence success, we computed adjusted flight abilities, where the percentage of fliers was based only on normal, fully emerged adult flies.

'Relaxed' storage, routine chilling (R-C). Thirty mL of pupae (≈ 1,600 individuals) were placed in each of 4 plexiglass cages (30 × 30 × 40 cm; 2 with GT flies, 2 with HW flies) with abundant food (a slab of sugar agar gel, 7.5 × 9 × 1 cm). When adults were 2 d old, the cages were placed in a 4 °C room for 2 h. Three samples of 100 chilled males were then taken from each cage (yielding 6 samples for GT and HW flies, respectively). Flight tests for this treatment, as well as the following 2 treatments, were conducted immediately after chilling.

Tower storage, no long-term chilling (T-NC). Pupae were placed in emergence towers as part of routine PRP procedures. Each tower held 46-52 trays, each of which was loaded with 350 mL of GT or HW pupae (18,000-19,000 pupae) and a sugar agar slab (15 × 9 × 1 cm). Immediately before the emergence towers were moved to the chilling room, we aspirated 500-1,000 adults from each of 6 trays (at different heights from a single tower for GT or HW flies, respectively) through a hole drilled in 1 side of each tray. These flies were chilled just long enough (< 15 min) to remove 100 individuals from each tray collection.

Tower storage, routine chilling (T-C). As a regular component of PRP quality control, flight tests were conducted using flies from 3 towers holding GT and HW flies, respectively, that had been chilled for 2 h. For both sources, flight tests were conducted on 5 samples (each of 100 males) that included similar numbers of males taken from the different towers.

Andress et al. (2012) provide details regarding the protocol followed for the flight ability tests. For all treatments, we computed the proportion of fliers for each test day as the grand mean of all samples taken on a given test day. These daily values were used in a 2-way ANOVA with source (GT or HW) and treatment (the 4 listed above) as main factors. Arcsine was used to transform all percentage values.

Both source ($F_{1, 48} = 4.8, P = 0.03$) and treatment ($F_{3, 48} = 133.8, P < 0.001$) had significant effects on flight ability. However, the interaction term was also significant ($F_{3,48} = 9.1, P < 0.001$), indicating the effect of source varied significantly with the treatment considered. Accordingly, the Tukey multiple comparisons test was used to identify pair wise differences separately within each of the levels of the main effects (Fig. 1). For GT flies, flight ability differed significantly between each of the conditions. For HW flies, flight ability differed significantly between all treatments, except no difference was detected between R-C and T-NC conditions. Comparing the 2 sources within each treatment revealed that there were no significant flight differences between GT and HW flies for either the NS-NC or the R-C treatments. However, HW flies had significantly higher flight ability than GT flies for both the T-NC and the T-C treatments.

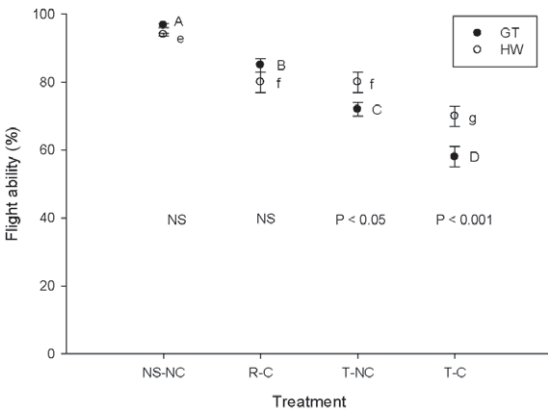

Fig. 1. Flight ability of sterile males produced at the Guatemala (GT) or Hawaii (HW) mass-rearing facilities subject to 4 different treatments: NS-NC: no storage, no chilling (flight measured on newly emerged adults); R-C: relaxed storage, chilling; T-NC: tower storage, no chilling; T-C: tower storage, chilling. Values represent means (± 1 SE) of daily measurements (N = 7 d in all cases). Means sharing the same (upper case, GT; lower case, HW) letter were not significantly different. For each treatment, the results of GT-HW comparisons are given below the corresponding points (NS – no significant difference).

We conclude i) the combination of tower storage plus chilling (T-C) depressed flight ability more than tower storage without prolonged chilling (T-NC) or relaxed storage with chilling (R-C) for both GT and HW flies, and ii) tower storage had a greater negative effect on GT than HW flies as evidenced by the significantly greater flight ability of GT flies held in relaxed conditions but subjected to chilling (R-C) compared to GT flies held in towers but not chilled for 2 h (T-NC). In contrast, there was no flight ability difference between groups of HW flies that were held for comparison in these same 2 conditions.

Reasons underlying the greater adverse effects of tower storage on the GT flies are unknown, but there are 2 likely possibilities. (i) Given the greater emergence rate and smaller size of GT flies, more GT flies were held per tray relative to HW flies, resulting in higher crowding for the HW adults. Based on emergence values and volumetric conversions to pupal counts, we estimate that, on average, each tower tray held 17,200 GT adults compared to 15,800 HW adults, a difference of 9%. While a relatively small difference, it nonetheless could have contributed to the lower flight ability of GT males held in the towers. (ii) Of greater importance, perhaps, GT pupae are irradiated and shipped at a later age (i.e., closer to emergence) than HW pupae (1.5 – 1.0 versus 2 days pre-emergence, respectively), and correspondingly complete emergence of GT flies occurs 12-24 h before that of HW flies. Thus, GT males were subject to tower conditions for longer time intervals than HW flies, which may have resulted in their lower flight performance.

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Summary

Although Mediterranean fruit flies (*Ceratitis capitata* (Wiedemann); Diptera: Tephritidae) of the same *tsl* Vienna-7 genetic sexing strain are mass-reared in Guatemala (GT medflies) and Hawaii (HW medflies), the GT flies appeared to suffered greater adverse effects (in terms of flight ability) from crowding in the emergence towers than did the HW flies. Possible reasons for this difference involve source-related differences in fly size and shipping time (relative to pupal age) and suggest that managers adopt flexible procedures in handling flies from different sources and with differing biological traits.

Key Words: chilling, emergence tower, pre-release knockdown, relaxed condition, tower storage

RESUMEN

Aunque las moscas Mediterráneo de la fruta (*Ceratitis capitata* (Wiedemann), Diptera: Tephritidae), criadas en masa en Guatemala (GT mosca de la fruta) y Hawaii (HW mosca de la fruta), son de una cepa genética sexual Viena-7 *tsl* idéntica, las moscas GT parecían sufrir más adverso efectos (en términos de capacidad de vuelo) de amontonamiento en las torres de emergencia comparadas con las moscas HW. Las posibles razones para esta diferencia implican diferencias en el tamaño de las moscas y el tiempo de envío relacionados con el fuente de las moscas (en relación a la edad de pupa) y sugieren que los gerentes adopten procedimientos flexibles en el manejo de las moscas de diferentes fuentes y con características biológicas diferentes.

Palabras Clave: enfriamiento, torre de emergencia, tumbado pre-liberación, condición relajada, almacenamiento en torres

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