THE UNIVERSITY OF ARIZONA
COLLEGE OF AGRICULTURE AND LIFE SCIENCES
COOPERATIVE EXTENSION
Arizona Pest Management Center

2016 Decon7 Commercial Scale Efficacy Trials

University of Arizona Maricopa Agricultural Center
Peter C. Ellsworth, Ph.D., IPM Specialist; Alexa Brown, Project Puente Summer Intern & Research Laboratory Aide; and Isadora Bordini, International Scholar & Assistant in Extension
15 December 2016

Introduction

The global transport and commerce of products present special challenges when infestations of insects and their relatives are detected. Expensive quarantine procedures are costly in time and materials, and currently depend on a greenhouse gas biocidal fumigant, methyl bromide, which is undergoing phase-out from most uses worldwide. In addition to these obvious biosafety concerns, the handling of methyl bromide requires special training, permits and licenses to safeguard human health and the environment. Cost-effective and safe alternatives are needed. Decon7 (D7) is a non-corrosive, non-toxic, multicomponent decontaminating product developed to address biological and chemical security threats. There exist data to support the log-7 reduction in microbial contamination from bacteria, molds, viruses, etc. as well as controlled, laboratory studies demonstrating the complete control of macro-invertebrates including 9 species from 8 families and 5 orders of insects. The studies reported here were designed to examine the biocidal impacts of D7 on a model macro-invertebrate in a commercial-scale proof-of-concept shipping container system. We specifically examined application method and minimum interval to achieve maximum kill rates.

Methods

A commercial-scale proof-of-concept model system was established with ProBox shipping containers, each a 10 X 10 X 10 ft sealed, steel box with wooden floors (Figure 1). A series of studies were designed with our model macro-invertebrate species, *Zophobas morio* larvae (a.k.a. "superworm"; Coleoptera: Tenebrionidae). As a very large insect (50–60 mm) with thick integument, this species has served as an excellent indicator of broad efficacy across many macro-invertebrates. In 3 different studies, 12 ProBoxes were infested with 100–200 large larvae, randomly assigned to treatments inclusive of an untreated control (UTC), and arranged into 3–4 replicates in a series of separate experiments: *Experiment 1* tested 24-h exposures to foaming or fogging application methods (Figure 2); *Experiment 2* tested 2–8 h exposures to foaming application; and *Experiment 3* carefully controlled pest harborages and tested 2.5–3.5 h exposures to two dilutions / equipment for foaming and full strength fogging. Decon7 was used at full strength except in Exp. 1 and 3, where foam was applied in a 25:1 dilution using a AR Blue Clean Pressure washer; in Exp. 2 and 3, foaming was with a commercial unit operating at 65–85 psi. The commercial containers were empty except for plastic container harborages stacked in the middle of the box in Exp. 1; however, food (ca. 500 ml of bran meal) was provided on a portion of the floors in all boxes up to 1 cm in thickness in Exp. 1 & 2 (Figure 3). In Exp. 3, the bran meal food source was raked out to minimize any chance of harborage within the media (Figure 4). Metal doors were constructed with rubber gasket door sweeps that sealed the doors to the floor, but provided a narrow harborage that was less accessible to directed sprays or fog (Figure 5). At the end of each testing period, all larvae were recovered and classified as dead, "sick", or alive. A small number were eaten (cannibalized) partially in Exp. 1 & 2 and were eliminated from the data. Apparent and corrected mortalities (using Henderson-Tilton's formula) were calculated and analyzed.

Results

After 24 h, *Experiment 1* showed excellent efficacy of the 25:1 diluted foaming treatment, which was significantly better than the fogging treatment. UTC mortalities were low, averaging 7%; a small number of "sick" larvae were recovered from most boxes. However, there was survival in the D7

treatments (Table 1). In *Experiment 2*, very high mortalities were found in each D7 foamed box and UTC mortality after 24 h was low. Nearly all surviving larvae were recovered within a narrow layer of bran feed harborage present and available in Exp. 1 & 2 (Figure 3). The very few live larvae recovered outside the bran matrix may have exited after D7 potency was reduced over time. Very high mortalities were found for each exposure interval, 2, 4, 6, and 8 h, with no apparent trend with duration of exposure. This suggests that most of the mortality was occurring in less than 2 h.

Experiment 3 was carefully adjusted to minimize any chance of pest harborage within the food medium (Figure 4). The results, when all larvae were directly exposed to D7, were dramatically improved (Figure 7). The foaming applications, either full strength or lower, diluted concentration, were far superior to the full strength fogging under the conditions of this trial. Virtually complete mortality was observed by 2.5 h after treatment, suggesting again that direct contact exposures of D7 foam work very quickly. Foam has the advantage of coating all surfaces and seeping into inaccessible areas and potential harborages (Figure 7).

Table 1. Commercial scale summaries for each of 3 experiments (No.) bioassaying Decon7 against *Zophobas morio* larvae. Observations were made at 24 hours after treatment (HAT) unless otherwise noted. A total of 5480 insects were assayed.

No.	Treatment	Concentration	HAT	Reps	N, Alive	N, Total	Mortality (%)	Corrected Mortality (%)	
1	Low Foam	Diluted 25:1	24	4	72	776	90.5 ±5.7%	89.1 ±6.9%	A
1	Fog	Full strength	24	4	354	720	50.1 ±12.6%	46.5 ±12.5%	B
1	UTC	–	24	4	655	706	7.2 ±3.2%	0 ±0%	C
2	Foam	Full strength	2	2	23	347	93.2 ±2.9%	92.7 ±2.7%	AB
2	Foam	Full strength	4	2	21	359	94.1 ±0.4%	93.1 ±0.6%	AB
2	Foam	Full strength	6	2	46	356	87.1 ±2.9%	86 ±2.4%	B
2	Foam	Full strength	8	3	21	534	96.1 ±0.6%	95.5 ±0.7%	A
2	UTC	–	24	3	435	495	11.5 ±4.1%	0 ±0%	C
3	Foam	Full strength	2.5	4	1	403	99.8 ±0.2%	99.8 ±0.2%	A
3	Low Foam	Diluted 25:1	2.5	3	0	299	100 ±0%	100 ±0%	A
3	Fog	Full strength	3.5	2	154	192	19.7 ±5.8%	19.7 ±5.8%	B
3	UTC	–	6	3	293	293	0 ±0%	0 ±0%	C

Corrected mortality based on Henderson-Tilton's formula, Henderson & Tilton, 1955

HAT, Hours after treatment for assessment

Mean corrected mortalities not sharing a letter within an experiment are significantly different from each other by Tukey's HSD (P<0.05)

Conclusions

This series of experiments confirmed the high rates of rapid insect mortality possible with Decon7 foaming applications. When conditions are optimized (no pest harborages) and insects are contacted by Decon7, 100% mortality is possible within 2.5 h of foaming at even diluted application rates. We also conclude that fogging, even at full strength and held for 24 h, is likely not effective enough against large bodied insects to achieve 100% mortality. Fogging is likely much more effective against smaller bodied, flying insects, out of reach of other application methods. The results of these studies underscore the importance of basic sanitation within containers (e.g., removal of soil or other organic debris that could serve as harborages) to achieve complete decontamination with Decon7 foam, making containers free of all arthropod pests. The lower pressure, more diluted Decon7 foaming application delivered more liquid volume with more opportunity for Decon7 to reach its intended targets. Decon7 foam provides users with an excellent, safer and more efficient alternative to more hazardous methyl bromide applications for container arthropod decontamination.

Figure 1. Commercial storage containers used in these studies, each a 10 ft cube with steel walls & wooden floors.

Figure 2. *Zophobas morio* larvae (a.k.a. "superworm") in commercial containers of Experiment 1 at the time of infestation. Note the variable thickness of food medium on floor and trails through the nearly 1 cm layer of bran.

Figure 3. Condition of food medium at the conclusion of Experiment 1. Note crusting of top layer where D7 was absorbed by the bran. Two surviving larvae are visible in the bran harborage.

Figure 4. Condition of food medium at the conclusion of Experiment 3. Note the bran was raked out to minimize potential for pest harborage. All food was wetted by the D7 foaming applications. All larvae contacted were killed; note dead superworms shown, left and right.

Figure 5. Note rubber gasket in door jam between steel door and steel floor. Superworms would tightly appress themselves against this rubber seal and avoid exposure to the fogging application. Many survivors were recovered from this area.

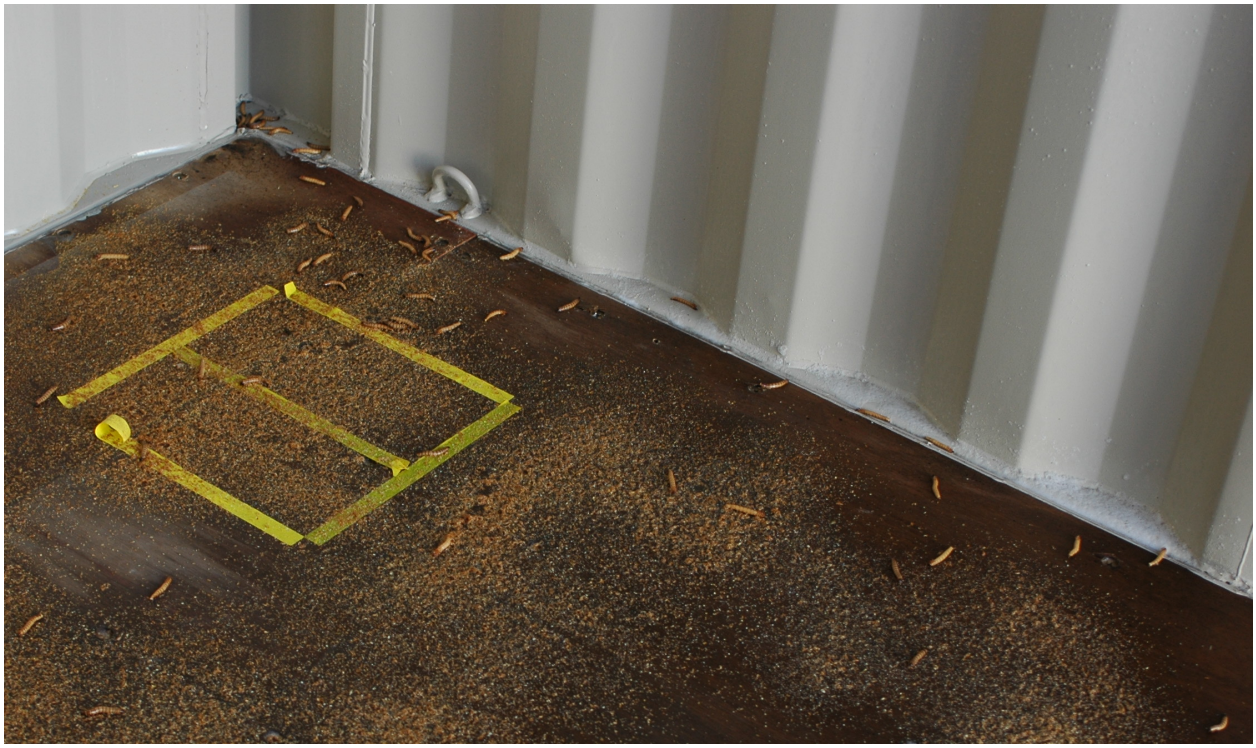

Figure 6. Dead bodies of superworms scattered over the floor at 2.5 h after treatment with diluted foaming of D7. Note wetted bran food media across the floor. No larvae survived this application.

Figure 7. Foaming at full or diluted concentrations had the advantage of coating all surfaces and seeping into potential pest harborages like under the door jam, rubber gasket. Note the coat of foam still visible in Experiment 3 2.5 h after foaming in the door threshold (right).

References

Henderson, C.F. and E. W. Tilton, 1955. Tests with acaricides against the brown wheat mite, J. Econ. Entomol. 48:157-161.