WTFRC Project # _____ Organization Project # _58-5352-4-445

Project title: Confirmatory Tests of an Organic Quarantine Treatment Against Codling Moth in Apples

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Objectives:

- 1. Treat 30,000 codling moth 4th instars in apples with zero survivors using the CATTS quarantine treatment.
- 2. Submit these data to USDA-ARS and USDA-APHIS for proposal of this treatment to our trading partners currently requiring methyl bromide treatment of apples for import.

Significant findings: We began the first treatments at the commercial CATTS unit in George on November 30, 2004. We performed 5 treatments at the commercial CATTS unit. We found that the transport of tarp covered fruit in a ½ ton truck caused fruit temperatures to drop to approximately 5°C, this caused a significant delay in acclimating the fruit to 23°C prior to the application of CATTS. We subsequently rented an enclosed 14 ft. moving van for the remainder of the treatments.

Cold winter temperatures caused the commercial CATTS unit not to heat as rapidly as compared to treatments performed in the previous spring and summer. There was also a programming problem which caused the continuous injection of nitrogen or carbon dioxide throughout the duration of the treatment, causing heat to be lost from the unit. There was also a problem with using liquid carbon dioxide gas tanks in cold weather, even with the use of hot water baths or heating tape. The production of CO_2 gas was often not sufficient to maintain atmosphere targets. There was also a problem with the sample chamber air not being properly dried prior to being injected over the O_2 and CO_2 analyzers. This caused an atmosphere mis-read in at least 1 treatment. We have since corrected all the program and equipment problems in the commercial CATTS chamber. Out of the 5 treatments conducted, we were able to perform 2 acceptable treatments in which all the larvae in the fruit were killed. These resulted in a total of 9,539 larvae killed in the commercial CATTS.

After the December 28th treatment, we decided to use the laboratory CATTS unit for conducting the confirmatory tests. In discussions with members of USDA-APHIS, they thought that this was an acceptable approach and that the data would be acceptable to MAFF-Japan. As you will recall, methyl bromide fumigations are performed in the laboratory in a ½ ton unit. A total of 20 treatments were performed on a total of 31,331 larvae with a result of 100% mortality. Control mortality was 1.5% over all the tests. So, accounting for control mortality, a total of 30,861 larvae were controlled by the CATTS treatment. A total of 41,145 larvae were used in the test and controls combined.

We found that the laboratory CATTS unit gave consistent and reproducible treatments. We believe that this type of treatment reproducibility is necessary for future acceptance of this treatment by foreign countries. We are able to obtain this type of treatment reproducibility in the spring and summer in the commercial CATTS.

Methods:

Commercial CATTS: Insects were reared to the desired stage and sufficient numbers. Insects were removed from the artificial diet medium, placed on fruit to allow for entry into the fruit. We used a ratio of 200 larvae per 15 lbs of fruit. It took 8 people 10 hours to infest 18 pans worth of insects. We obtained approximately 300 4th instar codling moth from 1 pan of diet. So, including controls, we infested an average of 5,277 larvae in a day. One day following infesting, any insects not inside the fruit were counted and disposed, since only insects actually inside the fruit at time of treatment are included in the total. We averaged 4,285 larvae in the fruit per treatment. We averaged about 81% entry of insects into the fruit. The infested fruit was transported to the treatment facility (PacOrganic in George, WA). Due to cold weather, we had to transport the fruit in a rented moving van to maintain fruit temperatures above freezing. The infested fruit was then placed into the treatment bins and temperature probes were set inside the fruit and the treatment applied. The treatment consisted of a heating rate of 12°C/hr, to a final chamber temperature of 46°C with a soak time of 1 hr, under a 1% oxygen, 15% carbon dioxide environment. Core low temperatures must remain at 45°C for 15 min. Following treatment, the fruit was transported back to YARL for evaluation. The bin was placed into a 2°C cold room after reaching YARL to prevent accidental re-infestation from laboratory insects. The infested fruit was cut open and searched for live and dead insects. It took 8 people 10 hours to cut the treated fruit. Untreated, control infested fruit was also evaluated.

<u>Laboratory CATTS</u>: Infesting and evaluation of the fruit was the same as for the commercial CATTS treatments with the exception that infesting took place over two consecutive days. We infested 15# of fruit with 200 4th instar larvae and used 100 larvae in 7.5# of fruit for untreated controls for each day. We infested 16 trays of 15# of fruit on Mondays and 8 trays of 15# on Tuesdays. Two trays of infested fruit were transferred into one CATTS treatment lugs. A total of 4 lugs were placed into the laboratory CATTS unit. Two treatments were conducted on Tuesdays and one treatment on Wednesdays. Following CATTS treatment, lugs of fruit were stored at 2°C to prevent accidental infestation by laboratory insects. Treated fruit were evaluated the next day as previously described.

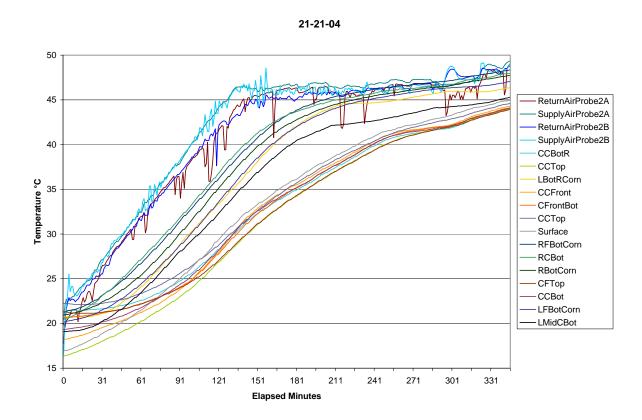
Results and discussion:

We were able to demonstrate that the commercial CATTS unit can perform effective quarantine treatments against codling moth in apples, providing all systems are operating properly.

We performed 5 treatments in the commercial CATTS unit in George. Only 2 resulted in 100% mortality of the 4th instars. These two treatments controlled a total of 9,539 larvae in organic apples.

Dan Black of Techni-Systems has already designed improvements for the commercial chamber to improve air flow, prevent temperature overshoots, and protect against inclement weather. He is also in negotiations with a company in the Neatherlands, ECO2, for large-scale production of CATTS units. This company intends to own the units they construct and rent the units out to the industry, but will also offer units for purchase.

Figure 1. Chamber air and fruit temperatures during CATTS treatment in commercial unit on December 21, 2004..

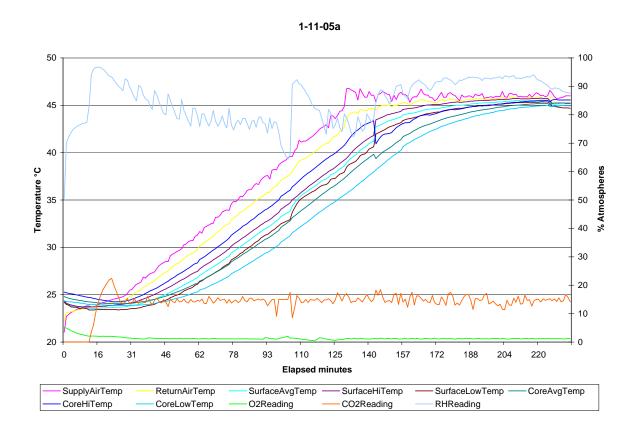


We were able to kill over $30,000 \ 4^{th}$ instar codling moth in organic apples with zero survivors using the laboratory scale CATTS unit. Treatment reproducibility and efficacy were well within acceptable parameters.

Table 2. Summary of control and treatment larval mortalities.

							Corrected
							Number
	# Infested	# Out	# Treated	# Live	# Dead	% Mort	Treated
Control	1345	324	0	1005	16	1.5	_
Treated	39800	8469	31331	0	31331	100	30861

Figure 2. Treatment conditions in laboratory CATTS during the first run on January 11, 2005. Size 100 organic Golden Delicious apples were used. O_2 , CO_2 and relative humidity (RH) values are indicated by the % Atmospheres axis. Air and fruit temperatures are indicated by the Temperature °C axis.



Budget: We obtained matching funds from the Washington Commission on Pesticide Registration. Matching funds promised from USDA-APHIS were not able to be transferred prior to the end of the fiscal year.

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PI: Lisa G. Neven, Ph.D. **Project duration**: 2004-2005

Current year: 2005

Project total (1 year): \$15,090

Year	Year 1 (2004)
Total	\$15,090
Item	Year 1 (2004)
Salaries ¹	7,000
Benefits ² (%)	2,100
Wages	
Benefits (%)	
Equipment	
Supplies ³	5,590
Travel 4	400
Miscellaneous	
Total ⁵	15,090

¹Salaries for 3 GS-3 technicians for 1 month.

Actual cost expenditures for this project are available upon request.

²Benefits are 30% of salary.

³Supplies include diet ingredients for rearing insects, disposable pans for rearing insects, nylon insect containment material, carbon dioxide gas, nitrogen gas.

⁴Travel back and forth to PacOrganic from USDA-ARS in Wapato.

⁵We obtained matching funds from Washington Commission on Pesticide Registration.