Controlled Atmosphere/High Temperature Forced Air: A Non-Chemical Quarantine Treatment for Stone Fruit

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This work is part of a continuing project designed to determine if controlled atmospheres combined with high temperature forced air (CATTS) can be used as a quarantine treatment for peaches and nectarines. An important part of this season's work was to initiate experiments using a semi-commercial scale CATTS chamber, and applying the treatment to boxed/palletized fruit, to begin evaluating whether this treatment is feasible on a commercial scale. Entomological work included research into two different technologies (controlled atmosphere (CA) water bath and CA heating block) designed to speed the development of CATTS treatments for new insect pests and the initiation of testing of the effectiveness of CATTS against San Jose scale (SJS), European red mite (ERM) and spider mites (SM).

METHODS AND MATERIALS

Semi-commercial CATTS chamber installation

The new semi-commercial scale CATTS chamber arrived on July 9 and was installed by the manufacturer (Techni-Systems, Chelan, WA) over a period of 11 days. This included assembly and testing of heating and controlled atmosphere capabilities. The chamber is capable of simultaneously treating two full pallets of boxed fruit.

Semi-commercial CATTS chamber testing

Commercially-packed fruit were utilized due to the great difficulty that there would be in obtaining sized fruit from culls. Initial fruit temperatures were adjusted to 23 °C by placing packed fruit which had not been cooled into an environmental room set at 23 °C or higher overnight prior to the run. Boxes were removed from the pallet stack to insert temperature probes into the fruit from boxes throughout the stack. Fruit were stacked into the chamber in standard configuration, with 80 boxes per each of the two pallets. The standard run conditions were the same as for prior treatments using the laboratory chamber. The initial chamber temperature (23 °C) was rapidly achieved by heating at 60 °C/hour. Dew point was maintained during the entire run at 2 ° C below the chamber temperature. Fan flow through the box stacks reversed every 15 minutes. The chamber temperature was held at 23 °C until the CA reached 1% oxygen and 15% carbon dioxide, which required at least 30 to 60 minutes. When the correct CA was reached the chamber was heated at 12 °C/h until the chamber reached 46 °C (115 °F) where it was held until all probed fruit core temperatures had reached and remained at 43.8 °C for 30 minutes. This was the end of the treatment. Since the testing was preliminary and heat flow through the boxes was of the greatest interest, no quality data was taken and instead fruit were used for at least two runs.

RESULTS

Semi-commercial CATTS chamber

A total of seven treatment runs were conducted with the semi-commercial chamber. The results for each run are as follows:

Run 1: *2-layer boxes filled with size 64 yellow nectarines, 56 fruit/box, 80 boxes per pallet, 2 pallets.*

- Was a testing run utilized to help set up the chamber when the chamber designer was still in the installation process. Problems with the heating (software issues) makes it difficult to utilize the data for any type of comparisons.

Run 2: *2-layer boxes filled with size 56 August Red nectarines, 56 fruit/box, 80 boxes per pallet, 2 pallets.*

- Stopped run prior to cores reaching final temperature due to hardware problems with the CA system. Later resolved hardware issues.

Run 3: *2-layer boxes filled with size 56 August Red nectarines, same fruit as Run 2.*

- Total run time would have been around 7 hours (including time for CA establishment). This is largely due to a very slow heating rate (Figure 1).
- The heating rate among the boxes was very uneven, as the coolest box required 329 minutes to reach 43.8 °C core and the warmest 163 minutes (Figure 2). Warmest were in the outside corners of the stack and coolest were inside boxes in the middle of the stack.

Run 4: *2-layer boxes, size 56 yellow nectarines, 56 fruit/box, 80 boxes per pallet, 2 pallets. Replaced some of the standard boxes with boxes with enlarged vents and others with enlarged vents and apple trays, all done to test the effects of improved airflow on heating.*

- a software problem caused loss of data from this run.

Run 5: *2-layer boxes, size 56 yellow nectarines, same fruit as Run 4. Replaced some of the standard boxes with boxes with enlarged vents, enlarged vents and apple trays, and others with plastic RPCs, all done to test the effects of improved airflow on heating.*

- Heating rates (time needed to reach 43.8 °C) were faster in the altered boxes/RPC's, especially for enlarged vent + apple trays:

	Normal box	Enlarged vents	Enlarged vents + apple trays	RPCs
Time needed for coolest box	338 minutes	326 minutes	232 minutes	306 minutes

- Difficult, however, to use this data to make comparisons with other runs as the full stack was not modified.

Run 6: Volume-filled boxes, size 56 September Red nectarines.

- Purpose was to test whether volume-filled boxes without trays might heat more effectively than boxes with trays.
- Very long treatment time needed. More than 9 hours would be needed if the time to establish CA was included. A problem with the seals around the boxes may have contributed to the long treatment time.

Run 7: Picking totes (8 high), size 56 September Red nectarines (same fruit as Run 6).

- Used picking totes to try to enhance airflow. The totes were used for the entire 2 pallets.
- Total run time would have been 6.9 hours, which is very similar to the time required using 2-layer boxes. The lack of much improvement in the time needed could have been partly due to the greater amount of fruit in each tote as compared to a 2-layer box.
- The average heating rate was similar to that of the 2-layer boxes, but much slower than that for the laboratory chamber (Figure 1).
- Heating was very uneven among the totes (Figure 2) with corner, outside boxes being the warmest and inside boxes the coldest.

Entomological Research – CA water bath

The regular air (RA) treatments on codling moth CM indicated that the 5th instar was the most thermotolerant larval stage (Table 1). However, when the controlled atmosphere was applied the mortality of the instars was not significantly different from each other. Previous in-fruit tests identified the 4th instar as being the most tolerant, but the tests are difficult to compare as the heating rates were different.

In agreement with test results from CM eggs treated on fruit in the CATTS chamber, the egg stages did not differ from each other in terms of tolerance for both the RA and CA treatments (Table 2).

For Oriental fruit moth (OFM) larvae under RA conditions, the 4th and 3rd instars were equally thermotolerant to each other and more thermotolerant that the 1st and 2nd instars (Table 3). Under CA treatments, the 1st instar was significantly less tolerant than the 2nd – 4th instars, which were equally tolerant to one another. This is in contrast with previous results obtained from in-fruit treatments, where the 4th instar was determined to be the most tolerant to CATTS. Again, the difference in heating rate could have been responsible.

For OFM eggs under RA, all of egg stages had a similar tolerance (Table 4). Under CA conditions there were also no differences in tolerance. These results are in agreement with previous findings using fruit treated in a CATTS chamber.

When the larval mortalities of both species were compared under CA conditions, neither species was significantly different from each other. When only the 4th and 5th instars of CM were compared with the 4th instar of OFM, no stage or species were significantly different from each

other. These findings are in contrast with previous research where the 4th instar of CM was determined to be more tolerant than the 4th instar OFM.
When the response of the egg stages are compared, codling moth were more tolerant of the CA and RA treatments than the OFM eggs. This is in contrast to previous studies where the whitehead stage of oriental fruit moth was more tolerant than the CM whitehead stage. However, the red ring and black head stages of both species were not significantly different from each other.

Entomological Research – CA heat block
We evaluated the CA heat block system using codling moth and found the results to be compatible to those obtained using the CA water bath system. We are now in the process of testing oblique banded leafroller (OBLR) in the CA heat block system. Initial results indicate that the last two instars of OBLR, the most thermotolerant stages, are less resistant to CATTS treatments than codling moth and Oriental fruit moth.

Entomological Research – Other Tests
We have obtained fruits infested with San Jose scale (SJS), European red mite (ERM) diapausing eggs, and diapausing spider mites (SM) for tests in the CATTS system. These tests need to be performed on infested fruits in the laboratory test chamber. Initial trials with diapausing eggs of ERM resulted in 100% mortality (0% egg hatch) following treatment of infested apples using a 12°C/h heating rate to 46°C under a 1% O_2, 15% CO_2 environment. Preliminary tests on non-diapausing spider mites indicated that the deuteronymph was the most resistant stage to the 24°C/h, 46°C CATTS treatment. There were no diapausing females in this trial. Test on SJS are a bit more difficult since the adults and immatures are more sessile, and determination of mortality requires numerous evaluations over time. Our goal is to treat over 5,000 of the most resistant stage of these pests with the same treatments currently developed to control CM and OFM.

SUMMARY

Results from a series of seven separate treatment runs using the new semi-commercial CATTS chamber indicated that the rate of heating of the fruit in the large chamber is substantially slower than that previously observed in the laboratory CATTS chamber, resulting in much longer treatment times. Total treatment times using the large chamber were around 7 hours versus 3.5 hours for the laboratory chamber. Although it is yet to be determined, these longer treatment times could be injurious to the quality of the fruit and may affect the insect efficacy. Also observed was the rate of heating among the boxes in the stacks is not equal, and will result in some fruit receiving a much greater heat dose than others. The long treatment times and unequal heating are likely partially attributable to the requirement in the large chamber that the treatment be conducted using boxed fruit. Altering the boxes to improve airflow or using plastic containers with greater venting did act to enhance the heating rate but further experimentation is needed to determine the full effect of these alterations.

The CA water bath system was developed to provide a lower cost and faster alternative to the conventional means of quarantine testing using large treatment chambers and infested fruit. This system was evaluated on the immature stages of CM and OFM and found to be effective for this

purpose. Another system that is being developed to facilitate treatment development is the CA heat block system. This system has an advantage over the CA water bath system in that it is able to treat larger numbers of insects at the same time, enabling a more robust statistical analysis and more accurate determination of treatment tolerance. Testing using OFM found the two systems to give comparable results, indicating that the CA heat block system can be used to speed the development of CATTS quarantine treatments.

Figure 1. Temperature curves (internal fruit seed surface temperatures) from runs performed in a laboratory CATTS chamber or in the semi-commercial CATTS chamber using 2-layer boxes or picking totes.

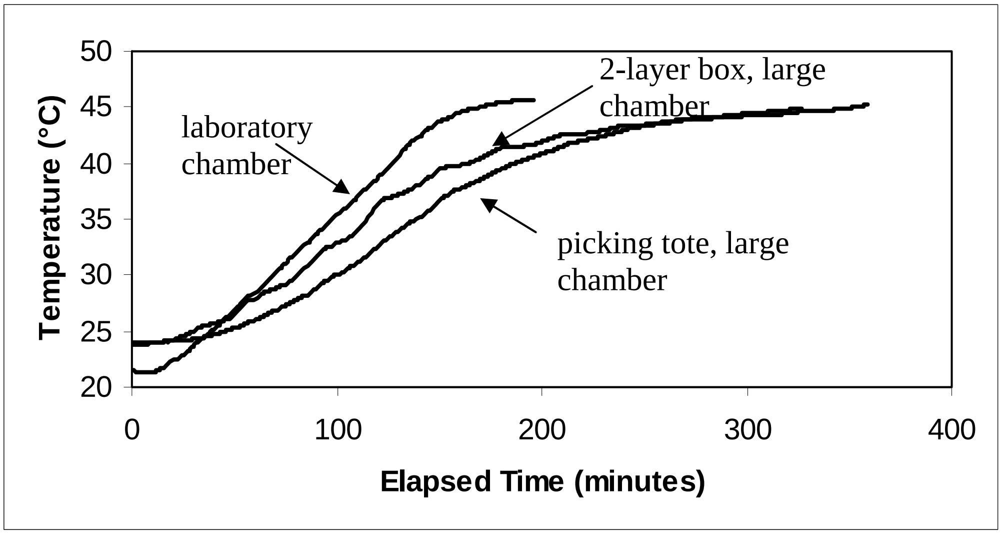

Figure 2. Time in minutes needed for the probed fruit in each box to reach 43.8 °C for Run 3 (2-layer boxes) and Run 7 (plastic totes). Four diagrams are given for each run, indicating the rear pallet (front and back row) and front pallet (front and back row), with each square indicating an individual box.

2-Layer Boxes (Run 3)

REAR PALLET

Front Side

210		264	189
	263	258	
	292	297	
263	303	329	253
	266	301	
	283	278	
		297	
	280		
187	237	248	223

Back Side

261	266	266	
	258	234	
	244	254	
	239	255	
249	269	285	260
	265	282	
	283		
		268	
	264		
173		199	264

FRONT PALLET

Front Side

198		236	220
		225	
	195		
234	204	230	
	241		
	251		
	238	224	
	197	213	163

Back Side

181	230	253	223
	266	231	
	262	262	
	249	274	
230	240	250	226
	247	257	
	261		
		246	
186		222	185

Picking Totes (Run 7)

REAR PALLET

Front Side

297		338	307
	322	313	
	303		321
		322	
267	287	317	
front			

Back Side

292	327	305	
		317	
	327	321	
302	327	336	307
	322		
		327	
	337		
230, 260		322	230

FRONT PALLET

Front Side

297		307	279
		296	
		322	251
	298		
		278	
	297		
			221

Back Side

297	322	306	
299	332	328	321
	327		
	325		287

- 100 – 200 min
- 201 – 300 min
- 300+ min
- never reached 43.8 °C

Table 1. Proportion corrected mortality (± SEMs) of codling moth larval stages following heat treatments in CAWB system under air (RA) and controlled atmosphere (CA) environments. CA conditions were 1% O_2, 15% CO_2. Beginning temperature was 23°C and ending temperature was 45.5°C. Heating rate was 24°C/hr. Times are total times from the start of the heat treatment.

	1st		2nd		3rd		4th		5th	
Time	RA	CA	RA	CA	RA	CA	RA	CA	RA	CA
0.5 h	0.150±0.084	0.223±0.128	0.461±0.176	0.362±0.097	0.113±0.065	0.362±0.087	0.233±0.134	0.460±0.177	0.105±0.092	0.056±0.089
1.0 h	0.493±0.103	0.978±0.081	0.311±0.092	0.532±0.151	0.311±0.193	0.532±0.156	0.215±0.085	0.204±0.036	0.144±0.101	0.290±0.112
1.5 h	0.785±0.051	0.975±0.049	0.424±0.049	0.672±0.082	0.424±0.045	0.672±0.067	0.503±0.035	0.934±0.034	0.067±0.121	0.857±0.119
2.0 h	0.940±0.138	1.00±0.00	0.844±0.040	1.00±0.00	0.844±0.049	1.00±0.00	0.896±0.049	0.986±0.012	0.288±0.140	1.00±0.00

Table 2. Proportion corrected egg hatch (± SEMs) of codling moth egg stages treated in CAWB system under air (RA) and controlled atmosphere (CA) environments. CA conditions were 1% O_2, 15% CO_2. Beginning temperature was 23°C and ending temperature was 45.5°C. Heating rate was 24°C/hr. Times are total times from the start of the heat treatment.

	White		Red Ring		Black Head	
Time	RA	CA	RA	CA	RA	CA
0.5 h	1.00±0.022	0.97±0.038	0.941±0.022	0.875±0.044	0.956±0.044	0.769±0.087
1.0 h	0.877±0.067	0.657±0.128	0.908±0.019	0.684±0.068	0.672±0.195	0.352±0.155
1.5 h	0.781±0.019	0.381±0.068	0.793±0.044	0.434±0.091	0.574±0.067	0.204±0.080
2.0 h	0.422±0.120	0.00±0.00	0.300±0.068	0.00±0.00	0.289±0.062	0.00±0.00

Table 3. Proportion corrected mortality (± SEMs) of oriental fruit moth following heat treatments in CAWB system under air (RA) and controlled atmosphere (CA) environments. CA conditions were 1% O_2, 15% CO_2. Beginning temperature was 23°C and ending temperature was 45.5°C. Heating rate was 24°C/hr. Times are total times from the start of the heat treatment.

	1st		2nd		3rd		4th	
Time	RA	CA	RA	CA	RA	CA	RA	CA
0.5 h	0.00±0.041	0.973±0.027	0.336±0.140	0.259±0.033	0.218±0.0454	0.391±0.074	0.00±0.009	0.167±0.012
1.0 h	0.905±0.063	1.00±0.00	0.897±0.083	0.654±0.019	0.351±0.217	0.670±0.076	0.333±0.174	0.986±0.009
1.5 h	0.911±0.059	0.971±0.028	0.800±0.046	1.00±0.00	0.814±0.123	1.00±0.00	0.796±0.086	1.00±0.00
2.0 h	0.882±0.068	1.00±0.00	0.701±0.131	1.00±0.00	0.707±0.062	1.00±0.00	0.572±0.147	0.974±0.026

Table 4. Proportion corrected egg hatch (± SEMs) of oriental fruit moth egg stages treated in CAWB system under air (RA) and controlled atmosphere (CA) environments. CA conditions were 1% O_2, 15% CO_2. Beginning temperature was 23°C and ending temperature was 45.5°C. Heating rate was 24°C/hr. Times are total times from the start of the heat treatment.

	White		Red Ring		Black Head	
Time	RA	CA	RA	CA	RA	CA
0.5 h	0.436±0.069	0.121±0.038	0.430±0.040	0.178±0.029	0.203±0.029	0.076±0.033
1.0 h	0.360±0.096	0.022±0.017	0.184±0.040	0.00±0.00	0.302±0.112	0.024±0.022
1.5 h	0.534±0.250	0.054±0.022	0.472±0.135	0.012±0.011	0.536±0.204	0.00±0.00
2.0 h	0.303±0.145	0.00±0.00	0.301±0.123	0.020±0.009	0.251±0.086	0.00±0.00