FINAL PROJECT REPORT WTFRC Project Number:

Project Title: Protocols for conditioning Anjou pears with ethylene release capsules

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Other funding Sources
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Agency Name: Amount awarded: Notes:

Total Project Funding: US\$19,500

Budget History:

Item	Year 1:	Year 2:	Year 3:
Salaries			
Benefits			
Wages			
Benefits			
Equipment	900		
Supplies	2,145		
Travel	16,455		
Miscellaneous			
Total	19,500		

Objective To establish protocols for conditioning pre-packed loads of Anjou pears using Ethylene Release Capsules (ERCs) at three different times during the pear season (fall, winter, spring*). *N.B. The spring trial was dropped, with agreement from the Fresh Pear Committee, based on advice from our collaborating packer/shipper that it was unlikely that conditioned fruit in spring would survive delivery in a saleable state.

Pallet scale conditioning using Ethylene Release Capsules

The usefulness of smaller prototype ERCs for conditioning pears on an individual clamshell or box scale has been demonstrated previously (Sharrock and Henzell, 2007). The ERC tested in the trials reported here to condition whole pallets was a much larger device, which is basically contained within an aerosol can (Fig. 1). A key feature is its unique internal valve (patent pending) that releases compressed ethylene at a constant rate for an adjustable period of up to seven days. This is achieved without electronics or moving parts, thus maximizing its reliability and cost-effectiveness.



Figure 1. Covered pallet conditioning using an ERC (enlarged in insert)

Significant findings

- ERCs provide an effective means of conditioning early season Anjou pears within covered pallets of either double-layer Euro-boxes or standard cartons of individually wrapped fruit.
- A single ERC at the top of a covered pallet resulted in >100 ppm ethylene throughout the pallet within 17 hours and maintained this for a pre-defined period of up to 7 days.
- Early in the Fall, Anjou conditioned by ERC for 5-7 days were **distinctly more aromatic and flavorsome**, once fully ripe, than those conditioned conventionally at higher temperatures for a shorter period. Fruit that were simply warmed without ethylene failed to ripen acceptably.
- In February, using CA-stored fruit, ethylene conditioning had no effect on subsequent rates of softening but still resulted in detectable aroma and flavor enhancement.
- Covering alone had little impact on rates of warming and softening of fruit, and no detrimental effects became evident during subsequent ripening in either Winter or Fall.

Results and discussion

Winter Conditioning Trial (February 07)

This trial involved five pallets each containing 12 layers of double layer Euro-boxes of 70 ct Anjou (42 per box). The fruit had been picked on 13 Sept. 07, sealed in CA at Underwood Fruit Packers, White Salmon on 16 Sept, removed from CA on 2 February, packed on 8 February, and held at 30°F in RA

storage until the trial commenced on 14 February. Each pallet except the uncovered control C3 was rebuilt on top of a sock created by cutting down a polyethylene pallet cover, which in turn was seated on a slip sheet to protect it from damage by the slats of the pallet base. Miniature "iButton®" temperature loggers (Maxim Integrated Products Inc.) were attached directly to fruit with rubber bands, within five labeled representative boxes located on layers 1 (top), 3, 6, 10, 12 (base) within each pallet. Each pallet was then subjected to one of the following five treatments in a disused store room with minimal electric heating sufficient to achieve and maintain an air temperature of 60-65°F.

Treatments:

T1. 3 days conditioning with ERC inside a sealed pallet cover; after 2 days uncovered;

T2. 5 days conditioning with ERC inside a sealed pallet cover, with no pre-warming period.

Controls

- C1. As for T1 but without an ERC;
- C2. As for T2 but without an ERC;
- C3. Simply left uncovered for 5 days.

Temperatures of fruit and the surrounding room, and concentrations of CO_2 , O_2 and ethylene inside the boxes at various levels within the pallets were monitored throughout conditioning using portable meters (PBI Dansensor and Dräger). Typical examples of the data obtained are shown in Figs 2 & 3.



Figure 2: Temperatures of fruit in selected layers of the T2 covered pallet, and of the surrounding room air during conditioning in February 07.

Figure 3: Carbon dioxide and ethylene concentrations in layers 1 and 6 of the T2 covered pallet in February 07.

6

5

2

1

0

Carbon dioxide (%)

Effective warming of the pallets only began at the end of the first day, when electric heaters were introduced to counter the effects of cold weather. The fruit in the top layer warmed up much more rapidly than those further down the pallet. The middle (layer 6) was the slowest to warm up, remaining 10° F and 5° F colder than the top (layer 1) and bottom (layer 12) respectively, after 3 days of warming (Fig. 2). Layers 3, 10 and 12 all warmed at similar rates. In contrast, concentrations of ethylene and CO₂ showed no significant difference between the middle and top layers Fig. 3). This knowledge of relative temperatures and gas distribution within the pallets is pertinent to interpretation of positional effects on responses to ethylene.

Ethylene concentrations within the covered pallets T1 and T2, which each contained an ERC, exceeded 100 ppm within 17 hours of covering (possibly sooner, but no earlier assessment was done). Levels of >100 ppm ethylene, generally accepted as sufficient for conditioning pears, were maintained by the ERCs in covered pallets T1 and T2 for three and five days, respectively. The covered control pallets, with no ERCs, were found to also contain ethylene, albeit at a much lower concentration of around 10 ppm, from the first assessment at 17 h after covering until the end of the conditioning period. This was due to

production of ethylene by the pears (that had been in storage for 5 months), since regular monitoring of the store room atmosphere revealed < 0.5 ppm ethylene.

After conditioning, the pallets were dismantled so that sample boxes of fruit from layers 1, 3, 6, 10 and 12 of each pallet could be removed and ripened at 68°F at MCAREC. The remaining bulk of each pallet was returned to 30°F storage and sold. During ripening, samples of each batch of fruit were assessed daily in terms of firmness and aroma production (using ripeSense® sensors).

Sealing the pallets inside a polyethylene cover during the conditioning period had no detectable effect on subsequent softening (Fig. 4B & C). Exposure to ethylene within the pallet cover also had little effect on the rates of softening (Fig. 4A & B). Fruit of treatments T1 and C1 (not shown) exhibited almost identical softening curves to the above. In all treatments, fruit of the top layer of the pallet began softening sooner than those of lower layers (Fig. 4), which is probably a reflection of their different rates of warming during the conditioning period, as revealed in Fig. 2.





Figure 4: Firmness changes in Anjou pears from the February 07 trial during the conditioning period (until day 5) and subsequently during ripening at 68° F, involving samples from five layers within each pallet (.01 = top layer; .12 = bottom layer).

A: Treatment T2 (5 days of ethylene conditioning in covered pallet).

B: Covered control C2 (5 days covered, but without ethylene).

C: Uncovered control pallet C3.

Ethylene production by fruit from all five pallets, including the controls, was evident when the fruit were placed in ripeSense® clamshells during the final 20 h of ripening. There appeared to be no relationship between the rate of ethylene production during ripening and whether fruit had earlier been artificially exposed to >100 ppm ethylene during conditioning. The capacity of these fruit, following 5 months storage, to produce their own ethylene is a key point of difference between this trial and the subsequent investigation in September 07 using freshly harvested fruit (see below).

Aroma development by the ripening fruit proved difficult to assess reliably by the method employed during the February trial, in which fruit were sealed in clamshells containing ripeSense® sensors for just 20 h before colors of the sensors were assessed and the fruit were subjected to destructive firmness testing. This incubation period proved too short to obtain reliable and meaningful sensor responses, and

consequently this was increased substantially during the following trial in Fall 07 (below). Based on sense of smell alone, the two researchers involved were usually able to distinguish between ethylene-treated and control fruit in blind testing, but the differences in aroma were subtle. In contrast with controls in the Fall trial (which remained virtually inedible), the February control fruit produced appreciably more aroma, and could be enjoyably eaten.

Taste comparisons performed "blind" by the two researchers on February trial fruit, fully ripened to around 2 lb firmness, revealed definite flavor differences between ethylene conditioned and control fruit. Ethylene treatment resulted in a more intense and attractive flavor. However, the flavor difference was far less marked than in the September trials (see below), probably because the February fruit, including the controls, were capable of producing their own ethylene.

Fall Conditioning Trial (September 07)

Green Anjou pears, all from the same lot, were harvested at commercial maturity on September 13th 2007, and kept in cold storage until grading and packing 12 days later. Packed 90 ct trial fruit (seven pallets of standard cartons and six pallets of double-layer Euros) were returned to 30°F storage overnight.

On September 26th, all thirteen pallets were rebuilt to permit gas sampling tubes, thermocouples and iButton® temperature loggers to be attached to fruit within the bottom, middle and top layers of each and then either sealed within pallet covers, as in February (see above), or left open, either as controls, or to be treated with ethylene in the conventional manner. Pallet labels denoted the treatments to be employed during conditioning (listed below), preceded by either an "S" (for standard carton) or an E (for Euro). Standard carton pallets received all seven treatments below. Euro-boxes received only six treatments (not T4). Ten of the pallets spent the conditioning period in an unheated dry-store warehouse, with an air temperature that fluctuated around 60°F. Exceptions were ST3 & ET3, treated in a conventional heated conditioning room at Underwood Fruit, and ST4, which was conditioned in the Underwood apple packing room (warmer than the warehouse but containing variable, and sometimes significant amounts of ethylene from the apples).

Ethylene treatments

T1: Two days uncovered, then five days covered with ERC in dry goods warehouse

T2: Seven days covered with ERC in dry goods warehouse

T3: Conventional conditioning (one day warming, **two days ethylene from catalytic generator** in Underwood's normal conditioning room)

T4: Two days uncovered, then four days covered with ERC in apple packing room throughout

Controls

- C1: Two days uncovered, then five days covered without ERC in dry goods warehouse
- C2: Seven days covered without ERC in dry goods warehouse

C3: Seven days uncovered without ERC in dry goods warehouse

Ethylene in the room atmosphere in the warehouse was generally <0.5 ppm, but in the apple packing room sometimes reached 20 ppm. Temperatures of fruit and the surrounding room, and concentrations of CO₂, O₂ and ethylene inside the boxes at various levels within the pallets were monitored throughout conditioning. Gas samples were collected via tubes to the interiors of boxes in the top, middle and bottom layers. Examples typical of the data obtained are shown in Figs 5, 6 & 7.



Figure 5: Temperatures of fruit within pallets in the top (T) middle (M) and bottom (B) layers. Graphs **A & B** relate to uncovered control pallets; **C & D** to covered control pallets; **E & F** to covered ERC-treated pallets; **G** to conventional conditioning of an uncovered pallet of pears in Euroboxes (ET3) in a commercial ethylene room; **H** to a covered ERC-treated pallet of pears in standard cartons in the Apple Packing Room at Underwood Fruit. Air temperatures of the surrounding room (dry goods warehouse) shown in A also apply to B-F.

iButton® temperature loggers attached to fruit at various levels within the pallets clearly demonstrated the very significant thermal inertia of full, tightly packed pallets of pears in both box types (Fig. 5). The middle layer warmed up much more slowly than the top layer, resulting in temperature differentials of up to 15° F within the same pallet. This temperature gradient was less apparent in the uncovered (Fig 5 A & B) than in the covered pallets (C & D), but still existed even in the pallet of Euro-boxes subjected to forced air warming and ethylene conditioning in a commercial conditioning room (Fig. 5G). Metabolic warming of the middle layers due to ethylene-induced acceleration of fruit respiration was barely detectable, based on comparison of rates of internal warming of pallets ET2 and ST2 (Fig 5 E & F) vs the covered controls without ethylene (C & D).



Figure 6. Oxygen and carbon dioxide concentrations within covered pallets during conditioning. A & B compare ethylene treatment ST4, involving a pallet of standard boxes, with the most relevant control, SC1, covered for a similar period but without an ERC included. C & D compare the longest (7 day) conditioning period (ET2) with its corresponding no-ethylene control (EC2). T = top; M = middle; B = bottom layer.

Substantially modified atmospheres developed within the pallets sealed inside polyethylene covers. Examples shown in Fig. 6 A & B displayed the greatest atmospheric modification, with O_2 falling below 1% and CO_2 rising to 13% by the end of the period of enclosure (5 days). No detrimental effects of these short exposures to such modified atmosphere were detected during subsequent cold storage by Underwood's quality inspectors, and excess fruit from covered pallets were deemed fit-for-sale. Inclusion of an ERC had no obvious impact on the rate of accumulation of CO_2 and depletion of O_2 within the covered pallets. There was considerable variation between pallets in the final levels of CO_2 and O_2 , almost certainly attributable to variable losses of accumulated gases during daily sampling of fruit from the top of each sealed pallet, followed by an attempt to completely reseal the damaged polyethylene. In commercial use, internal sampling and resealing during the course of conditioning of a covered pallet would of course not normally be necessary.



or ST), compared with corresponding controls (EC or SC). "E" signifies Euro-box; "S" signifies standard carton. Treatments shown in the figure for pallets of Euro-boxes were the same as those applied to pallets of standard cartons graphed on that same level.



Ethylene within the five covered pallets each containing an ERC rose rapidly to around 50 ppm by 3.5 h after sealing. The ethylene concentrations in each of these pallets continued to climb, exceeding 100 ppm by one day after sealing. The levels fluctuated and varied between pallets during the period of conditioning (Fig. 7), due primarily to loss of ethylene during daily opening and closing of the top of the pallet cover to remove fruit, and, in some cases, to inadequate resealing. Despite these difficulties, the rate of continued ethylene release from the ERCs was in all cases sufficient to maintain at least 100 ppm ethylene around the fruit in the covered pallets for the entire conditioning period of up to 7 days (Fig. 7).





The control pallets that were covered but contained no ERCs did not accumulate any detectable ethylene (in contrast with the February trial, when the control fruit was capable of producing its own ethylene). Ethylene concentrations in the conventional conditioning room during conditioning of pallets ET3 and ST3 were 155 and 175 ppm during the first 24 h of ethylene exposure, but had fallen to 40 ppm by the end of the 48 h ethylene treatment.

Ripening following conditioning was carried out using fruit from sample boxes from the top, middle and bottom of each pallet. The remaining boxes were returned to 30° F after conditioning, inspected and deemed fit-for-sale. The sampled fruit were ripened in ripeSense® clamshells in the same dry goods warehouse in which most had been conditioned, at an uncontrolled temperature that averaged around 60° F. Ethylene, CO₂ and O₂ levels (not shown) and fruit firmness (Figure 8) within representative clamshells were measured daily. Aroma development (Figure 9) was monitored during ripening using ripeSense® sensor labels.

Ethylene production during ripening of these early season fruit, in marked contrast with the February trial, was restricted to only those fruit that had been conditioned with ethylene. Those that had been conditioned for the longest period (pallets ET2 and ST2) produced the most ethylene, which accumulated in their clamshells to almost 10 ppm by the end of ripening (data not shown).

Softening rates during ripening were also clearly dependent upon conditioning with external ethylene (Fig. 8). Controls (C1, C2 and C3), which were warmed and, in some cases, covered but not ethylene treated during conditioning, were invariably far slower to soften than the corresponding ethylene treatments (Fig. 8). Fastest softening occurred in the top layer of each ethylene-treated pallet. The T3 treatments (conventional conditioning room, Fig. 8 E&F) produced the fastest mean rates of softening across the whole pallets. This probably reflects the faster rates of warming of the T3 and top layer fruit during conditioning (Fig. 5), which meant that they more rapidly reached temperatures at which they were able to respond to ethylene.

Aroma production during softening was distinctly greater in fruit that had received the longer exposures to ethylene during conditioning (Fig. 9). These data are based on ripeSense® sensor responses, but corresponded with what our noses and taste buds told us. T1 & T2 fruit, conditioned with ethylene from ERCs for 5 and 7 days respectively in covered pallets, were clearly preferable in aroma and flavor criteria over T3 fruit, which had received the standard commercial conditioning involving one day of warming and two days of ethylene exposure. The ripened T1 and T2 fruit were generally acknowledged as excellent eating quality, which for Anjou is remarkable in view of the fact that they had been harvested just 10 days prior to commencement of the September conditioning trial. The conventionally conditioned T3 fruit softened to the same extent, but developed very little aroma as they softened (Fig 9 E&F), in comparison with fruit of T1 (A&B) and T2 (C&D). T4 fruit (Fig. 9G), which received a shorter, 4 day conditioning at a slightly higher mean temperature than T1 and T2, did not produce as much aroma as fruit of the latter two treatments, but were still more aromatic than the conventionally conditioned fruit.

These observations of the aroma and flavor benefits resulting from prolonged (5-7 days) ethylene exposure of Anjou pears during the first month after harvest are in keeping with the results of our previous project, obtained over several seasons (Sharrock and Henzell, 2007). The ERC, expected to be commercially available soon, offers a practical means of achieving such long conditioning periods at minimal extra cost, and with improved logistical flexibility.

Reference: Sharrock, K. R. and Henzell, R. F. (2007) Ethylene ripening of pears by unconventional means. Final Project Report. NW Pear Research Review 2007, 10 pp. (N.B. Complete report, including last 4 pages omitted from Review Proceedings and CD, is obtainable from the authors).

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