PROGRESS REPORT

Project number: PMG 02-24

1. PROGRAMME AND PROJECT LEADER INFORMATION

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2. PROJECT INFORMATION

Project title	Develop a cold storage protocol to maintain the overall fruit quality of Wonderful pomegranates
Short title	Storage protocol to maintain the overall fruit quality

Fruit kind(s)	Pomegranates		
Start date (mm/yyyy)	03/2024	**End date** (mm/yyyy)	09/2024

Key words	Pomegranates, cold storage, protocol, cooling, postharvest, quality

POMASA	R 102 030.00
APPROXIMATE PROJECT COST	

3. ABSTRACT

Objectives and rationale

Post-harvest cold storage protocols are defined to maintain the fruit quality and limit cold storage disorders. The mandate was to assess the current cold-storage protocol used for pomegranates and conduct preliminary testing with the view to identify the best protocol.

Methods

Different rapid and slow cooling procedures were tested as well as the impact of different holding temperatures during subsequent cold storage. We suspect that rapid cooling of pomegranates might damage the fruit cells and that this may manifest as surface and pulp membrane browning after cold storage.

Key results

Step-down cooling to 7.5 °C had a positive effect in reducing pulp membrane and albedo browning, followed closely by pomegranates forced-air cooled to 7.5 °C. The key here was the slightly higher cold storage temperature, compared to the traditional 5.0 °C. If “Black heart” decay can be controlled, these fore-mentioned treatments show potential for commercial application. However, these initial research findings need to be confirmed in more research.

4. PROBLEM IDENTIFICATION AND MOTIVATION

Problem identification

Post-harvest cold storage protocols are defined to maintain fruit quality and limit possible cold storage disorders. ExperiCo received a mandate to assess the current cold-storage protocol used for pomegranates and do preliminary testing to identify best protocol possibilities. The main requirement is to develop a handling protocol that will limit external and internal disorders for cold stored pomegranates.

Motivation

This project aims to identify a cold storage protocol that will assist in reducing or limiting surface and pulp membrane browning with pomegranates subjected to sea export to overseas markets. Different rapid and slow cooling procedures were tested, as well as the impact of different holding temperatures during subsequent 6 weeks cold storage. We suspect that rapid cooling might damage the fruit cells during cooling and that this may manifest as surface and pulp membrane browning after cold storage.

5. ACCUMULATED PROGRESS TABLE

Objectives	Milestones (Significant event or stage in a project)	Date achieved
Objective 1: Provide essential information on how to cold store Wonderful pomegranates	Identify the correct holding temperature to cold store Wonderful pomegranates to reduce disorders, control decay and maintain general quality	August 2024
Objective 2: Determine the effect of forced-air cooling versus step-down cooling	Determine the effect of forced-air cooling versus step-down cooling on fruit quality after cold storage	August 2024
Objective 3: Further research will be identified and a separate proposal will be made	Submit written report on research findings and make recommendations for next steps if appropriate	Report date – Sept 2024

6. WORKPLAN (MATERIALS AND METHODS)

Treatments:

Two populations of Wonderful pomegranates packed for export were sourced from different commercial packhouses. The fruit was packed in the same standard commercial packaging.

The following treatments were used:

T1: Forced-air cooling (FAC) to 5 °C
T2: Forced-air cooling (FAC) to 7.5 °C
T3: Step down cooling to 5 °C (→ 10 °C (24 Hours) → 7.5 °C (24Hours) → 5.0 °C)
T4: Step down cooling to 7.5 °C (→ 10 °C (24 Hours) → 7.5 °C)

Eight (8) single carton replicates per treatment was used (14 Fruit per carton).
Fruit quality data after cold storage were analysed by a One-way ANOVA.

Cold storage:

Fruit was cold stored for 6 weeks at the respective temperature treatments followed by 7 days shelf life at 20 °C.

Examination parameters:

<u>At harvest:</u>

For each population of pomegranates sourced, the following parameters were assessed on one representative carton of fruit (14 fruit evaluated per parameter):

- Total soluble solids (TSS) (%), on a pooled sample of juice of each replicate using an Atago DBX-30 digital refractometer
- Malic acid (MA), on a pooled sample of juice by titrating a 10 g aliquot with 0.1N NaOH to a pH end point of 8.2 using a Metrohm 605 Dosimat titrator

<u>After cold storage and shelf-life:</u>

The following parameters were assessed for each replicate carton of fruit (7 fruit evaluated per parameter):

- Photographs were taken at storage examinations (Figure 1)
- Incidence of crown rot, by expressing the number of pomegranates with decay, as a percentage of the sample size
- Incidence of 'black heart' decay, by expressing the number of pomegranates with internal decay as a percentage of the sample size
- Incidence of fruit with surface decay (incl. *Botrytis* sp. and *Penicillium* sp.), by expressing the number of pomegranates with surface decay as a percentage of the sample size
- The severity of fruit with surface browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), also given as a percentage of each rating

- Severity of fruit with pulp membrane (septum) browning (rating 0 – 3, with 0 = none, 1 = slight, 2 = moderate, 3 = severe), also given as a percentage of each rating
- Severity of fruit with albedo browning (rating 0 – 3, with 0 = none, 1 = slight, 2 = moderate, 3 = severe), also given as a percentage of each rating
- Severity of fruit with aril browning (rating 0 – 3, with 0 = red, 1 = a few light brown arils, 2 = less than half of arils brown, 3 = majority of arils are brown)

7. RESULTS AND DISCUSSIONS

Table 1a: Browning severity of population 1 on Wonderful pomegranates after 42 days cold storage at various temperatures

Treatment	Parameters[2]											
	Surface browning				Pulp membrane browning				Albedo browning			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)
FAC to 5 °C	0.0	0.0	0.0	0.0	17.9	12.5	3.6	33.9bc	8.9	8.9b	0.0	17.9
FAC to 7.5 °C	0.0	0.0	0.0	0.0	10.7	1.8	0.0	12.5ab	3.6	0.0a	0.0	3.6
Step cooling to 5 °C	0.0	0.0	0.0	0.0	21.4	10.7	5.4	37.5c	12.5	8.9b	0.0	21.4
Step cooling to 7.5 °C	0.0	0.0	0.0	0.0	1.8	0.0	1.8	3.6a	5.4	0.0a	0.0	5.4
Prob.>F[1]	-	-	-	-	0.0606	0.1599	0.6400	0.0082	0.4758	0.0477	-	0.0680

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.
2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 1b: Browning severity and decay incidence of population 1 on Wonderful pomegranates after 42 days cold storage at various temperatures

Treatment	Parameters							
	Aril browning[2]				Incidence (%)			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Crown Rot	Black heart decay	Surface decay	Total decay
FAC to 5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
FAC to 7.5 °C	0.0	0.0	0.0	0.0	0.0	5.4	7.1	12.5
Step cooling to 5 °C	0.0	0.0	0.0	0.0	0.0	1.8	1.8	3.6
Step cooling to 7.5 °C	0.0	0.0	0.0	0.0	0.0	7.1	0.0	7.1
Prob.>F[1]	-	-	-	-	-	0.2834	0.0740	0.1900

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.
2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 2a: Browning severity of population 1 on Wonderful pomegranates after 42 days cold storage at various temperatures and 7 days shelf life at 20 °C

Treatment	Parameters[2]											
	Surface browning				Pulp membrane browning				Albedo browning			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)
FAC to 5 °C	0.0	0.0	0.0	0.0a	12.5	26.8b	8.9	48.2b	10.7	19.6	10.7ab	41.1b
FAC to 7.5 °C	0.0	0.0	0.0	0.0a	8.9	1.8a	1.8	12.5a	5.4	3.6	3.6a	12.5a
Step cooling to 5 °C	0.0	0.0	0.0	0.0a	10.7	8.9a	7.1	26.8ab	3.6	7.1	16.1b	26.8ab
Step cooling to 7.5 °C	0.0	3.6	1.8	5.4b	5.4	5.4a	3.6	14.3a	1.8	3.6	1.8a	7.1a
Prob.>F[1]	-	0.0955	0.4074	0.0142	0.7115	0.0166	0.4945	0.0185	0.3322	0.1088	0.0221	0.0106

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.
2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 2b: Browning severity and decay incidence of population 1 on Wonderful pomegranates after 42 days cold storage at various temperatures and 7 days shelf life at 20 °C

Treatment	Parameters							
	Aril browning[2]				Incidence (%)			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Crown Rot	Black heart decay	Surface decay	Total decay
FAC to 5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
FAC to 7.5 °C	0.0	0.0	0.0	0.0	1.8	5.4	0.0	7.1
Step cooling to 5 °C	0.0	0.0	0.0	0.0	0.0	4.9	1.8	10.7
Step cooling to 7.5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Prob.>F[1]	-	-	-	-	0.4074	0.1158	0.4074	0.1701

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.
2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 3a: Browning severity of population 2 on Wonderful pomegranates after 42 days cold storage at various temperatures

Treatment	Parameters[2]											
	Surface browning				Pulp membrane browning				Albedo browning			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)
FAC to 5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
FAC to 7.5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Step cooling to 5 °C	0.0	0.0	0.0	0.0	1.8	0.0	0.0	1.8	0.0	0.0	0.0	0.0
Step cooling to 7.5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Prob.>F[1]	-	-	-	-	0.4074	-	-	0.4074	-	-	-	-

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.

2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 3b: Browning severity and decay incidence of population 2 on Wonderful pomegranates after 42 days cold storage at various temperatures

Treatment	Parameters							
	Aril browning[2]				Incidence (%)			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Crown Rot	Black heart decay	Surface decay	Total decay
FAC to 5 °C	0.0	0.0	0.0	0.0	0.0	14.3	7.1	21.4
FAC to 7.5 °C	0.0	0.0	0.0	0.0	1.8	8.9	10.7	21.4
Step cooling to 5 °C	0.0	0.0	0.0	0.0	0.0	7.1	10.7	17.9
Step cooling to 7.5 °C	0.0	0.0	0.0	0.0	0.0	1.8	0.0	1.8
Prob.>F[1]	-	-	-	-	0.4074	0.2619	0.1158	0.2362

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.

2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 4a: Browning severity of population 2 on Wonderful pomegranates after 42 days cold storage at various temperatures and 7 days shelf life at 20 °C

Treatment	Parameters[2]											
	Surface browning				Pulp membrane browning				Albedo browning			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)
FAC to 5 °C	17.9	0.0	0.0	17.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
FAC to 7.5 °C	23.2	0.0	0.0	23.2	3.6	0.0	0.0	3.6	0.0	0.0	0.0	0.0
Step cooling to 5 °C	30.4	0.0	0.0	30.4	5.4	0.0	0.0	5.4	0.0	0.0	0.0	0.0
Step cooling to 7.5 °C	21.4	0.0	0.0	21.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Prob.>F[1]	0.7556	-	-	0.7556	0.5644	-	-	0.5644	-	-	-	-

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.

2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Table 4b: Browning severity and decay incidence of population 2 on Wonderful pomegranates after 42 days cold storage at various temperatures and 7 days shelf life at 20 °C

Treatment	Parameters							
	Aril browning[2]				Incidence (%)			
	Rating 1 (%)	Rating 2 (%)	Rating 3 (%)	Total (%)	Crown Rot	Black heart decay	Surface decay	Total decay
FAC to 5 °C	0.0	0.0	0.0	0.0	3.6	1.8	0.0	5.4
FAC to 7.5 °C	0.0	0.0	0.0	0.0	5.4	5.4	1.8	12.5
Step cooling to 5 °C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Step cooling to 7.5 °C	0.0	0.0	0.0	0.0	0.0	10.7	0.0	10.7
Prob.>F[1]	-	-	-	-	0.5644	0.4991	0.4074	0.6065

1. Values in the same examination parameter followed by different letters indicate significant differences (P<0.05) according to the Tukey HSD test.

2. Severity of fruit with browning, as related to chilling injury (rating 0 – 3, with 0 = none, 1 = slight, 3 = severe), given as a percentage of fruit with each rating

Figure 1: Photographs of the pomegranates from population 1 showing skin colour after 6 weeks of cold storage at the different temperature regimes

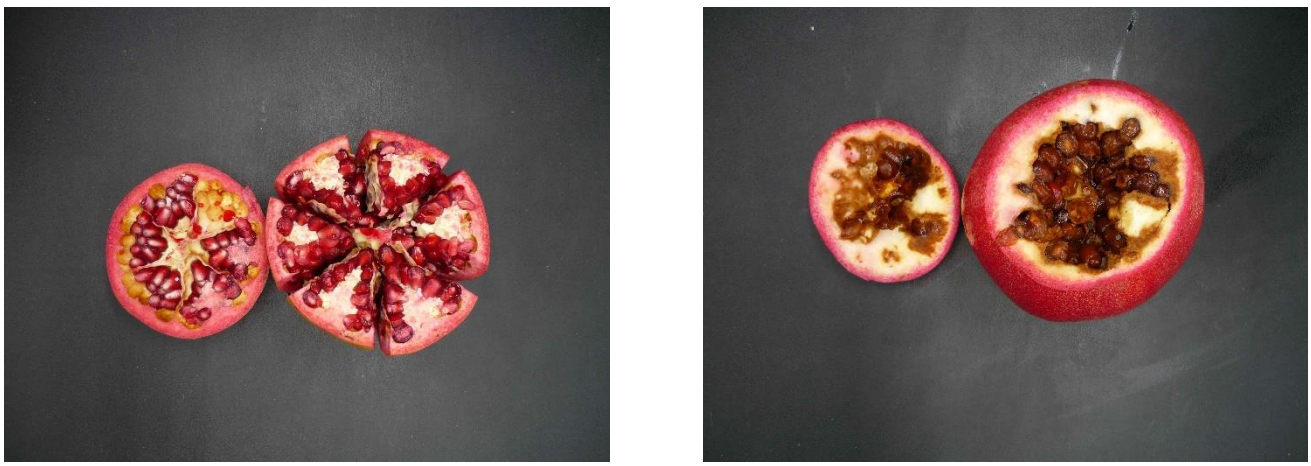

Figure 2: Pomegranates with albedo browning (left) and “Black heart” decay (right)

DISCUSSION

Population 1

After 6 weeks cold storage (Table 1a & 1b, Figure 1)

The severity of fruit with browning was assessed on a 0 to 3 rating with 0 = none, 1 = slight, 2 = moderate and 3 = severe and presented as the percentage of fruit in each rating category, which is indicative of chilling injury. After 6 weeks cold storage, no surface browning was exhibited on fruit from any treatments.

Wonderful pomegranates cold stored at 5.0 °C, irrespective of forced air or step-down cooling, exhibited higher levels of pulp membrane browning, compared to fruit stored at 7.5 °C. In the case of total pulp membrane browning, this difference was significant.

The rating 2 category albedo browning was significantly higher on the fruit cold stored at 5.0 °C (8.9%), irrespective of forced air or step-down cooling, compared to 0.0% for the fruit cold stored at 7.5 °C. Although not significant, a similar trend occurred in the rating 1 category fruit, as well as for the total albedo browning values.

No aril browning occurred in fruit from any of the treatments at the end of cold storage. Similarly, crown rot was not evident in fruit from any treatments. “Black heart” tended to be slightly higher for the pomegranates cold stored at 7.5 °C, compared to those cold stored at 5.0 °C. Surface decay was visible, but with no clear interpretation possible at this point in the study. Looking at total decay, there might be a higher risk of cold-storing pomegranates at 7.5 °C if the decay potential is high, but this temperature may reduce browning levels after cold storage.

After 6 weeks cold storage and a shelf-life at 20 °C for 7 days (Table 2a & 2b)

While not significant, the surface browning for ratings 2 and 3 was higher on pomegranates step-down cooled to 7.5 °C, compared to all the other treatments. However, the total surface browning was significantly higher on pomegranates step-down cooled to 7.5 °C, compared to all the other treatments.

Pulp membrane browning recorded in the rating 2 category was significantly higher on pomegranates forced-air cooled to 5.0 °C, compared to the other treatments. The total pulp membrane browning was significantly higher on pomegranates forced-air cooled to 5.0 °C, compared to fruit cold stored at 7.5 °C, irrespective of forced air or step-down cooling. Like after cold storage, pomegranates cold stored at 5.0 °C exhibited higher levels of pulp membrane browning, irrespective of whether forced air or step-down cooling was applied.

The total albedo browning was significantly higher on the pomegranates forded-air cooled and cold stored at 5.0 °C (41.1%), compared to 7.1% for the pomegranates step-cooled and cold stored at 7.5 °C, and the pomegranates forded-air cooled and cold stored at 7.5 °C.

No aril browning was exhibited in fruit from any of the treatments. Crown rot was at 1.8% for the pomegranates forded-air cooled and cold stored at 7.5 °C, compared to 0.0% for the other treatments. “Black heart” tended to be slightly higher for the pomegranates forced-air cooled and cold stored at 7.5 °C and step-down cooling to 5.0 °C, compared to the other treatments, but it is not clear if this is a direct treatment effect. The only surface decay was identified (1.8%) on pomegranates step-down cooled to 7.5 °C.

Population 2

After 6 weeks cold storage (Table 3a & 3b)

No surface, albedo or aril browning was exhibited for fruit from any of the treatments after the 6 weeks of cold storage. The rating 1 pulp membrane browning was slightly elevated at 1.8% for the pomegranates step-cooled and cold stored at 5.0 °C, compared to 0.0% for all the other treatments.

Crown rot was at 1.8% for the pomegranates forced-air cooled and cold stored at 7.5 °C, compared to 0.0% for the other treatments. “Black heart” tended to be slightly higher for the pomegranates cold stored at 5.0 °C (14.3%), followed by pomegranates forced-air cooled and cold stored at 5.0 °C (8.9%) and pomegranates step-cooled to 5.0 °C (7.1%). Pomegranates step-down cooled to 7.5 °C (1.8%) exhibited lowest “black heart”. Similarly, pomegranates step-down cooled to 7.5 °C exhibited lower levels of surface decay, compared to the other treatments.

After 6 weeks cold storage and a shelf-life at 20 °C for 7 days (Table 4a & 4b)

The surface browning in the rating 1 category fruit was highest across rating categories, with no significant differences between treatments. This disorder developed over shelf life. Pulp membrane browning was between 3.6% and 5.4% for the pomegranates forced-air cooled and cold stored at 7.5 °C and fruit step-down cooled to 5.0 °C, with the other two treatments at 0.0%. No albedo and aril browning were exhibited after shelf-life.

Crown rot was higher for both treatments with forced-air cooling to 5.0 °C and 7.5 °C, compared to 0.0% for pomegranates step-down cooled to 5.0 °C and 7.5 °C. “Black heart” decay was higher with pomegranates step down and forced-air cooled to 7.5 °C when compared to 5.0 °C, but despite these differences not being significant, this is a concern. The only surface decay (1.8%) was identified on pomegranates forced-air cooled to 7.5 °C. This population of pomegranates exhibited higher levels of total decay when cold stored at 7.5 °C, irrespective of step-down cooling or forced-air cooling. A large proportion of this decay was Black heart”.

8. CONCLUSIONS SINCE THE LAST REPORT

This was a first report. However, further research is suggested to confirm these findings and to try find a reliable way to balance control of browning without exacerbating unacceptable decay levels.

9. PLANNED OUTPUTS

a) TECHNOLOGY DEVELOPMENT, PRODUCTS AND PATENTS

This research can be transferred to the growers and pack-houses via articles or presentations as requested by POMASA and Hortgro.

b) STUDENTS

	Nationality	Study year	Race (B,C,I,W)	Gender (M,F,X)	Bursary cost 2024	Bursary cost 2025	Bursary cost 2026
Honours							
Masters							
PhD							
Postdoc							

DISCLOSING OF OTHER BURSARIES

Student name and surname	Other funder	Other cost

c) LIKELY PUBLICATIONS (POPULAR, PRESS RELEASES, SCIENTIFIC)

Articles as requested by POMASA

d) PRESENTATIONS/PAPERS THAT COULD BE DELIVERED

Presentations as requested by POMASA

10. PROJECT OUTCOME AND IMPACT

New knowledge	Benefits supply chain	Direct grower application	Direct packhouse application	Other
X	X	X	X	

Other is:

The value of the project to industry

Creates new knowledge for sea export of pomegranates.

11. TOTAL COST IN REAL TERMS

PROJECTED AMOUNTS FOR FUTURE YEARS	CFPA	RAISIN SA	HORTGRO	SATI	SA WINE	ARC	OTHER	TOTAL
2024			R 102 030.00					R 102 030.00
3025								
TOTAL								

RESPONSE TO PREVIOUS FEEDBACK