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# ANABP 01 Apple Storage Report 2020-2021

Kristen Brodison, Steele Jacob, Sophie Brockmann, John Sutton, Kevin Lacey and Yu-Yi Liao.

**Summary**

ANABP 01 is an apple variety bred by the Australian National Apple Breeding Program (ANABP) based at the Department of Primary Industries and Regional Development (DPIRD). ANABP 01 apples have an attractive dark burgundy colour and are crisp with sweet juicy white flesh that is slow to brown.

ANABP 01 resulted from the controlled cross-pollination of ‘Cripps Red’ and ‘Royal Gala’ in 1992 and was released to industry in 2014. Fruit West Co-operative Ltd is licenced to distribute ANABP 01 plant material to registered growers, to sell and distribute fruit that meets the defined quality specifications as Bravo™ branded apples in Australia.

An important aspect in any new apple variety bred by the ANABP is its ability to maintain fruit quality for extended periods of time to enable storage for ongoing retail supply. DPIRD have been investigating the capability of ANABP 01 to maintain fruit quality in storage since 2019. This report details further research into the storage capability of ANABP 01 undertaken in 2020-21.

In this study, storage treatments for ANABP 01 were selected based on information learnt from trials undertaken in 2019-2020. Building on the 2019-2020 trial results, ANABP 01 fruit was either harvested and then (1) stored in Controlled Atmosphere (CA); (2) treated with 1-Methylcyclopropene (1-MCP) and air stored in a cold room; (3) treated with 1-MCP and stored in CA; or (4) harvested later, treated with 1-MCP and CA stored.

Apples removed from storage environments underwent shelf life tests to investigate fruit quality characteristics post storage. For this study, all apples removed from storage were exposed to shelf life conditions and examined for fruit quality after 7 and 14 days.

Key outcomes from this study demonstrate the suitability of ANABP 01 fruit to store for long durations up to 42 weeks and the high capability to respond to the positive influence of 1-MCP on fruit storability and quality. A summary of key outcomes from the storage trial is presented in Table 1.

Of note in these results is the ability of late picked ANABP 01 fruit to store well for extended periods of time, up to 32 weeks, with 1-MCP application and CA storage. ANABP 01 fruit treated with 1-MCP was found to exhibit lower ethylene concentrations during shelf life tests at 7 and 14 days (than non 1-MCP treated fruit). Interestingly, after 40 weeks in storage, 1-MCP treated fruit increased total soluble solids content by exactly 10% confirming results from our storage trials in 2019-20.

| Fruit Treatment | Optimal fruit storage quality (duration) | Shelf life quality post storage (7 & 14 days) | Duration ethylene concentration is maintained in storage |
|---|---|---|---|
| **1-MCP treated and air stored** | 20 weeks | good quality for 20 weeks then variable | 24 weeks |
| **Untreated fruit CA stored** | 42 weeks | quality decreased from 24 weeks storage | 42 weeks |
| **1-MCP treated and CA stored** | 42 weeks | some variability, good firmness for 40 weeks | 42 weeks |
| **Late picked 1-MCP treated and CA stored** | 32 weeks+ | good quality for 32 weeks | 32 weeks |

**Table 1.** Summary of key outcomes from the ANABP 01 storage trials for 2020-21.

## Background

The ANABP aims to breed unique apple varieties with excellent eating and storage qualities, preferred by consumers and key export markets and well adapted to Australian growing conditions. The process of apple breeding is comprehensive and time consuming with it taking approximately 20 years to create and test a new variety prior to release. Thousands of crosses are made and repeatedly tested to find superior varieties prior to commercial release.

Of key importance in any new apple variety is the ability to maintain excellent fruit quality in storage to meet the market requirements of fruit availability for an extended time period. Investigations of apple storage ability through formal storage experiments commenced at DPIRD in 2019. The purchase of a gas analyser with the capability to measure ethylene provided additional information on the response of apples within the storage environments.

Previous ANABP 01 storage trials have demonstrated the ability of ANABP 01 to maintain excellent fruit quality for up to 40 weeks in storage. The 2019-20 storage trials showed ANABP 01 treated with 1-MCP and CA stored to maintain a consistent ethylene concentration and quality eating characteristics for up to 40 weeks (Brodison et al. 2020). ANABP 01 fruit harvested later to allow for further colour development also stored well for up to 36 weeks in CA storage after treatment with 1-MCP. ANABP 01 fruit not treated with 1-MCP showed a pronounced postharvest increase in ethylene.

Results produced from the 2019-20 storage trials enabled the development of storage protocol for 2020-21. The 2020-21 trial treatments include:

- Treatment with 1-MCP and air stored in a cold room;
- Untreated and stored in CA;
- Treatment with 1-MCP and stored in CA; or
- Harvested 20 days later (late pick), treated with 1-MCP and stored in CA.

Some of these treatments were new and others were undertaken to confirm previous work.

The defined quality specifications for premium grade ANABP 01 apples are paramount in the determination of the apple being branded as Bravo™. For 2021, ANABP 01 apples can be branded as Bravo™ when they meet the required specifications of:

- Flesh firmness: Average greater than 8.0 kg at harvest (minimum 7.5 kg);
- Sugar content: No more than 5% fruit less than 13% Brix;
- Eating characteristic: free from foreign taints (crisp, sweet, firm and juicy);
- Fruit appearance and shape: no misshapes / malformed fruit, milky coating or surface residue;
- Skin colour: At least 60% of the surface area to be black – dark burgundy colour;
- Skin ground colour: no more than 10% of the surface area to be uncoloured (Fruit West Co-operative Ltd., 2021).

All ANABP 01 apples branded as Bravo™ are required to be treated with 1-MCP within 7 days after picking. The majority of treatments in this trial used 1-MCP however one treatment did not use 1-MCP for comparison purposes.

## Methodology

### *Storage conditions*

Based on previous storage trials, ANABP 01 fruit was stored in a variety of cold storage environments for up to 42 weeks postharvest. The duration of each storage treatment varied depending on the condition of the fruit and storage environment. The storage environments and conditions used in this study included:

- Fruit treated with 1-MCP and Air Stored in Cold Room:
  - 1°C +/- 0.3°C, 85-95% humidity.
  - Fruit was harvested on the 14/04/2020, evaluated at 12 weeks then every 4 weeks until the 27/10/2020 (a total of 28 weeks).
  - Fruit was measured at removal from storage and at 7 days and 14 days of shelf life simulation.
  - Evaluations concluded when the fruit texture began to soften and the skin felt rubbery and displayed a withered appearance.

- Untreated fruit stored in Controlled Atmosphere (CA)
  - 1°C +/- 0.3°C, 85 -95% humidity, 1.5 to 2.5% oxygen, < 1% carbon dioxide.
  - Fruit was harvested on the 14/04/2020, evaluated at 12 weeks then every 2 to 4 weeks until the 02/02/2021 (42 weeks).
  - Fruit was measured at removal from storage and at 7 days and 14 days of shelf life simulation.
  - Evaluations concluded when all stored fruit had been utilised for testing and some of the fruit texture had softened below acceptable levels.

- Fruit treated with 1-MCP and stored in CA
  - 1°C +/- 0.3°C, 85 -95% humidity, 1.5 to 2.5% oxygen, < 1% carbon dioxide.
  - Fruit was harvested on the 14/04/2020, evaluated at 12 weeks then every 2 to 4 weeks until the 02/02/2021 (42 weeks).
  - Fruit was measured at removal from storage and at 7 days and 14 days of shelf life simulation.
  - Evaluations concluded when all stored fruit had been utilised for testing with minor detection of fruit softening below acceptable levels.

- Fruit picked at a later maturity (late picked), treated with 1-MCP and stored in CA
  - 1°C +/- 0.3°C, 85 -95% humidity, 1.5 to 2.5% oxygen, < 1% carbon dioxide.

- Fruit was harvested 20 days later than the normal harvest timing on the 04/05/2020, evaluated at 12 weeks then every 4 weeks until the 16/12/2020 (32 weeks).
- Fruit was measured at removal from storage and at 7 days and 14 days of shelf life simulation.
- Evaluations concluded when all stored fruit had been utilised for testing.

All fruit used in this study was produced, stored and tested at DPIRD's Manjimup Horticulture Research Facility. Where used, 1-MCP was applied to fruit as soon as possible after harvest (within 48 hours).

### *Apple quality tests*

Tests to determine the quality of ANABP 01 apples were undertaken at harvest and at key intervals during the storage period. Every 4 weeks for the first 32 weeks, then at 2 weekly intervals. The two weekly interval testing was adopted after 32 weeks in order to identify the critical timing when ANABP 01 quality had deteriorated in storage.

All quality measurements were undertaken as per the 'ANABP 01 Apple Maturity Testing' guide (Jacob and Murphy White 2020) on ten single apples at each sampling occasion. Measurements included apple firmness (kg-f), total soluble solids (% Brix), starch pattern index (1 to 6 scale), acid (%), fruit weight (g) and diameter (mm).

Ethylene concentration was measured on each single apple with a Felix F-950, as per the methodology used in previous ANABP 01 storage trials (Brodison et al. 2020). Apples were sealed within clear plastic containers (1.2 L) fitted with two Vacutainer® rubber stoppers to prevent the diffusion of ethylene. The Felix F-950 was set up to draw and analyse the headspace gas sample via an input and output probe.

For measurement of ethylene from apples in storage, apples were removed from the cold storage environments to allow to adjust to room temperature (22°C) for 24 hours prior to ethylene being recorded. All factors that could contribute to the measurement of ethylene were recorded including the duration of the apple within the container (hours), container volume (litres) and size of the apple (grams). Ethylene calculations were adjusted for all these factors and where these figures show a positive correlation with ethylene (ppm), are presented here as the ethylene concentration (ppm).

## Results and discussion

*Harvest fruit evaluation*

At harvest, the ANABP 01 fruit achieved the optimal fruit maturity specifications required to be marketed as Bravo™. The fruit Brix (15.5%) and firmness (9.6 kg-f) exceeded the required quality specifications (Figure 2). The fruit also had an average starch level of 4.7 and acidity of 0.79%.

The late picking of ANABP 01 fruit had limited impact on the fruit quality measurements although from visual observations was found to increase the overall dark burgundy colour of the fruit (Figure 1.). The starch (4.9) and Brix (15.9%) levels were all within 5% of the earlier harvested fruit (Figure 2). However, the firmness was slightly lower (8.14 kg-f) but remained within the quality specification range for Bravo™. It's important to note that late picked fruit needed to meet required variety specifications.

**Figure 1.** Photo of ANABP 01 fruit taken during fruit maturity testing showing the colour of fruit at (A) commercial harvest time on 14/4/20 and (B) the late pick on 4/5/20.

Interestingly, the ethylene concentration of late picked fruit was lower than that of fruit picked at the normal harvest date. The late picked fruit had an ethylene concentration of 8.28 ppm whilst the normal harvest date fruit was 9 ppm (Figure 2). This is within 10% of the ethylene concentration at harvest and was not determined to be significantly different ($P < 0.05$). The decrease in ethylene during the ripening period also fits with the generalised pattern of ethylene production during the ripening of apples (Paul et al, 2011) and may indicate fruit in senescence.

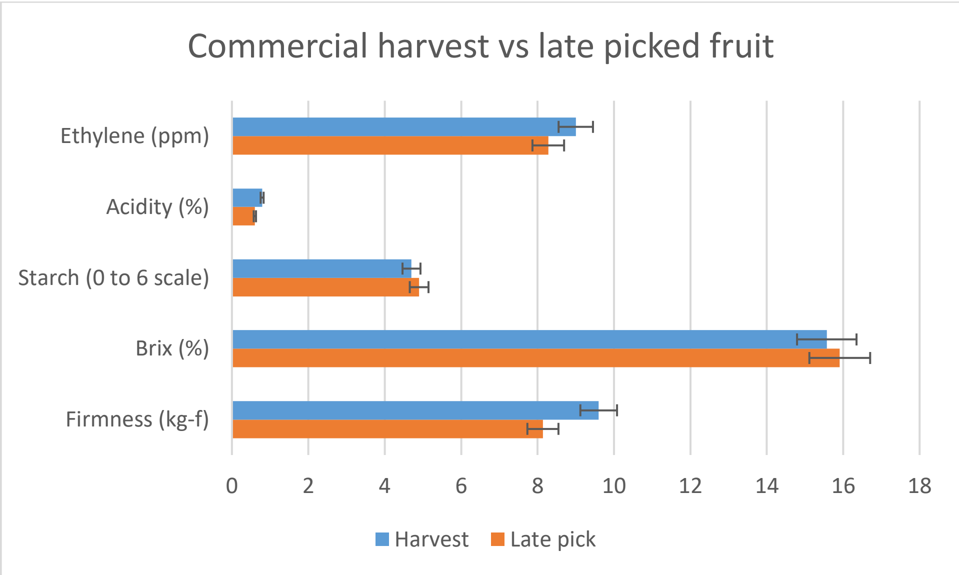


**Figure 2.** Maturity of ANABP 01 at commercial harvest and picked 20 days later ('late pick') including ethylene (ppm), acidity (%), starch (0 to 6 scale), Brix (%) and firmness (kg-f). Error bars indicate percentage difference (5%).

*Fruit treated with 1-MCP and air stored in cold room*

A low and consistent ethylene concentration was maintained in 1-MCP treated ANABP 01 fruit for 24 weeks in air store (Figure 3). The median ethylene concentration in fruit treated with 1-MCP and air stored for 24 weeks was 8.02 ppm (+/- 0.98). A noticeable increase in ethylene concentration (14.7 ppm) was detected in fruit at 28 weeks postharvest. This rapid increase in ethylene concentration was coupled with the onset of deterioration in fruit quality indicating the decrease in commercial viability of the fruit.

Shelf life simulation tests of ANABP 01 fruit, undertaken at both 7 and 14 days post storage, show ethylene to increase. Initially the rise in ethylene was low at removal after 12 weeks, increasing by 0.6 ppm at 7 days shelf life and 2.55 ppm at 14 days shelf life. The escalation in ethylene continued and peaked at 34 ppm in shelf life tests of fruit removed from storage at 28 weeks and kept on the shelf for a further 7 days.

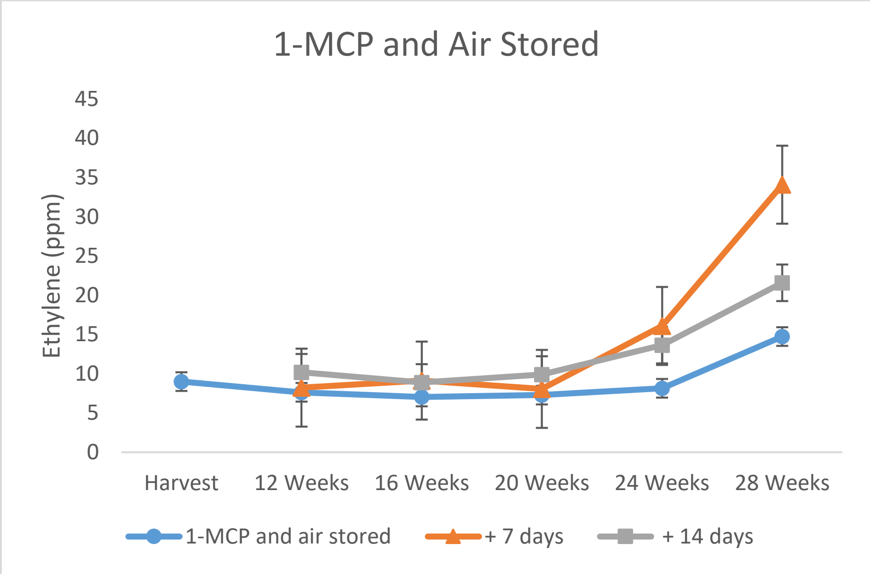


**Figure 3.** Ethylene concentration from ANABP 01 fruit treated with 1-MCP and air stored for 28 weeks. Ethylene was measured from fruit at removal and at both 7 days and 14 days shelf life. Error bars indicate standard errors.

The fruit quality of 1-MCP treated apples stored in the cold room was maintained at premium grade specification for up to 20 weeks in storage. During this period, the fruit Brix and firmness either met or exceeded the specifications of 8 kg-f firmness and 13% Brix (Figure 4). The starch pattern index (0 to 6 scale) also reached a maximum starch degradation mark 6 at 20 weeks storage. After 20 weeks in storage the fruit quality became variable with some fruit ranking below premium specifications.

At the conclusion of this storage trial (28 weeks) fruit undergoing shelf life evaluation (for 7 and 14 days) was of a lower quality than fruit straight out of storage. At 7 days shelf life a small reduction in Brix (13.8 %) and firmness (7.79 kg-f) were recorded (Figure 4). These figures continued to decline at 14 days shelf life with further reduction in Brix (12.7%) and firmness (7.53 kg-f), probably due to increased respiration and metabolism resulting in sugar degradation.

The quality of ANABP 01 fruit under similar conditions was maintained for a longer period in the experiments in 2019-20 than in 2020-21. Evaluations in 2019-20 showed fruit to produce lower ethylene concentrations (average 5ppm) for longer durations (up to 40 weeks) than 2020-21 (Brodison et al. 2020). Fruit in 2019-20 trials also had a lower sugar level (12.3% Brix) at harvest than fruit in 2020-21 (15.57%). The higher level of ethylene and Brix in fruit at harvest in 2020-21 could imply that fruit were harvested at a more advanced physiological maturity stage and therefore, it may have contributed to the high ethylene concentrations and reduced ability of the fruit to store as long.

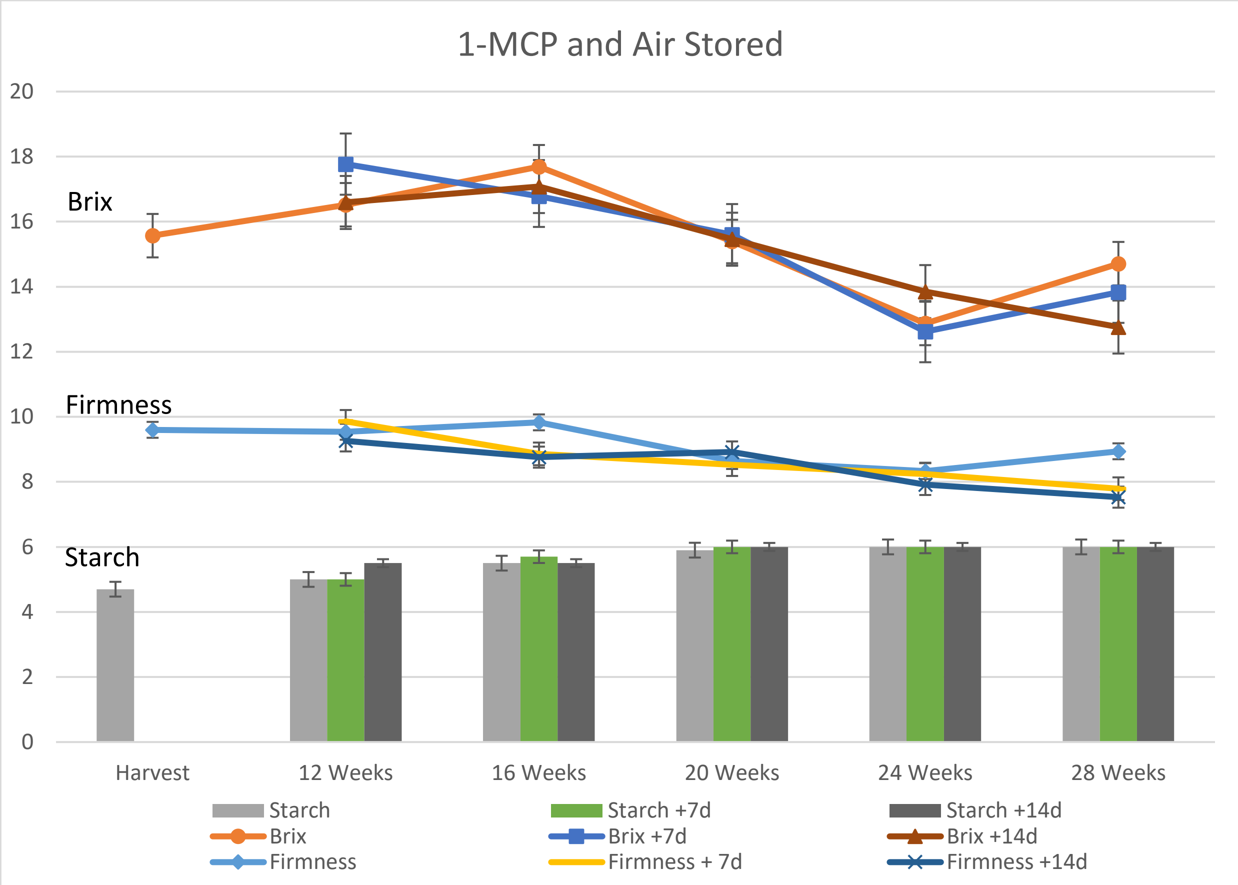


**Figure 4.** Maturity indices of ANABP 01 fruit treated with 1-MCP and air stored for 28 weeks including firmness (kg-f), total soluble solids (% Brix), starch pattern index (1 to 6 scale). Error bars indicate standard errors.

*Untreated fruit stored in CA storage*

A low and consistent ethylene concentration was also maintained in untreated ANABP 01 fruit CA stored for up to 42 weeks. The ethylene concentration commenced at 9 ppm at harvest and was 8.19 ppm at 42 weeks in CA storage (Figure 5). Overall, the ethylene concentration averaged 9.36 ppm whilst in storage.

The ethylene concentration increased in shelf life tests of untreated ANABP 01 fruit. In comparison to fruit tested at removal from storage (average 9.36 ppm ethylene), fruit tested after being on the shelf for 7 days and 14 days showed an increased ethylene concentration (16.1 ppm and 18.1 ppm respectively) (Figure 5). This was expected since fruit would restart full respiration rates during shelf life, therefore resuming rates of ripening and consequently ethylene production.

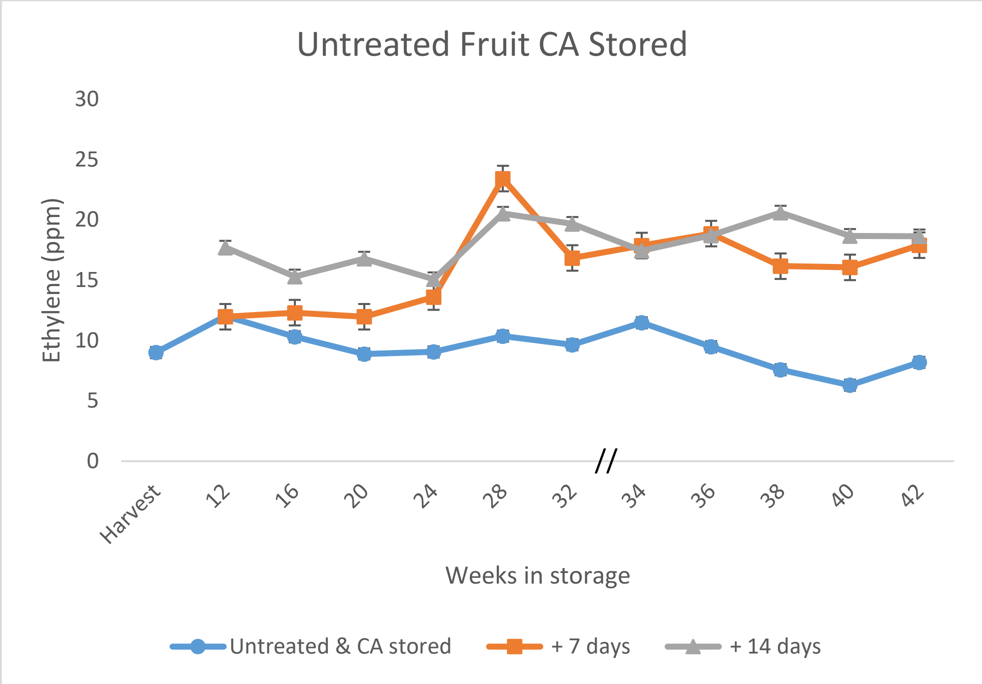


**Figure 5.** Ethylene concentration from untreated ANABP 01 fruit CA stored for 42 weeks. Ethylene was measured from fruit at removal and at 7 days and 14 days shelf life. Error bars indicate standard errors.

ANABP 01 fruit which was untreated and CA stored showed variable fruit quality on removal from the storage environment and during shelf life tests. For the duration of CA storage the Brix remained above the minimum specifications required for premium grade (13%) detected in fruit immediately out of storage and in shelf life tests for 7 and 14 days (Figure 6). During the storage period, the Brix gradually increased to an average high of 16.9% at 42 weeks post storage for all treatments.

Optimal fruit firmness was retained in fruit straight out of storage which remained above the required firmness of 8 kg-f for the duration of the trial. However, fruit firmness was found to decrease after 36 weeks storage in fruit undergoing shelf life tests for 7 days and after 24 weeks for fruit undergoing shelf life tests for 14 days (Figure 6).

The starch pattern index reached its minimum content (starch degradation mark 6) in fruit tested on removal from the storage environment and in shelf life tests after 20 weeks in CA storage (Figure 6).

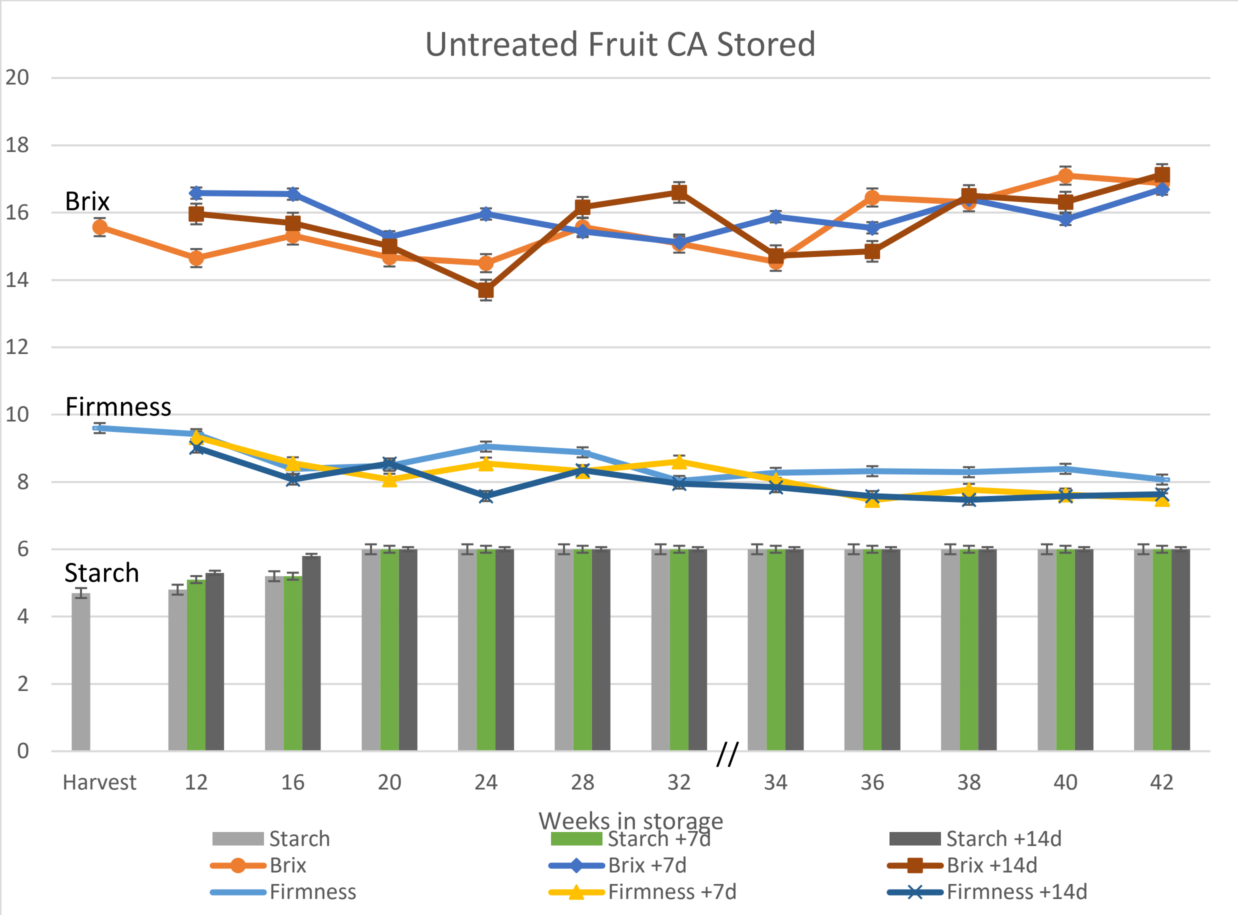


**Figure 6.** Maturity indices of untreated ANABP 01 fruit, CA stored for 42 weeks including firmness (kg-f), total soluble solids (% Brix) and starch pattern index (1 to 6 scale). Error bars indicate standard errors.

*Fruit treated with 1-MCP and CA stored*

Ethylene concentration was maintained at a low and consistent level in fruit treated with 1-MCP and CA stored for 42 weeks. From fruit exiting storage, ethylene concentration was found to decrease from concentrations recorded at harvest (8.65 ppm) to a low of 5.29 ppm at 42 weeks storage (Figure 7), as expected due to the inhibiting effects of 1-MCP and fruit not having resumed ripening yet. Overall, the average ethylene concentration of fruit at removal was 6.2 ppm.

The ethylene concentration of fruit from shelf life experiments was maintained low due to the influence of 1-MCP. Fruit retained on the shelf for 7 days modelled similar ethylene concentrations (average 6.8 ppm) to fruit directly removed from storage up until 42 weeks where levels dramatically increased to 12 ppm (Figure 7). However, the 14 day shelf life fruit had ethylene concentrations consistently higher (average 11 ppm) than the fruit at removal from storage due to the restarting of the ripening process.

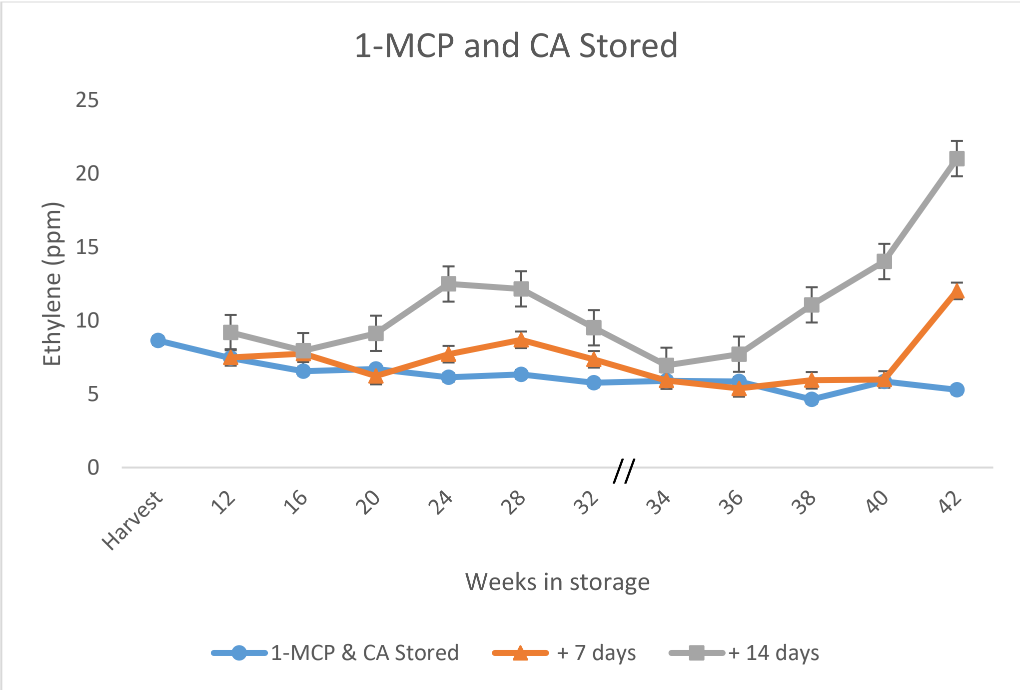


**Figure 7.** Ethylene concentration from 1-MCP treated ANABP 01 fruit CA stored for 42 weeks. Ethylene was measured from fruit at removal and at 7 days and 14 days shelf life. Error bars indicate standard errors.

The quality characteristics of 1-MCP treated fruit on removal from CA storage was high. Both the fruit Brix (average 14.4%) and firmness (average 8.9 kg-f) remained above the premium quality Bravo™ specifications for the entire 42 weeks of storage (Figure 8). However, the quality of fruit from the 7 and 14-day shelf life tests was variable. The sugar content and firmness of fruit from 7-day shelf life test was irregular however within required quality specifications. The sugar content of fruit from 14-days shelf life examinations fluctuated to a low of 12.4% Brix at 28 weeks and a high of 17.05% Brix at 36 weeks (Figure 8). The starch pattern index reached its minimum content (starch degradation mark 6) for all fruit at 16 to 20 weeks. Overall, the observed Brix variability during the various experiments and storage conditions, is probably due more to the fruit variability within the canopy in the field than the effect of storage itself, since normally sugars tend to increase during storage from starch and acid degradation.

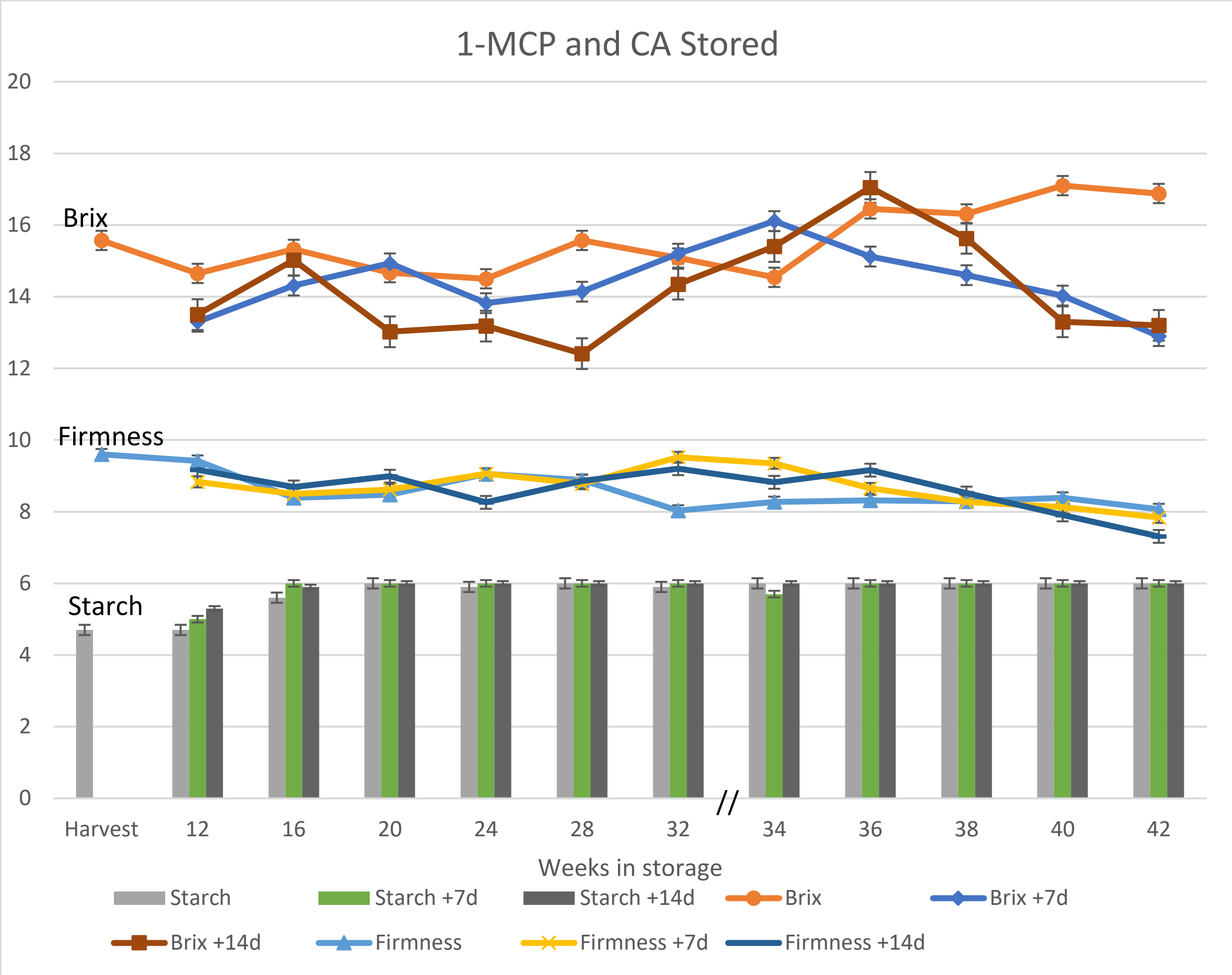


**Figure 8.** Maturity indices of ANABP 01 fruit, treated with 1-MCP and CA stored for 42 weeks including firmness (kg-f), total soluble solids (% Brix), starch pattern index (1 to 6 scale). Error bars indicate standard errors.

The storage results for 1-MCP and CA stored fruit showed similar trends in both years of testing even with different levels of maturity. For both 2019-20 and 2020-21 the ethylene levels were maintained low and consistent in 1-MCP treated and CA stored fruit for 40 weeks (Brodison et al. 2020). The fruit firmness was also maintained above 8 kg-f. Remarkably from harvest to 40 weeks in storage the total soluble solids content of fruit increased by exactly 10% in both years (Table 2).

**Table 2.** Total soluble solids (% Brix) comparison in 1-MCP treated and CA stored ANABP 01 fruit at harvest and at 40 weeks storage.

| | **2019-20** | **2020-21** |
|---|---|---|
| Harvest | 12.38% | 15.57% |
| 40 weeks storage | 13.62% | 17.10% |
| % difference | +10% | +10% |

*Late picked fruit, treated with 1-MCP and CA stored*

A low and consistent ethylene concentration was maintained for the entire 32 week storage period of late picked ANABP 01 fruit treated with 1-MCP. The overall fruit ethylene concentration for this period was 6.56 ppm. At harvest the fruit ethylene concentration was 8.28 ppm and gradually decreased until it reached 5.88 ppm at 32 weeks postharvest (Figure 9).

Shelf life tests presented minor increases in ethylene content in fruit left on the shelf for 14 days. Whilst the ethylene content of fruit left on the shelf for 7 days mimicked that of fruit directly removed from storage, fruit left on the shelf for 14 days increased in ethylene when removed from storage at 20 weeks (7.23 ppm) through to 32 weeks (8.4 ppm) recovering from the 1-MCP effects and going back to harvest levels (Figure 9). However, the increase in ethylene at 14 days storage was only minor and ranged from a low increase of 1.05 ppm (at 20 weeks storage) to a high increase of 2.52 ppm (at 32 weeks storage).

The evaluation of late picked fruit concluded when all stored fruit had been utilised for testing.

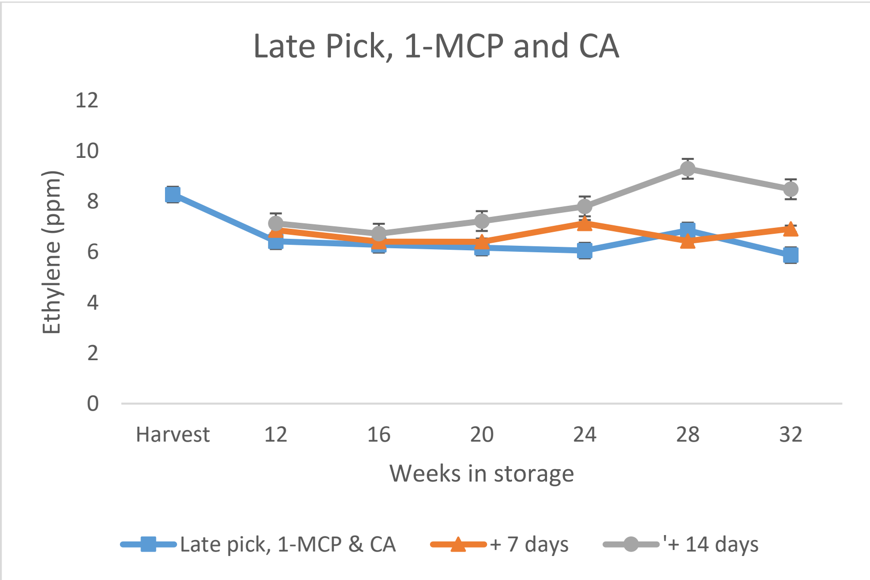


**Figure 9.** Ethylene concentration from late picked, 1-MCP treated ANABP 01 fruit CA stored for 32 weeks. Ethylene was measured from fruit at removal and then at 7 days and 14 days shelf life. Error bars indicate standard errors.

All late picked ANABP 01 fruit, treated with 1-MCP and CA stored maintained premium grade fruit quality during the 32 weeks of storage. All treatments, including the 7-day and 14-day shelf life tests maintained optimal sugar content (ranging from 13.4 to 16.1 % Brix) and firmness (all above 8 kg-f) (Figure 10). The starch pattern index reached its minimum content (starch degradation mark 6) for all fruit at 16 weeks storage. These results are consistent with those recorded in storage trials in 2019-20 (Brodison et al. 2020).

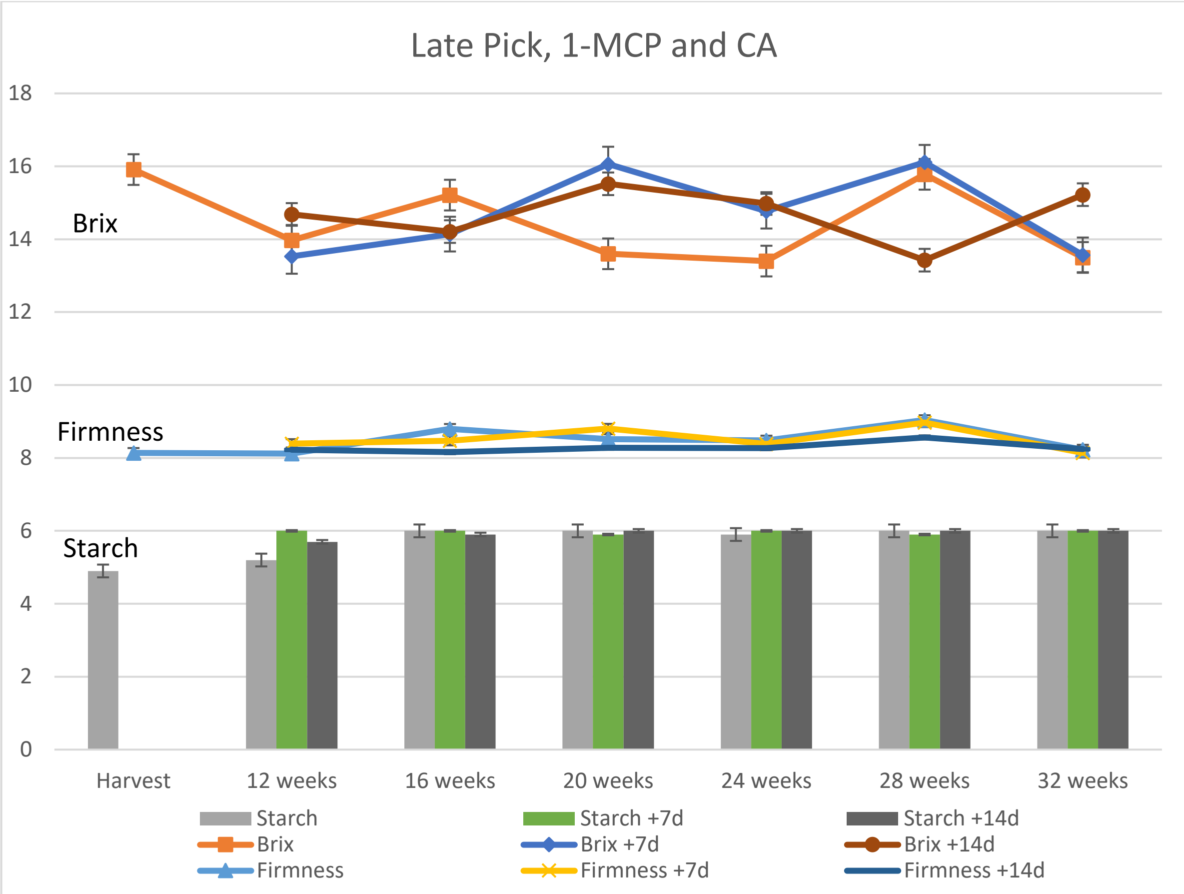


**Figure 10.** Maturity indices of late picked ANABP 01 fruit, treated with 1-MCP and CA stored for 32 weeks including firmness (kg-f), total soluble solids (% Brix), starch pattern index (1 to 6 scale). Error bars indicate standard errors.

## Acknowledgements

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## References

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## Important disclaimer