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# Maturity indices for optimum harvest time of apple fruit cv. Monalisa

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**Abstract:** This study was carried out to evaluate the changes in maturity and quality of 'Monalisa' apple fruit at harvest and after storage and to determine maturity indices for the optimum harvest time. Experimental treatments were harvesting time, storage atmosphere and duration and 1-MCP exposure. Fruit from multiple harvest date and three harvest years were stored at 0.8 °C in air or controlled atmosphere for 3, 5, 6 or 9 months. Half of the fruit were treated with 1-MCP in two years. The increase in ethylene production, respiration, starch degradation and soluble solids content, and the decline in flesh firmness and titratable acidity during on-tree maturation followed the expected pattern of early season cultivars such as Gala, the 'Monalisa' progenitor. After storage, late harvested fruit had higher severity of decay, and physiological disorders compared to early harvested fruit. Skin browning was the predominant disorder in 'Monalisa', which was affected by harvest maturity, 1-MCP treatment, storage atmosphere and duration. The results showed that 'Monalisa' apple intended for immediate marketing should be harvested between 131 to 149 days after full bloom, with starch index ranging from 3.3 to 7.5 (1-9 scale), flesh firmness from 87.1 to 69.3 N, soluble solids content from 12.7 to 14.7 %, and titratable acidity from 0.66 to 0.56 %. 'Monalisa' apple intended for mid- and long-term storage should be harvested earlier between 124 to 131 days after full bloom, with starch index ranging from 2.4 to 3.4, flesh firmness from 90.7 to 86.2 N, soluble solids content from 12.7 to 14.3 %, and titratable acidity ranging from 0.67 to 0.59 %.

**Index Terms**: *Malus domestica*, starch index, flesh firmness, soluble solids, acidity, physiological disorders.

# Índices de maturação para o ponto ideal de colheita de frutos de macieira cv. Monalisa

**Resumo:** Este estudo foi realizado para avaliar as mudanças na maturação e na qualidade da maçã 'Monalisa' na colheita e após armazenagem, e para determinar os índices de maturação para o ponto ideal de colheita. Tratamentos experimentais foram datas de colheita, atmosfera e tempo de armazenagem e tratamento 1-MCP. Frutas de diferentes datas de colheita e de três anos de

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produção foram armazenadas a 0,8 °C sob atmosfera do ar ou atmosfera controlada por 3; 5; 6 ou 9 meses. Metade das frutas foi tratada com 1-MCP em dois anos. O aumento na produção de etileno, respiração, degradação do amido e teor de sólidos solúveis, e o declínio na firmeza da polpa e acidez titulável, durante a maturação na planta, seguiram o padrão esperado de cultivares precoces, como Gala, o progenitor da 'Monalisa'. Após a armazenagem, as frutas colhidas tardiamente apresentaram maior severidade de podridões e de distúrbios fisiológicos em comparação às frutas colhidas precocemente. O escurecimento da epiderme foi o distúrbio predominante em 'Monalisa', sendo afetado pela maturidade na colheita, tratamento com 1-MCP, atmosfera de armazenagem e duração. Os resultados mostraram que a maçã 'Monalisa', destinada à comercialização imediata, deve ser colhida entre 131 e 149 dias após a plena floração, com índice de amido variando de 3,3 a 7,5 (escala de 1 a 9), firmeza de polpa de 87,1 N a 69,3 N, teor de sólidos solúveis de 12,7 % a 14,7 % e acidez titulável de 0,66 % a 0,56 %. A maçã 'Monalisa' destinada ao armazenamento, por médio e longo períodos, deve ser colhida mais cedo, entre 124 e 131 dias após a plena floração, com índice de amido variando de 2,4 a 3,4, firmeza de polpa de 90,7 N a 86,2 N, teor de sólidos solúveis de 12,7 a 14,3 % e acidez titulável variando de 0,67 a 0,59 %.

**Termos para indexação:** *Malus domestica*, índice de amido, firmeza de polpa, sólidos solúveis, acidez, desordens fisiológicas.

# Introduction

The quality and duration of apple fruit's shelf- and storage life rely on harvest maturity. Fruit harvested at more advanced maturity have enhanced red coloration, flavor and are larger, particularly at harvest (PLOTTO et al., 1997; FELLMAN et al., 2000; DELONG et al., 2014; MAGRIN et al., 2017). However, mature fruit are more susceptible to decay, greasiness, senescent related disorders (CAMELDI et al., 2016; DELONG et al., 2016; BETINELLI et al., 2017), low-temperature breakdown, chilling injury (PRANGE et al., 2011; DELONG et al., 2014; DOERFLINGER et al., 2024), $CO_2$ injury (ARGENTA et al., 2002) and shrivel (ARGENTA ; MONDARDO, 1994; MAGUIRE et al., 2000) after storage. On the other hand, fruit harvested at early maturity stages are more susceptible to physiological disorders such as bitter pit and superficial scald (WATKINS et al., 2005; PRANGE et al., 2011; LURIE; WATKINS, 2012; DELONG et al., 2014).

The compromise between quality attributes at harvest and after storage can be achieved by harvesting apples at a physiological maturity that precedes the rise in respiration rate and ethylene production characteristic of climacteric fruit (KNEE et al., 1989; WATKINS, 2003). However, physiological maturity is characterized by a wide range of maturity and quality attributes including, starch index, flesh firmness, soluble solid content (SSC), titratable acidity (TA) and skin background color (KINGSTON, 1992). These quality parameters are practical objective measures of maturity (maturity indices) that apple growers can use to determine the appropriate harvest time in each season and orchard plot (KNEE; FARMAN, 1989; KINGSTON, 1992; TOIVONEN, 2007). Therefore, the precise maturity indices for optimum harvest time must be determined for each cultivar and growing condition by assessing the relationship between harvest maturity and quality after storage (KINGSTON, 1992; ARGENTA; MONDARDO, 1994; TOIVONEN, 2007; PRANGE et al., 2011; DELONG et al., 2016). A small percentage of apples are shipped to the market briefly after harvest, whereas most are stored to keep fruit available for an extended pe-

riod (FELLMAN et al., 2000). Therefore, optimum harvest maturity must be determined for each marketing stage to maximize each apple cultivar's quality and consumer acceptance.

Although the optimum harvest maturity has been well determined for most commercial apple varieties, information is still required for new varieties recently launched on the market. 'Monalisa' is a new apple cultivar, obtained from the crossing between 'Gala' and 'Malus 4', which has attracted the attention of apple growers due to its high fresh eating and appearance qualities, and its high resistance to major diseases, such as Apple Scab and Glomerella Leaf Spot (DENARDI et al., 2013). The genetic resistance to these diseases is necessary for profitable commercial production of non-organic and organic apples under subtropical humid climate conditions, such as in Southern Brazil. Although 'Monalisa' apple have important quality traits for commercial fruit production, no information about the optimum maturity indices to harvest fruit intended for short- and long-term storage is available.

The objectives of this study were to evaluate the quality as a function of harvest maturity and determine the maturity indices for the optimal harvest time of 'Monalisa' apple produced in Southern Brazil for short and long marketing times.

# Material and Methods

## Orchard and plant material

'Monalisa' apple trees on Marubakaido rootstock and M.9 interstem were cultivated in two orchards at 1.4×4 m spacing, planted in 2006 (Orchard 1), and 2012 (Orchard 2) at the Agricultural Research and Rural Extension Company of Santa Catarina, EPAGRI, Caçador, SC, Brazil (26°50'8.42" S, 50°58'26.79"W). The study was composed of three experiments. Experiment 1 was carried out in 2011 in the orchard one, whereas Experiments 2 and 3 were carried out in 2017 and 2019 in the orchard two. Fruit of similar size that were representative of the whole tree in each harvest year and harvest date were sampled from 150 trees in each experiment. Fruit were harvested from the inner and outer canopy of both sides of the tree row at mid-canopy height. In the laboratory, visually unblemished fruit were randomly selected to prepare homogeneous samples of 20 fruit held on fiberboard trays.

## Storage

After harvest, fruit were moved into a cold room and were cooled to 0.8 °C within 36 h. The storage temperature was 0.8 ± 0.8 °C for both Air (~21 kPa $O_2$) and controlled atmosphere (CA) conditions. Fruit subjected to Air storage were packed in carton boxes (18 kg) that were internally lined with perforated low-density polyethylene bags (20 µm = 10 µm per wall) to prevent fruit shriveling. The relative humidity (RH) inside the bag was likely close to saturation. Fruit for CA storage were enclosed in 0.150 $m^3$ stainless steel chambers with a plexiglass lid. RH in CA chambers ranged between 92 to 95 %. Temperature and RH were monitored as described by (ARGENTA et al., 2023b). CA with low $O_2$ (1.5 kPa) and $CO_2$ (<0.5 kPa) partial pressures was established within 54 h after fruit cooling. Concentrations of $O_2$ and $CO_2$ were monitored and maintained as described by (ARGENTA et al., 2023b). The CA with low $pCO_2$ was used to avoid $CO_2$ injury in the fruit, as recommended by other studies (THEWES et al., 2023). Shelf life simulation was in Air atmosphere at 22 ± 1 °C.

## Experiment 1

In 2011, fruit were harvested periodically at 4 to 7-day intervals between January 24th and February 21st, corresponding to 120 and 149 days after full bloom (DAFB). Fruit were assessed 24 hours after harvest and after six months of storage in CA plus 1- and 7-days shelf life. For each combina-

tion of harvest time (6) and shelf life period (2), there were six replicates ($n = 6$) of 8-fruit batches analyzed for respiration, ethylene, soluble solid concentration (SSC) and titratable acidity (TA), and 60 individual fruit ($n = 60$) were analyzed for starch index, flesh firmness and disorders (Section 2.6) in a complete randomized design.

## Experiment 2

In 2017, fruit were harvested on January 30th, February 8th and 17th, corresponding to 119, 128 and 137 DAFB. Fruits were assessed 24 hours after harvest and after three and six months of storage in air (21 kPa $O_2$) plus 7 days shelf life. Half of the stored fruit were exposed to 1 µL $L^{-1}$ of 1-MCP within 24 h of harvest in a sealed steel container (1 $m^3$) for 12 h at ambient temperature. The 1-MCP gas was generated by mixing cyclodextrin-1-MCP powder (EthylBloc™, AgroFresh Inc. Spring House, USA) and water. The 1-MCP concentration inside the treatment container was monitored as previously described (MATTHEIS et al., 2005). For each combination of harvest time (3), 1-MCP treatment (2), and storage period (2) there were three replicates ($n = 3$) of 8-fruit batches for SSC and TA analysis and 60 individual fruit ($n = 60$) for starch index, flesh firmness and disorders in a complete randomized design.

## Experiment 3

In 2019, fruit were harvested on February 1st, 11th, and 18th, corresponding to 126, 136 and 143 DAFB. Fruits were assessed 24 hours after harvest and after five and nine months of storage in air or CA plus 7 days shelf life. Half of the stored fruit were exposed to 1 µL $L^{-1}$ of 1-MCP within 24 h of harvest, as described above. There were three replicates ($n = 3$) of 8-fruit batches for SSC and TA analysis and 40 individual fruit ($n = 40$) for the other variables, for each combination of harvest time (3), 1-MCP treatment (2), storage atmosphere (2), and storage period (2) in a complete randomized design.

## Maturity and quality analyses

Fruit analyses were performed one day after harvest and after storage and shelf-life conditions. Fruit fresh weight, background and red skin color, flesh firmness, starch index (1-9 scale), SSC, TA, and SSC/TA ratio were assessed as described by (ARGENTA et al., 2023b). Ethylene production and respiration rate were assessed as described by (MATTHEIS et al., 2005). External and internal disorders were visually assessed using subjective scales of severity, where a score of one indicates the absence of disorders. Internal disorders were assessed from four transverse slices across the fruit. The severity of disorders was recorded according to the area of fruit surface or cortex cross-section affected or the number of lesions per fruit. Assessment of fungal decay, skin browning (scald-like browning), shriveling, bitter pit, flesh browning, fruit cracking and senescent breakdown were accomplished as previously described by (ARGENTA et al., 2023a; ARGENTA et al., 2023b). Fruit affected by wrinkly skin was scored as 1, absence; 2, 1–30 % of the fruit surface with light wrinkle; 3, 31–60 % of the fruit surface with light to deep wrinkle; or 4, >60 % of the skin with light to deep wrinkle.

## Statistical analyses

Data from Experiment 1 were subjected to regression analysis using the Equation Dynamic Fit Wizard of SigmaPlot software version 14 (Systat Software Inc., San Jose, USA). Statistical models for each treatment and variable were initially selected based on the Akaike information criterion (AIC) and later by the determination coefficient and regression residuals. Means were compared by Fisher's least significant difference LSD test ($\alpha = 0.05$). In Experiments 2 and 3, flesh firmness, starch index, SSC and TA data were subjected to the analysis of variance (ANOVA). Means were compared by Tukey's HSD test ($p<0.05$). Physiological disorders and decay incidence data did not

show normal distribution and homogeneity of variances, which were then subjected to non-parametric Kruskal-Wallis test (α = 0.05). All statistical analyses were performed using R (R-CORE-TEAM, 2021) with the add-on package 'Agricolae' (MENDIBURU, 2020).

# Result and Discussion

## *Experiment 1*

The 'Monalisa' apple fruit size increased during on-tree maturation, especially between 120 and 136 DAPF (Figure 1). However, the percentage of red coloration on the surface increased throughout a longer period, between 120 and 149 DAPF (Figure 1). The on-tree maturation of the 'Monalisa' apple was characterized by the increase in ethylene production, respiration, starch degradation (decrease in starch content) and SSC, the decline in flesh firmness and acidity, and a change of background color from green to yellow (Figures 1 and 2). These results are a typical pattern of maturation as described for other cultivars such as Gala (ARGENTA et al., 2018b), 'Monalisa' progenitor, and 'Fuji' (ARGENTA et al., 2022), the most planted cultivars in Brazil.

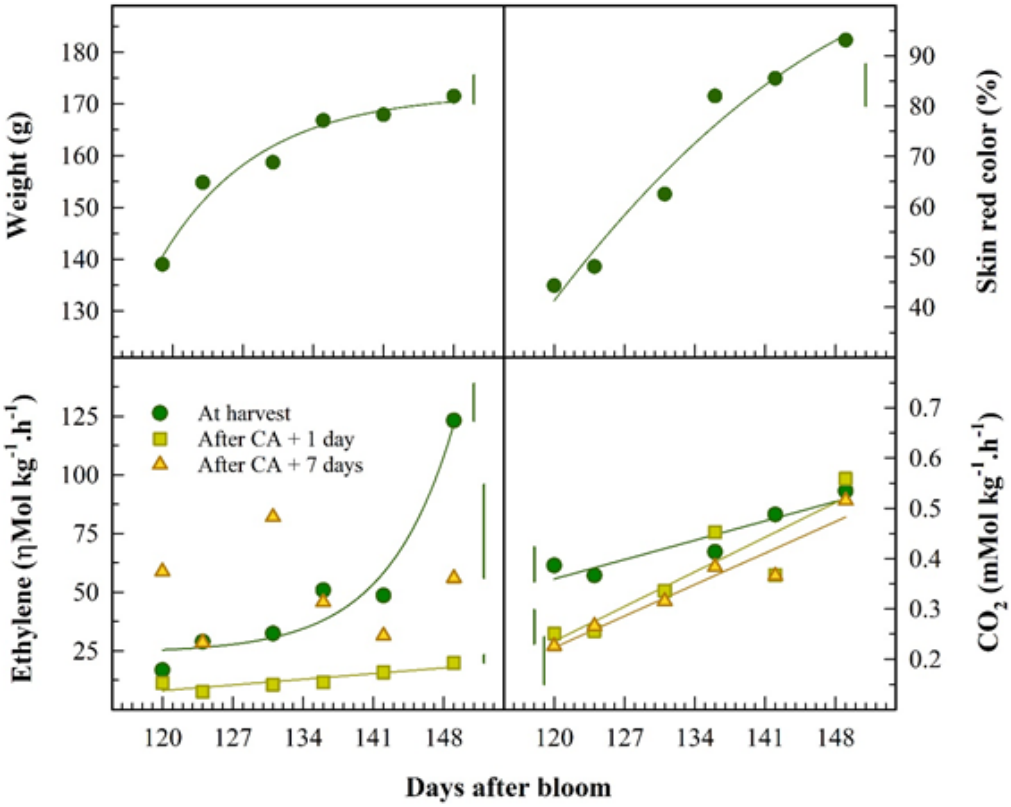


**Figure 1.** 'Monalisa' apple weight and skin red color at harvest, as well as ethylene production and respiration rate at harvest and after storage in response to harvesting on different days after full bloom. The fruit were analyzed at harvest and after six months of storage under a controlled atmosphere plus one and seven days of shelf life at 22 °C. Lines represent statistical models with $P < 0.05$. Vertical bars represent the least significant differences ($P < 0.05$) for the harvest time. Data obtained in Experiment 1 (2011).

The growth rate of 'Monalisa' apple (12 g/week) at the early maturity stages, when the starch index ranged from 1.9 to 4.7, was higher than that observed in 'Gala' apple, 9.5 g/week (ARGENTA et al., 2018b). However, the growth rate of 'Gala' apple is higher than 'Monalisa' apple at more advanced maturity stages, when the starch index ranges from 4.7 to 7.5 (Argenta et al., 2018b).

The rates of ethylene production by 'Monalisa' apple during on-tree maturation (40 to 120 ηMol $kg^{-1}$ $h^{-1}$) were similar to the

rates observed in 'Gala' apple (ARGENTA et al., 2018b), which increased about 2.8-fold from 120 to 142 DAFB (Figure 1, Table 1). The exponential increase in ethylene production rate observed in 'Monalisa' apple was mainly due to the 2.5-fold increase in one week between 142 and 149 DAPF (Figure 1, Table 1).

**Table 1**. Statistical models for 'Monalisa' apple quality traits at harvest, and after storage and shelf life in response to harvesting on different days after full bloom. The fruit were analyzed at harvest and after six months of storage under controlled atmosphere plus one and seven days of shelf life at 22 °C. Data obtained in Experiment 1 (2011).

| | **Fresh weight (g)*** | **$R^2$** | **Skin red color (%)** | |
|---|---|---|---|---|
| At harvest | y=-7127406.06+7127578.10*(1-exp(-0.10*x)) | 97.9*** | y=-671.11+9.25*x+(-0.02)*x^2 | 98.2** |
| | **Starch index (1-9)** | | **Soluble solids content (SSC, %)** | |
| At harvest | y=2.25+5.51/(1+exp(-(x-136.97)/4.09)) | 99.9*** | SS y=5.05+0.06*x | 76.8*** |
| | **Ethylene production (nMol $kg^{-1}$ $h^{-1}$)** | | **Respiration rate (mMol of $CO_2$ $kg^{-1}$ $h^{-1}$)** | |
| At harvest | y=24.12+1.96*exp(0.15*x) | 97.5** | y = - 0.3012+0.0055*x | 88.2 ** |
| 1 day | y=-33.48+0.34*x | 86.8** | y= -0.94+0.009*x | 90.4** |
| 7 days | | | y= -0.85+0.009*x | 95.6*** |
| | **Flesh firmness (N)** | | **Titratable acidity (TA, %)** | |
| At harvest | y=-241.1+5.6*x+(-0.024)* x^2 | 99.7*** | y=-0.99+0.02*x+(-0.0001)*x^2 | 97.9*** |
| 1 day | y= 254.8+(-1.36)*x | 98.6*** | y=0.97+(-0.004)*x | 92.8*** |
| 7 days | y= 266.6+(-1.48)*x | 96.9*** | y=1.11+(-0.005)*x | 92.9*** |
| | **Background color (1-5)** | | **SSC/TA ratio** | |
| At harvest | y=17.08+(-0.27)*x+0.001*x^2 | 97.0** | y=152.08+(-2.21)*x+0.09*x^2 | 97.8*** |
| 1 day | | | y=35.21+(-0.446)*+x+0.003*x^2 | 99.2*** |
| 7 days | y=3.77+0.08*x | 80.1*** | y=195.48+(-2.96)*x+0.01*x^2 | 97.6*** |

*Only statistically significant models at $P < 0.001$ (***), $P < 0.01$ (**), and $P < 0.05$ (*) are presented.

The simultaneous increase of ethylene production and starch index in 'Monalisa' apple (Figure 2, Table 1) was similar to that observed in other cultivars such as Gala (ARGENTA; MONDARDO, 1994; PLOTTO et al., 1995) and Ambrosia (DELONG et al., 2016). However, it differs from cultivars such as Honeycrisp (DELONG et al., 2014) and Fuji (PLOTTO et al., 1995), in which the significant increase in starch degradation begins two to three weeks before the ethylene production rise. The earlier rise of the starch index, compared to the rise of ethylene production, suggests that starch hydrolysis can be initially triggered by increasing fruit sensitivity to ethylene, which has been demonstrated in other studies by pre-harvest treatments with 1-methylcyclopropene (ARGENTA et al., 2018a). The close relationship between starch index and ethylene action, associated with the simplicity of the starch index analysis, makes it one of the most valuable maturity indices for apples (KNEE et al., 1989; KINGSTON, 1992; TOIVONEN, 2007).

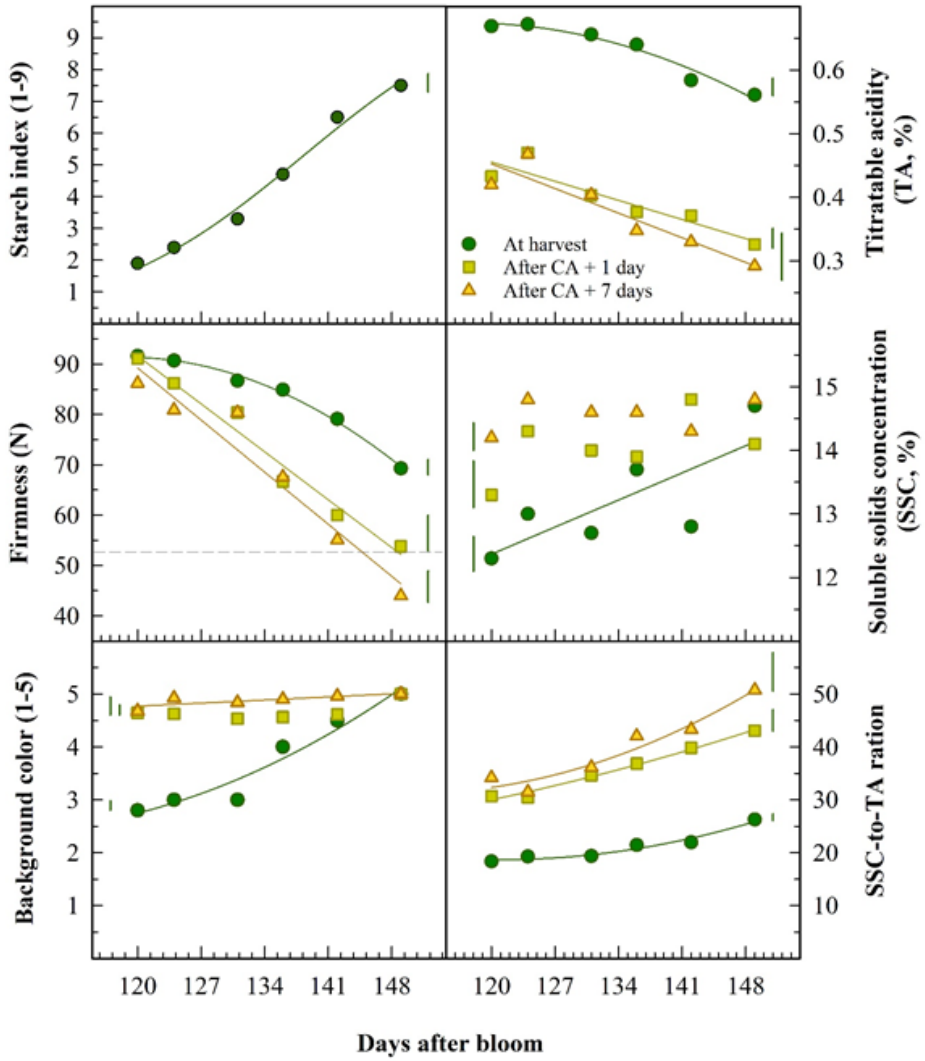


**Figure 2.** 'Monalisa' apple starch index at harvest, as well as titratable acidity, flesh firmness, soluble solids content, background color, and SSC/TA ratio at harvest and after storage in response to harvesting on different days after full bloom. The fruit were analyzed at harvest and after six months of storage under a controlled atmosphere plus one and seven days of shelf life at 22 °C. Lines represent statistical models with $P < 0.05$. Vertical bars represent the least significant differences ($P < 0.05$) for the harvest time. Data obtained in Experiment 1 (2011).

The starch index and flesh firmness rates of change along on-tree apple fruit maturation are practical benchmarks to predict the beginning and the end of the harvest season, especially for fruit intended for long-term storage (ARGENTA et al., 1995). In 'Monalisa' apple, the starch index increased at a rate of 1.6 per week from 124 DAFB (Figure 2, Table 1), which was higher than that observed in 'Gala' apple with a starch index increase rate of 0.94 per week (ARGENTA et al., 1995). On the other hand, the flesh firmness loss rate in 'Monalisa' apple was lower (3.6 N per week) at the beginning (120 to 131 DAFB), and higher (7.1 N per week) at more advanced maturity stages (131 to 149 DAFB), compared to that of 'Gala' apple (5.3 N per week) (ARGENTA et al., 1995). The rate of flesh firmness loss in 'Fuji' apple is approximately 3.6 N per week in Brazil (ARGENTA et al., 2022) and 1.8 N per week in North America (PLOTTO et al., 1995) indicating environmental effects on pace of on-tree apple fruit maturation. In 'Monalisa' apple, the high rate of firmness loss during on-tree maturation was consistent with the rapid fruit softening during cold storage (ARGENTA et al., 2023a).

The effect of harvest date on fruit quality at harvest compared to after CA storage

remained similar for TA, decreased for SSC, and increased for flesh firmness (Figure 2). The greater impact of harvest date on flesh firmness after storage (Figure 2) and the incidence of decay and physiological disorders such as, skin browning (scald-like browning), wrinkly skin, cracking, and shrivel in late (from 142 DAPF) harvested fruits (Table 2) demonstrates that late-harvested fruits' have a lower storability (storage life) than earlier harvested fruit.

**Table 2**. Decay and physiological disorders severity indices in 'Monalisa' apple after six months of storage under controlled atmosphere plus one and seven days of shelf life at 22 °C in response to harvesting on different days after full bloom (DAFB). Data obtained in Experiment 1 (2011).

| Days at 22 °C | DAFB | Decay (1-3)[1] | Skin browning (1-4)[1] | Flesh browning (1-4)[1] | Wrinkling (1-3)[1] | Cracking (1-3)[1] | Withering (1-3)[1] |
|---|---|---|---|---|---|---|---|
| 1 | 120 | 1 | 1 | 1 | 1 | 1 | 1 |
| | 124 | 1 | 1.08 | 1.12 | 1 | 1 | 1 |
| | 131 | 1 | 1.02 | 1.24 | 1 | 1 | 1 |
| | 136 | 1 | 1 | 1 | 1 | 1 | 1 |
| | 142 | 1.18 | 1.16 | 1.06 | 1 | 1.12 | 1 |
| | 149 | 1 | 1.71 | 1 | 1.12 | 1.41 | 1 |
| | Linear | ns | *** | ns | * | **** | - |
| | Quadratic | ns | ns | ns | ns | ** | - |
| 7 | 120 | 1 | 1 | 1 | 1 | 1 | 1 |
| | 124 | 1 | 1 | 1.04 | 1 | 1 | 1 |
| | 131 | 1 | 1 | 1.23 | 1 | 1 | 1 |
| | 136 | 1 | 1.05 | 1.54 | 1.05 | 1 | 1 |
| | 142 | 1.04 | 1.31 | 1.35 | 1.19 | 1.12 | 1.85 |
| | 149 | 1.25 | 1.25 | 2.31 | 1.19 | 1.56 | 2.25 |
| | Linear | ** | ** | **** | *** | **** | **** |
| | Quadratic | * | ns | * | ns | **** | **** |

[1]Disorder severity index. Linear and quadratic effects of harvesting date. ns = not significant at $P < 0.001$ (***), $P < 0.01$ (**), and $P < 0.05$ (*).

## *Experiment 2*

The starch index and SSC differed among harvest dates one day after harvest, but firmness and TA did not (Table 3). Fruit from all harvest dates were at early maturity stages with starch index ranging from 1.2 at 119 DAFB to 4.5 at 137 DAFB (Table 3). At harvest, fruit from Experiment 2 presenting similar starch index to those of Experiment 1 had a similar flesh firmness and TA, but higher SSC (Figure 2 and Table 2).

**Table 3.** Starch index, flesh firmness, soluble solids content (SSC), and titratable acidity (TA) of 'Monalisa' apple at harvest in Experiment 2 (2017) and Experiment 3 (2019) in response to harvesting on different days after full bloom (DAFB).

| | DAFB | Starch index (1-9)* | Flesh firmness (N) | SSC (%) | TA (%) |
|---|---|---|---|---|---|
| Experiment 2 (2017) | 119 | 1.2 c | 90.7 a | 12.5 b | 0.628 a |
| | 128 | 2.4 b | 88.0 a | 14.3 a | 0.594 a |
| | 137 | 4.5 a | 87.1 a | 14.6 a | 0.574 a |
| Experiment 3 (2019) | 126 | 3.4 c | 86.2 a | 12.8 a | - |
| | 136 | 4.6 b | 83.6 a | 13.2 a | - |
| | 143 | 7.1 a | 72.0 b | 14.1 a | - |

*Means in the same experiment (year) followed by different letters are statistically different according to the Tukey HSD test ($P < 0.05$).

After storage in an air atmosphere, 1-MCP fruit from the later harvest date softened more than those from the earlier harvest (Table 4), as observed for fruit harvested at an advanced maturity and stored in CA in Experiment 1. Fruit treated with 1-MCP maintained a higher firmness, TA, and SSC during storage than untreated fruit regardless of harvest date.

**Table 4.** Flesh firmness, titratable acidity (TA), and soluble solids content (SSC) of 'Monalisa' apple in response to harvesting on different days after full bloom (DAFB). The apples were treated with (1-MCP) or without (C) 1-methylcyclopropene one day after harvest and were stored for three and six months at 0.8 °C plus seven days of shelf life at 22 °C. Data obtained in Experiment 2 (2017).

| | DAFB | Flesh firmness (N)* | | | TA (%) | | | SSC (%) | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | C | 1-MCP | Mean | C | 1-MCP | Mean | C | 1-MCP | Mean |
| 3 months of cold storage + 7 days of shelf life | 119 | 52.9 Ba | 88.0 Aa | 70.2 | 0.337 | 0.489 | 0.413 | 13.6 | 13.9 | 13.7 b |
| | 128 | 52.4 Ba | 86.2 Aa | 69.3 | 0.328 | 0.441 | 0.385 | 14.2 | 14.7 | 14.5 a |
| | 137 | 50.7 Ba | 76.0 Ab | 63.6 | 0.333 | 0.451 | 0.392 | 13.8 | 14.2 | 14.0 b |
| | Mean | 52.0 | 83.5 | | 0.333 B | 0.460 A | | 13.8 B | 14.3 A | |
| 6 months of cold storage + 7 days of shelf life | 119 | 48.0 Ba | 85.8 Aa | 67.1 | 0.308 | 0.412 | 0.360 a | 13.5 | 13.8 | 13.7 b |
| | 128 | 48.0 Ba | 84.0 Aa | 66.2 | 0.279 | 0.387 | 0.333 ab | 13.9 | 14.4 | 14.2 a |
| | 137 | 45.3 Ba | 78.7 Ab | 64.4 | 0.270 | 0.358 | 0.314 b | 13.7 | 14.3 | 14.0 ab |
| | Mean | 47.1 | 83.1 | | 0.285 B | 0.386 A | | 13.7 B | 14.2 A | |

*Means in each storage + shelf-life time followed by different uppercase letters in each DAFB (row) and lowercase letter in each treatment (column) are statistically different according to the Tukey HSD test ($P < 0.05$).

Severity indices of decay, flesh browning (FB), cracking, and wrinkly skin increased with late harvest, especially for fruit not treated with 1-MCP that were stored for a longer period (Table 5). The skin browning was the predominant disorder, and its severity was greater after six months than after three months of storage and in fruits not treated with 1-MCP. This disorder only occurred on the third harvest date for 1-MCP fruit but was not consistently affected by the harvest date in fruit not treated with 1-MCP. The treatment with 1-MCP reduced decay and physiological disorders except for wrinkly skin.

**Table 5.** Decay and physiological disorders severity indices in 'Monalisa' apple in response to harvesting on different days after full bloom (DAFB). The apples were treated with (1-MCP) or without (C) 1-methylcyclopropene one day after harvest and were stored for three and six months at 0.8 °C plus seven days of shelf life at 22 °C. Data obtained in Experiment 2 (2017).

| | DAFB | Decay (1-3)[1] | | Skin browning (1-4)[1] | | Flesh Browning (1-4)[1] | | Cracking (1-2)[1] | | Wrinkling (1-3)[1] | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | C* | 1-MCP | C | 1-MCP | C | 1-MCP | C | 1-MCP | C | 1-MCP |
| 3 months of cold storage + 7 days of shelf life | 119 | 1.02Ab | 1.03Aa | 1.90Aa | 1.00Ba | 1.00Aa | 1.00Aa | 1.00Ab | 1.00Aa | 1.00Aa | 1.00Aa |
| | 128 | 1.02Ab | 1.00Aa | 1.90Aa | 1.00Ba | 1.10Aa | 1.00Aa | 1.00Ab | 1.00Aa | 1.00Aa | 1.00Aa |
| | 137 | 1.30Aa | 1.03Ba | 1.60Ab | 1.03Ba | 1.03Aa | 1.10Aa | 1.20Aa | 1.00Ba | 1.00Aa | 1.00Aa |
| 6 months of cold storage + 7 days of shelf life | 119 | 1.10Ac | 1.00Ab | 3.40Ab | 1.00Bb | 1.10Ab | 1.03Ab | 1.02Ab | 1.00Aa | 1.00Ab | 1.00Ab |
| | 128 | 1.30Ab | 1.02Bb | 3.80Aa | 1.00Bb | 1.60Aa | 1.10Bab | 1.10Ab | 1.00Aa | 1.00Ab | 1.00Ab |
| | 137 | 1.80Aa | 1.20Ba | 3.30Ab | 1.30Ba | 1.90Aa | 1.20Ba | 1.30Aa | 1.00Ba | 1.40Aa | 1.20Aa |

[1] Disorder severity index. *Means in each storage + shelf-life time followed by different uppercase letters in each DAFB (row) and lowercase letter in each treatment (column) are statistically different according to the Tukey HSD test ($P < 0.05$).

## *Experiment 3*

The starch index, firmness, and SSC at harvest differed among harvest dates (Table 3). Fruit from this Experiment were harvested one week later than those from Experiment 2 based on the date of full bloom and, therefore, were more mature at harvest.

Differences in flesh firmness between harvest dates were maintained or increased after storage, as observed in Experiments 1 and 2, regardless of 1-MCP treatment and storage atmosphere (Table 6). Both CA and 1-MCP increased firmness retention regardless of harvest time. There was an additive effect of 1-MCP treatment and CA on firmness retention. However, 1-MCP+Air fruit maintained a higher firmness than untreated CA fruit, regardless of storage duration. This marked response of 'Monalisa' apple to 1-MCP and CA is consistent with a previous study on 'Monalisa' (ARGENTA et al., 2023a), and is similar to other cultivars such as 'Gala' and 'Delicious' (BAI et al., 2005). However, other studies show that untreated 'Gala' apple stored in CA maintain a higher firmness than apple treated with 1-MCP and stored in air beyond the fifth month of storage (MATTHEIS et al., 2005).

**Table 6.** Flesh firmness (N) of 'Monalisa' apple in response to harvesting on different days after full bloom (DAFB). The apple were treated with (1-MCP) or without (C) 1-methylcyclopropene one day after harvest and were kept for five and nine months in cold Air (0.8 °C) or controlled atmosphere (CA) plus seven days of shelf life at 22°C. Data obtained in Experiment 3 (2019).

| | DAFB | Air | 1-MCP | CA | 1-MCP+CA | Mean |
|---|---|---|---|---|---|---|
| 5 months of cold storage + 7 days of shelf life | 126 | 50.2* | 84.4 | 59.1 | 85.3 | 69.8 a |
| | 136 | 44.9 | 77.3 | 53.8 | 80.9 | 64.0 b |
| | 143 | 39.1 | 63.1 | 49.8 | 68.4 | 55.6 c |
| | Mean | 44.9 C | 75.1 A | 54.2 B | 78.2 A | |
| 9 months of cold storage + 7 days of shelf life | 126 | 36.0 | 80.0 | 52.9 | 82.7 | 62.7 a |
| | 136 | 27.6 | 69.8 | 47.1 | 73.8 | 57.8 ab |
| | 143 | ** | 60.4 | 42.2 | 63.6 | 56.0 b |
| | Mean | 31.1 C | 70.2 A | 48.0 B | 73.8 A | |

*Means in each storage + shelf-life time followed by different uppercase letters in each DAFB (row) and lowercase letter in each treatment (column) are statistically different according to the Tukey HSD test ($P < 0.05$). **Fruit severely affected by decay.

The severity indices of most disorders and decay were increased with late harvest; however, the effect was dependent on 1-MCP treatment, storage atmosphere, and storage period (Table 7). There was no effect of harvest date when disorder indices were low, such as decay and senescent breakdown in 1-MCP+Air fruit, cracking for 1-MCP+Air and 1-MCP+CA fruit, and wrinkling in untreated Air fruit (Table 7).

Senescent breakdown was only observed in Experiment 3, which was characterized by smooth and moist brown areas on the skin, often associated with a disrupted epidermis (Figure 3). The 1-MCP treatment and CA reduced the severity index of senescent breakdown in fruit stored for nine months (Table 7). In addition, 1-MCP and CA reduced the severity index of decay, skin browning, cracking, and FB, mostly after a longer storage period (Table 7). In contrast, the wrinkling severity index was increased by 1-MCP and CA storage (Table 7).

**Table 7.** Decay and physiological disorders severity indexes in 'Monalisa' apple in response to harvesting on different days after full bloom (DAFB). The apples were treated with (1-MCP) or without (C) 1-methylcyclopropene one day after harvest and were kept for five and nine months in cold Air (0.8 °C) or controlled atmosphere (CA) plus seven days of shelf life at 22°C. Data obtained in Experiment 3 (2019).

| | | DAFB | Air | 1-MCP | CA | 1-MCP+CA |
|---|---|---|---|---|---|---|
| Decay (1=3)[1] | 5 months + 7 days of shelf life | 126 | 1.15Ab* | 1.1Aa | 1.0Ab | 1.0Ab |
| | | 136 | 1.15Ab | 1.0Ba | 1.3Aab | 1.05Ab |
| | | 143 | 1.85Aa | 1.1Ca | 1.4Ba | 1.3BCa |
| | 9 months + 7 days of shelf life | 126 | 1.2Ac | 1.0Ba | 1.0Bb | 1.05Bb |
| | | 136 | 2.1Ab | 1.1Ba | 1.25Bb | 1.00Bb |
| | | 143 | 3.0Aa | 1.05Da | 1.85Ba | 1.45Ca |
| Cracking (1-2)[1] | 5 months + 7 days of shelf life | 126 | 1.0Ab | 1.0Aa | 1.00Aa | 1.0Aa |
| | | 136 | 1.0Ab | 1.0Aa | 1.05Aa | 1.0Aa |
| | | 143 | 1.2Aa | 1.0Ba | 1.05Aba | 1.1Aba |
| | 9 months + 7 days of shelf life | 126 | 1.00Ac | 1.00Aa | 1.05Ab | 1.0Aa |
| | | 136 | 1.30Ab | 1.05Ba | 1.05Bb | 1.0Ba |
| | | 143 | 1.85Aa | 1.10Ca | 1.40Ba | 1.1Ca |
| Flesh browning (1-4)[1] | 5 months + 7 days of shelf life | 126 | 1.0Ab | 1.0Aa | 1.0Aa | 1.0Aa |
| | | 136 | 1.0Ab | 1.0Aa | 1.0Aa | 1.0Aa |
| | | 143 | 1.5Aa | 1.0Ba | 1.0Ba | 1.05Ba |
| | 9 months + 7 days of shelf life | 26 | 1.80Aa | 1.00Ba | 1.10Ba | 1.05Ba |
| | | 136 | 1.65Aa | 1.10Ba | 1.05Ba | 1.00Ba |
| | | 143 | - | 1.05Aa | 1.15Aa | 1.10Aa |
| Skin Browning (1-4)[1] | 5 months + 7 days of shelf life | 126 | 3.0Aa | 1.05Ba | 1.2Ba | 1.0Ca |
| | | 136 | 1.7Ab | 1.0Ba | 1.1Bab | 1.0Ba |
| | | 143 | 2.0Ab | 1.0Ba | 1.0Bb | 1.0Ba |
| | 9 months + 7 days of shelf life | 126 | 3.75Aa | 1.10BCa | 1.25Bb | 1.05Ca |
| | | 136 | 1.60Aab | 1.20Ba | 1.60Aa | 1.10Ba |
| | | 143 | 1.45Ab | 1.25Aa | 1.20Ab | 1.25Aa |
| Wrinkling (1-3)[1] | 5 months + 7 days of shelf life | 126 | 1.0Aa | 1.00Aa | 1.0Ab | 1.00Ab |
| | | 136 | 1.0Ba | 1.05Aba | 1.2Aba | 1.25Aa |
| | | 143 | 1.0Ba | 1.10Aba | 1.0Bb | 1.20Aab |
| | 9 months + 7 days of shelf life | 126 | 1.05Aa | 1.05Aa | 1.20Ab | 1.05Ab |
| | | 136 | 1.0Ba | 1.25Aba | 1.45Aab | 1.40Ab |
| | | 143 | 1.0Ca | 1.30BCa | 1.50Aba | 1.75Aa |
| Senescent breakdown (1-4)[1] | 5 months + 7 days of shelf life | 126 | 1.0Aa | 1.0Aa | 1.0Aa | 1.0Aa |
| | | 136 | 1.0Aa | 1.0Aa | 1.0Aa | 1.0Aa |
| | | 143 | 1.0Aa | 1.0Aa | 1.0Aa | 1.0Aa |
| | 9 months + 7 days of shelf life | 26 | 1.10Ab | 1.0Aa | 1.00Ab | 1.0Ab |
| | | 136 | 1.75Aa | 1.0Ba | 1.10Bab | 1.0Bb |
| | | 143 | 1.25Aab | 1.0Aa | 1.25Aa | 1.2Aa |

[1]Decay or disorder severity index. *Means in each storage + shelf life time followed by different uppercase letters in each DAFB (row) and lowercase letter in each treatment (column) are statistically different according to the Tukey HSD test ($P < 0.05$).

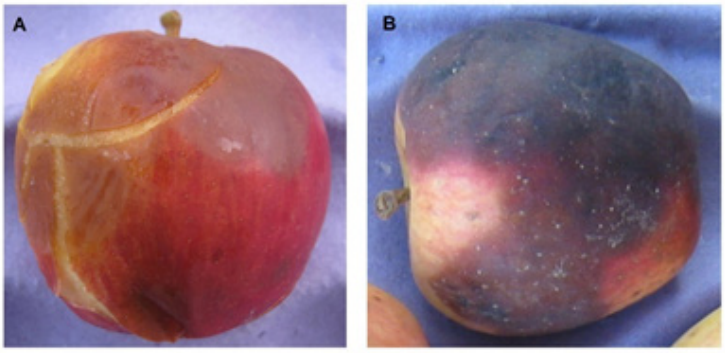


**Figure 3.** 'Monalisa' apple produced in Southern Brazil showing symptoms of senescence (A) and Alternaria rot (B).

In general, the on-tree maturation pattern of the 'Monalisa' apple was similar to that of 'Gala', based on the ethylene production rates and fruit softening, but it was slightly faster than 'Gala', considering the rate of starch degradation (Figures 1 and 2) (ARGENTA ; MONDARDO, 1994; PLOTTO et al., 1995). Additionally, 'Monalisa' apple maintained higher TA and lower SSC/TA ratio during on-tree maturation, compared to other cultivars such as 'Gala' and 'Fuji' (ARGENTA et al., 1995; PLOTTO et al., 1995).

The responses of 'Monalisa' apple to 1-MCP and CA were greater than those observed for other cultivars, such as 'Gala' and 'Fuji', considering traits such as flesh firmness and incidence of physiological disorders (BAI et al., 2005; MATTHEIS et al., 2005). Therefore, although 'Monalisa' apple have rapid softening during cold storage, fruit can retain a flesh firmness above 62.2 N for five to six months if treated with 1-MCP and/or stored under CA conditions (Figure 2, Tables 4 and 6) (ARGENTA et al., 2023a; THEWES et al., 2023).

The incidence of physiological disorders was the most important factor limiting 'Monalisa' apple storage, which was highly affected by harvest maturity. The severity indices of disorders were higher in late-harvested fruit in all three experiments, except for skin browning. 1-MCP and/or CA reduced fruit susceptibility to most of the disorders. However, 1-MCP is not registered for organic apple production and CA can trigger $CO_2$ injury in 'Monalisa' apple (THEWES et al., 2023).

Skin browning was the most prevalent physiological disorder in 'Monalisa' apple, which appeared as diffuse brown skin discoloration confined to the fruit surface, sometimes slightly roughened, and often on the unblushed side of the fruit (ARGENTA et al., 2023a). However, the symptoms can also develop on the blushed side resembling the senescent scald of 'Golden Delicious' apple (PIERSON et al., 1971). This disorder increases with the storage period and is inhibited by 1-MCP and CA (Tables 5 and 7) (ARGENTA et al., 2023a), consistent with other senescent-related disorders and superficial scald (WATKINS; MATTHEIS, 2019).

Unlike the ordinary superficial scald, the skin browning disorder in 'Monalisa' apple was more severe in late-harvested fruit and occurred a few times only on the blushed side of the fruit. Skin browning was not consistently affected by harvest maturity in two of the three experiments of the current study. For 1-MCP treated fruit, the severity of skin browning was higher in later harvested fruit, whereas harvest maturity had no effect on skin browning severity in untreated fruit. The lower severity of skin browning in late harvested fruit from Experiments 2 and 3 may be partly due to the development of severe decay symptoms that prevented the disorder's visual assessment.

Decay was the second predominant cause of postharvest 'Monalisa' fruit deterioration markedly reduced by early harvest matu-

rity, 1-MCP, and/or CA as observed in previous study with this cultivar (ARGENTA et al., 2023a). Indeed, early harvest maturity has been an essential strategy to reduce postharvest losses caused by fungal decay, especially for organic apple production system (BØRVE et al., 2013). However, a comprehensive study under commercial conditions shows no effect of 1-MCP on decay incidence in 'Gala' and 'Fuji' apple (ARGENTA et al., 2021). Alternaria was the main symptom of fungal decay observed in 'Monalisa' (Figure 3). The pathogen *Alternaria sp*. causing this symptom does not grow readily on healthy tissues but can attack tissues weakened by postharvest stress, such as low-temperature exposure (MCCOLLOCH; WORTHINGTON, 1952). Therefore, early harvest maturity, 1-MCP, and/or CA reduced susceptibility to decay in stored 'Monalisa' apple possibly by delaying ripening, reducing disorders associated with senescence, and maintaining stronger pathogen defense mechanisms in the fruit.

### *'Monalisa' apple maturity indices for the optimum harvest time*

The revenue of apple production is influenced by quality grade and fruit size (CAREW and SMITH, 2004). The grade standards for apple are based on the area of red coloration on the fruit skin (for red or bicolored apple cultivars), as well as the incidence and severity of defects (e.g., physiological disorders) (USDA, 2002; OECD, 2010). Additionally, apple fruit with suboptimal sensory taste decreases consumers' willingness to purchase (HARKER et al., 2008). All these quality attributes are generally more extraordinary in on-tree ripened apple fruit (i.e., late harvest) than stored apples that are usually harvested at an early maturity. However, for 'Monalisa' apple fruit, the risk of quality deterioration due to the incidence of decay and physiological disorders after storage was higher in fruit harvested at advanced maturity. Therefore, the optimal harvest window and respective maturity indices were defined by a tradeoff between the fruit quality at harvest (e.g., skin red color) and storability, especially the incidence of storage disorders. In this sense, the current results suggest that 'Monalisa' apple for immediate fresh market after harvest may be harvested from 131 to 149 DAFB, when the maturity indices are 3.3 to 7.5 for starch (1-9 scale), 87.1 N to 69.3 N for firmness, 12.7 % to 14.7 % for SSC, 0.66 % to 0.56 % for TA and 3 to 5 (scale 1-5) for ground color (Figure 2, Table 2). 'Monalisa' apple fruit intended for mid- or long-term storage should be harvested earlier in a narrower harvest window, between 124 and 131 DAPF, when the maturity indices are 2.4 to 3.4 for starch, 90.7 N to 86.2 N for firmness, 12.7 % to 14.3 % for SSC, 0.67 % to 0.59 % for TA and 3 (scale 1-5) for ground color (Figure 2, Table 2). Data from Experiment 2 indicate that the harvest window can be extended to up to 137 DAPF (4.5 for starch and 87.1 N for firmness) for 'Monalisa' apple fruit intended for short-term storage in cold air following 1-MCP treatment (Table 3, 4 and 5). This extended harvest window is possibly also applicable for short-term CA fruit, considering that the maintenance of 'Monalisa' fruit treated with 1-MCP and stored in air is similar to untreated fruit stored in CA for 2 and 4 months (ARGENTA et al., 2023a).

## Conclusions

The on-tree maturation pattern of 'Monalisa' apple was similar to that of early-season apple cultivars, such as its progenitor 'Gala'. 'Monalisa' apple exhibit a rapid loss of firmness during storage and are susceptible to many physiological disorders after storage. Severity indices of decay and physiological storage disorders are markedly higher in late- compared to early-harvested fruit. Skin browning was the predominant disorder, and harvest time's effect on this disorder was variable. The results suggest that the interactions among harvest date, year,

and postharvest factors (1-MCP treatment, storage atmosphere, and duration) can influence skin browning development. The study allowed the estimate of the maturity indices for the optimum harvest time for 'Monalisa' apple by relating the harvest date and fruit quality at harvest and after storage.

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