# Original article



# Early harvest of tomato cv. Absoluto combined with different postharvest conditions increases shelf life without compromising sensorial attributes

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# **Summary**

Introduction - Most studies state that ripe harvested tomatoes present superior sensorial attributes compared to unripe harvested. However, in many occasions, the harvest of unripe tomatoes is necessary to cope with the long distribution chain from field to consumers located in the greater consumption centres, since fruit tend to be firmer and more resistant to manipulation. In this context, tomato (cv. Absoluto), harvested at mature green and breaker stages, were tested for sensorial attributes and physicochemical parameters in order to evaluate the significance of harvesting stage and storage temperature. Materials and methods - Treatments consisted in 1-methylcycloprone (1-MCP) treated/non-treated and different storage temperatures (room temperature, 25 °C and 4 °C). Results and discussion - Tomato harvested at green stage, treated with 1-MCP (300 ppm) and stored at room temperature (RT) or 25 °C had an extended shelf life of 10 and 12 days, respectively, without compromising sensorial characteristics. This effect was not observed on fruit harvested at breaker stage, non-treated with 1-MCP and stored at RT or 25 °C. Lower storage temperatures (4 °C) increased by 3 days the shelf life of tomatoes harvested at breaker stage irrespective of 1-MCP treatment, maintaining the desired characteristics for consumption with exception of fruit color. Similar effect was not obtained when tomatoes at breaker stage treated or not with 1-MCP were kept at RT or 25 °C. Conclusion - The early harvest, at mature green stage, with application of 1-MCP (300 ppm) and storage at RT or without 1-MCP and storage at 4 °C, are feasible early-harvest strategies capable of extending the tomato shelf life without compromising the desirable characteristics of ripe fruit.

# Keywords

tomato, Lycopersicum esculentum, fruit quality, maturity stage, phenolics, shelf life

## Résumé

Une récolte précoce de tomates cv. Absoluto combinée avec différentes conditions de postrécolte augmente la durée de conservation

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# Significance of this study

What is already known on this subject?

 Harvest at later developmental stages and storage/ transport at room temperatures is a common practice to obtain mature fruit with good sensorial qualities.

What are the new findings?

 Fruit harvested at earlier stages (mature green) and allowed to ripe at room temperature or at 25 °C, did not present losses in sensorial attributes, and in addition improved shelf life.

What is the expected impact on horticulture?

• These results reinforce the possibility of conciliation of shelf life and sensorial quality of fruits and vegetables, and address the customers' increasing demand for more flavor and aroma allied with fruit homogeneity.

# sans compromettre les attributs sensoriels.

Introduction - La plupart des études indiquent que les tomates récoltées mûres présentent des attributs sensoriels supérieurs à ceux des tomates vertes. Cependant, en de nombreuses occasions, la récolte de tomates vertes est nécessaire pour faire face à la longue chaîne de distribution du champ aux consommateurs situés dans les grands centres de consommation, car les fruits tendent à être plus fermes et plus résistants à la manipulation. Dans ce contexte, la tomate (cv. Absoluto) récoltée à des stades vert et virage de couleur, a été testée pour les attributs sensoriels et les paramètres physico-chimiques afin d'évaluer l'importance du stade de récolte et de la température de stockage. Matériel et méthodes - Les traitements ont consisté en 1-méthylcicloprone (1-MCP à 300 ppm) traité/ non-traité, et différentes températures de stockage (température ambiante, 25 °C et 4 °C). Résultats et discussion - Les tomates récoltées au stade vert, traitées au 1-MCP et conservées à température ambiante (RT) ou à 25 °C ont eu une durée de conservation prolongée de 10 et 12 jours, respectivement, sans compromettre leurs caractéristiques sensorielles. Cet effet n'a pas été observé sur les fruits récoltés au stade du virage de couleur, non traités au 1-MCP et conservés à température ambiante ou à 25 °C. Les températures

de conservation plus basses (4 °C) ont augmenté de 3 jours la durée de conservation des tomates récoltées au stade du virage indépendamment du traitement au 1-MCP, en maintenant les caractéristiques de consommation souhaitées à l'exception de la couleur du fruit. Un effet similaire n'a pas été observé lorsque les tomates au stade du virage traitées ou non avec du 1-MCP étaient maintenues à température ambiante ou à 25 °C. Conclusion – Une récolte précoce, au stade vert mature, avec application de 1-MCP (300 ppm) et stockage à 4 °C, sont des stratégies de récolte précoce qui permettent de prolonger la durée de conservation de la tomate sans compromettre les caractéristiques souhaitables des fruits mûrs.

#### Mots-clés

tomate, *Lycopersicum esculentum*, qualité du fruit, stade de maturité, composés phénoliques, aptitude à la conservation

## Introduction

The loss of characteristic flavor and aroma is leading to the stagnation in consumption and increasing consumer dissatisfaction with the quality of in natura tomatoes. In response, sensorial attributes as flavor and aroma are being considered paramount factors to improve market acceptability of in natura fruit (Bruhn et al., 1991; Klee, 2010; Renard et al., 2013). A number of studies point to the inverse correlation, in most fruit, between early harvest and the development of sensorial attributes, even in climacteric species (Rombaldi et al., 2007). The majority of the tomato varieties follow the same inverse correlation, with fruit harvested at more advanced ripening stages (breaker and pink red) tending to have a shorter shelf life but also an increased market acceptance, due to improved flavor and aroma (Maul et al., 2000; Mir et al., 2004; Baldwin et al., 2011; Verheul et al., 2015). Still, several studies performed with older tomato varieties present different results, where harvest at early stages does not impair the development of desirable traits related to sensorial quality, like total soluble solids content (Watada and Aulenbach, 1979). In some cases, these traits are even improved by early harvest (Watada and Aulenbach, 1979; Kavanagh et al., 1986). These results point to a highly cultivar-specific behavior when it comes to the influence of harvesting stages in flavor and aroma development in tomato. In many cases, the production regions are located at long distances from the major consumption centres, this situation coupled with the fluctuations in production/consumption rates, require the adoption of early harvest strategies (Baldwin et al., 1998). Other reasons for early harvest include climatic limitations, with special emphasis to frosts. In this context, the harvest of tomatoes in early ripening stages and 1-MCP application is a feasible strategy to cope with the transport distances and production/consumption fluctuations. Based on this scenario, the present research aimed to determine the development of proper sensorial characteristics of the tomato local cultivar 'Absoluto' with emphasis in response to different harvest stages, combined with 1-MCP treatment and different storage temperatures.

# Materials and methods

#### Sample origin

The tomato seeds (*Lycopersicum esculentum* Mill., cv. Absoluto) were obtained from a local farm located at the countryside in the municipality of Pelotas-RS, Brazil. Seeding took place in the month of August 2012 and sampling started at the month of December 2012. During the cropping season, the average temperature and relative humidity were  $24\pm2$  °C and  $80.9\pm10.0$ %, respectively, according to local measurements. The harvest was performed based on fruit maturation; mature green (MG) stage is characterized by 100% green color in fruit surface, with shades of green varying from light to dark; breaker stage (BR) consisted of a "break" in color from green to tannish-yellow, pink or red on not more than 10% of the surface.

The fruits analysed in this study (a total of 192) belong to the hybrid cultivar 'Absoluto', which cycle varies between 110 and 120 days with semi-determinate growth habit. This cultivar is highly productive with fruit weight varying from 300 to 350 g and good density.

#### Treatments and experimental design

The experiment involved three factors: a) maturation stage (MG and BR); b) 1-MCP treated (1-MCPt) or not treated (1-MCPnt), and c) storage conditions (room temperature [RT], 25 °C and 4 °C). The storage temperatures were determined based on the normal practices adopted by producers/transporters (transport at RT) and consumers (storage in refrigerators at 4 °C). The treatment at 25 °C was used to simulate ideal ripening temperature. Relative humidity (RH) on the treatments at 4 °C and 25 °C ranged between 70% and 95%, while at RT treatments the RH was not controlled. The RT temperature was measured daily and achieved an average of  $25\pm3$  °C.

The tomato fruits were treated with 1-MCP (Ethylbloc<sup>™</sup>) at concentration of 300 ppm, according to fruit weight, following the manufacturer instructions. The fruit were exposed to 1-MCP inside sealed desiccators for a period of 24 h, and after that placed in polystyrene trays ( $40 \times 20$  cm) at the different temperatures abovementioned. Two trays with eight fruit inside (one experimental unit) composed each treatment; each tray was considered one biological repetition and three samples (technical repetition) were taken from each tray. The sample collection was performed when the fruit achieved complete ripening (CR), characterized by 90% of the fruit surface presenting red color. For comparison purposes, unripe MG and BR tomato samples were analysed for the same variables as ripe treatments.

#### Sample processing

All analyses were performed in triplicates at two different moments: immediately after harvest and at complete ripening. For all chemical analyses, the samples were grinded to a fine powder in a ball mill (Marconi/MA350) in the presence of liquid nitrogen.

## Physicochemical analyses

#### Soluble solids, sugars and acidity

Total soluble solids (TSS) were measured in the tomato juice using a digital refractometer (ATAGO Co. Ltd., PR-32 $\alpha$ ,) with automatic temperature compensation; the results were expressed as °Brix at 25 °C.



Total sugars (TS) were measured based on the phenol-sulphuric acid method described by Dubois et al. (1956). The sample was prepared by homogenizing (Vortex®) 1.0 g fruit with 45 mL of distilled H<sub>2</sub>O, followed by centrifugation (7,500 rpm for 10 min). The supernatant was collected for the spectrophotometric analysis (Jenway® 6705 UV/Vis Single Cell Spectrophotometer).

Reducing sugars (RS) were quantified based on method described by Miller (1959) with modifications. The sample preparation is the same described for TS. The reaction was made in screw capped glass tubes containing 200 µL of supernatant plus 200 µL of dinitrosalicylic acid reagent (DNS). The mixture was homogenised and let to rest in a thermostatic bath at 100 °C for 5 min. After that, 1.6 mL distilled water were added to the reaction, followed by homogenization and temperature stabilization. Spectrophotometric readings were performed at 540 nm.

Titratable acidity (TA) was obtained by titration of 0.1 M NaOH into a filtered sample solution (2 g fruit homogenised in 48 mL distilled water) until pH stabilization at 7.0 and expressed in citric acid equivalents (mg g-1 sample); the pH was measured with a potentiometer (HI 2221, Hanna).

# Fruit color and firmness

Fruit epidermis color (L\*, a\* and b\* coordinates) was measured using a colorimeter (Minolta CR-300); the measurements were made in opposite sides of the equatorial region of the fruit and the a\* and b\* values were used to calculate the hue angle:

$$(\tan^{-1} b \times a^{-1})$$

and chroma

$$\sqrt{(a)^2 + (b)^2}$$
.

Fruit firmness was determined using a texture analyser (TA.XT Plus, Texture Technologies) equipped with a 2-mm probe; each fruit was penetrated in 25% at a 1 mm s-1 speed, all the measurements were performed in the equatorial region of the fruit and expressed in Newton (N).

## Phenolics and related activity

Total phenolic compounds (TPC) were determined according to Singleton and Rossi (1965) with adaptations. Briefly, 1.0 g of homogenized fruit pulp were transferred to a 100-mL volumetric flask plus 60 mL distilled water and 5 mL Folin-Ciocalteu's reagent. After 8 min, 20 mL of 20% Na<sub>2</sub>CO<sub>3</sub> were added to the reaction and the flask volume was completed to 100 mL with distilled H<sub>2</sub>O. After a 2-h rest in the dark, the reaction was filtered and submitted to spectrophotometric analysis (725 nm).

Antioxidant activity (AA) was quantified according to methodology adapted from Brand-Williams et al. (1995). The samples (5 g homogenised fruit) were extracted with 20 mL methanol (100%). The extract (100 μL) was mixed with 3.9 mL DPPH solution and stored in the dark for 24 h. Spectrophotometric readings were performed at 515 nm.

The L-ascorbic acid (vitamin C or AsAc) content was quantified according to Vinci et al. (1995). A 2-g sample was homogenised in 10 mL meta-phosphoric acid solution (4.5%) and let to stand for 1 h in an aluminium foil-covered flask. The total volume was transferred to a 25-mL Erlenmeyer and the volume adjusted with ultrapure water. The solution was filtered with filter paper and centrifuged at 7,000 rpm for 10 min. The supernatant was used for determination of vitamin C by high-performance liquid chromatography (HPLC); the samples were compared with a HPLC grade commercial standard (Sigma® part no. A92902).

#### Sensorial analysis

The sensorial attributes were evaluated based on an intensity scale (0-9) with "0" being the absence of a given characteristic and "9" an extremely intense characteristic. Fifteen panellists (students, teachers and university staff) of both sexes, aged between 18 and 50 years were selected based on consumption habits. Only those who regularly consume tomato (2-3 times a week) were selected for the experiment. Panellists were trained to properly distinguish and measure the characteristics responsible for consumer acceptance, such as flavor (tomato characteristic flavor, acidity and sweetness), texture (flouriness and firmness) and color. During the training, each panellist tasted tomato samples obtained from local retailers and were instructed how to identify the acceptable limits of the abovementioned characteristics (e.g., acidity limits and color homogeneity).

## Statistical analyses

Different treatments were compared by parametric and non-parametric statistics. Differences in single variables were determined by variance analyses (One-way ANOVA) followed by multiple comparison procedure (Tukey's HSD test). Principal component analyses (PCA) was carried out to reduce variable redundancy, cluster variables/treatments and correlate physicochemical parameters with sensorial evaluation in different treatments. All calculations were performed in SAS® Studio University Edition V. 3.4.

# **Results and discussion**

### Fruit ripening

The period for fruit to achieve complete ripening (CR) varied among the different treatments (Table 1); 1-MCP-treated fruits had an increased shelf life of 7 days for (MG + RT), 9 days (MG + 25  $^{\circ}$ C), 3 days (BR + RT) and 5 days (BR + 25  $^{\circ}$ C) compared to 1-MCP non-treated fruits. Tomatoes stored at 4 °C presented the longest storage period (21–24 days), not being affected by 1-MCP treatment. Harvesting stage had a considerable impact on storage period in fruit kept at room temperature (RT) or 25 °C, where CR was delayed in approximately 7 days for breaker stage (BR). For those fruit stored at 4 °C CR was not affected by both 1-MCP treatment or harvesting stage. At temperatures below 18 °C the effects of 1-MCP or harvest stage seem to be less and less important regarding the shelf life period. According to Baldwin et al. (2011), at 13 °C, fruit ripening is delayed in just 2 days by 1-MCP treatment of fruit harvested at different stages (mature green, breaker, turning or pink).

#### Physicochemical parameters

## Tomato acidity

The determination of titratable acidity (TA) and ascorbic acid (AsAc) content is important considering the role of acids in fruit sensorial attributes; for MG, the highest AsAc occurred in RT + 1-MCPt (2.02 g 100 g-1) (Figure 1), even not differing statistically is possible to observe the tendency of acids maintenance in 1-MCPt fruit stored at 25 °C, compared to 1-MCPnt at the same temperature. This pattern is present in BR fruit as well, with significant statistical difference. The content of AsAc tends to increase to a certain point during

**TABLE 1.** Days until complete ripening (CR), total soluble solids/titratable acidity ratio (TSS/TA), and color indicators (chroma, hue angle and lightness) of tomatoes harvested at mature green and breaker stages, treated or not with 1-MCP and stored at different temperatures.

Ripening stages	1-MCP	Storage conditions	Days until CR	TSS/TA ratio -	Tomato fruit color		
					Chroma	Hue°	Lightness
Mature green	+	RT	18	12.7	47.03 ab	65.13 bc	50.88 bc
		25 °C	19	11.1	41.14 abc	57.07 cd	47.55 cd
		4 °C	24	7.6	49.16 a	84.99 a	61.05 a
	-	RT	11	8.9	41.23 abc	64.45 bc	49.51 cd
		25 °C	10	11.0	30.89 d	49.61 cd	46.49 e
		4 °C	24	9.2	40.53 abc	82.40 a	57.70 ab
Breaker	+	RT	11	12.4	42.39 abc	62.32 c	48.33 cd
		25 °C	13	11.7	43.07 abc	59.44 cd	48.67 cd
		4 °C	21	11.2	34.45 d	75.64 ab	54.02 abc
	-	RT	8	10.6	39.06 cd	57.95 cd	45.71 e
		25 °C	7	9.5	37.47 d	54.87 cd	46.19 e
		4 °C	21	11.2	37.16 d	65.91 bc	49.12bcd

Means (n=3) followed by the same letter in the column do not differ significantly at p  $\leq$  0.05 by Tukey's test.

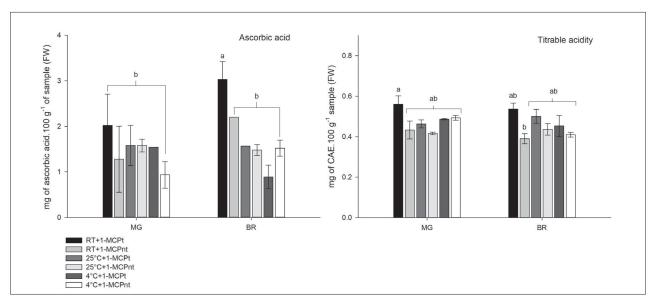
Mature green: 100% green color in fruit surface, with shades of green varying from light to dark; Breaker: A "break" in color from green to tannish-yellow, pink or red on not more than 10% of the surface; RT: Room temperature.

the ripening process, reaching a maximum at approximately the  $20^{\text{th}}$  day of storage and starting to decrease after that (Toor and Savage, 2006; Tigist *et al.*, 2013).

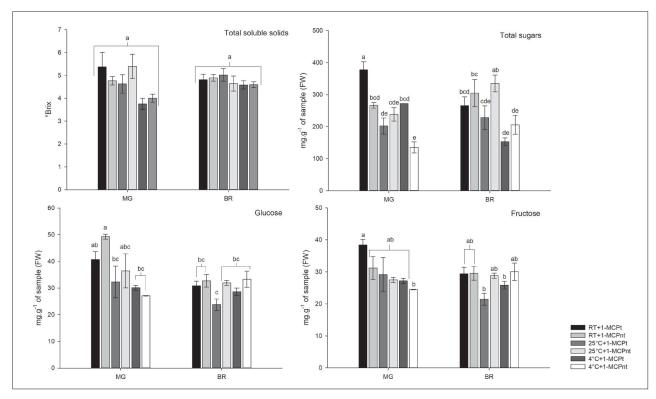
Mature green fruit treated with 1-MCP had a longer storage period (18 days) allowing fruit to develop a greater acid content, differently from those untreated which had a storage period of just 11 days. At 25 °C and 4 °C, the differences in AsAc content are much smaller and not statistically significant; 1-MCP treatment in these conditions did not produce significant differences between treatments. As expected, the variable TA behaves similarly to AsAc contents, with only RT + 1-MCPt treatments presenting a small increase in TA, the reasons for that behavior are probably the same discussed for AsAc contents and involve the longer shelf life period of 1-MCPt fruit.

#### Tomato sugar and sugar/acidity ratio

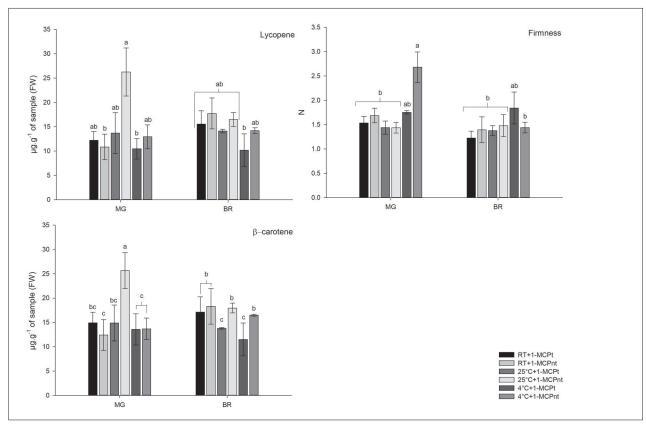
Despite the earlier ripening stage and 1-MCP treatment, MG tomatoes accumulated more TS and RS compared to BR tomatoes in the same conditions (Figure 2). This result is in contrast with similar experiments (at lower temperatures, 13 °C and 18 °C) where BR tomatoes treated with 1-MCP accumulated more soluble solids than their MG counterparts (Baldwin  $et\ al.,\ 2011$ ). The higher TS accumulation occurred in RT + 1-MCPt with 377.8 mg g¹ in a fresh weight basis (FW). This result was highly influenced by the contents of fructose in this treatment. Reducing sugars repeated the same accumulation pattern with high contents in RT + 1-MCPt, with 40.7 mg g¹ FW and 38.4 mg g¹ FW for glucose and fructose, respectively. The higher glucose content was obtained in 25 °C + 1-MCPt, with 49.23 mg g¹ FW.



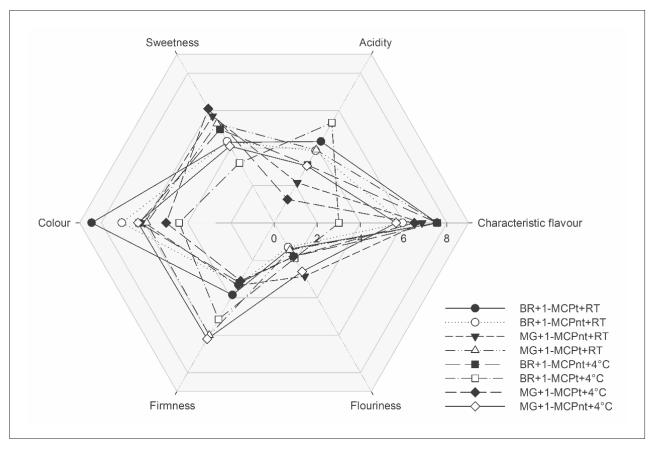
**FIGURE 1.** Contents of ascorbic acid and titratable acidity of tomatoes harvested at mature green (MG) and breaker stages (BR), treated or not with 1-MCP and stored at different temperatures. RT (Room temperature); 1-MCPt (1-MCP treated fruit); 1-MCPnt (1-MCP non-treated fruit). Means followed by the same letter do not differ significantly at  $p \le 0.05$  by Tukey's test. Error bars represent the Standard Error (SD) of the mean.



**FIGURE 2.** Total soluble solids, total sugars and reducing sugars (glucose and fructose) of tomatoes harvested at mature green (MG) and breaker stages (BR), treated or not with 1-MCP and stored at different temperatures. RT (Room temperature); 1-MCPt (1-MCP treated fruit); 1-MCPnt (1-MCP non-treated fruit). Means followed by the same letter do not differ significantly at  $p \le 0.05$  by Tukey's test. Error bars represent the Standard Error (SD) of the mean.



**FIGURE 3.** Contents  $\beta$ -carotene, lycopene and fruit firmness of tomatoes harvested at mature green (MG) and breaker stages (BR), treated or not with 1-MCP and stored at different temperatures. RT (Room temperature); 1-MCPt (1-MCP treated fruit); 1-MCPnt (1-MCP non-treated fruit). Means (n=3) followed by the same letter do not differ significantly at  $p \le 0.05$  by Tukey's test. Error bars represent the Standard Error (SD) of the mean.



**FIGURE 4.** Sensorial attributes of tomatoes harvested at mature green (MG) and breaker stages (BR), treated or not with 1-MCP and stored at different temperatures. RT (Room temperature); 1-MCPt (1-MCP treated fruit); 1-MCPnt (1-MCP nontreated fruit). Nine panelists (n = 9) assessed each attribute.

An important characteristic in tomato fruit acceptance is the sugar/acid ratio TSS/TA (Table 1); higher ratios are present at RT + 1-MCPt, irrespective of ripening stage. The 1-MCP non-treated fruits presented lower ratios in general, especially for MG fruits. Studies suggest that 1-MCP treatment alone is not capable of significant alteration in the TSS of fruit (Boggala, 2015). However, in some cultivars, 1-MCP may prevent the typical decrease in acids content that takes place during ripening, affecting the TSS/TA ratio and causing loss of quality with regard to sensorial parameters (Wills and Ku, 2002; Mir *et al.*, 2004; Opiyo and Ying, 2005; Cliff *et al.*, 2009). Only at RT the 1-MCP effect over TSS/TA ratio was significant due to a greater accumulation of acids in this treatment.

#### Tomato firmness

Fruit firmness presented a small variation among all treatments, with the smaller values found in fruit stored at RT and 25 °C in both MG and BR stages (Figure 3). Similar results were found by Brashlyanova *et al.* (2014), where tomatoes stored at 18–22 °C were less firm than fruit stored at 12 °C. As expected, fruit kept at 4 °C+1-MCPt were firmer, but only for BR stage; interestingly MG + 4 °C + 1-MCPnt fruit were significantly firmer (2.68 N) than 1-MCPt counterparts (1.755 N).

Maturation caused a slightly softening of tomato fruit during storage, irrespective of temperature or 1-MCP treatment, when compared to unripe samples (data not shown).

Since fruit firmness was measured at the end of the ripening period, the effect of 1-MCP and temperature of storage was not clearly detected, considering that the delay in

softening is commonly detected on the first days after harvest; as ripening develops, all fruit eventually softens. Park  $et\ al.\ (2016)$  describe the same pattern; after 9 days of storage, 1-MCP-treated tomatoes (1,000 ppb) were firmer than control, however after the  $15^{\rm th}$  day the differences were not significant any longer. According to Batu (2004), firmness values above 1.28 N are suitable for household preparation (mainly as salads) and are widely accepted by consumers, those with firmness above 1.46 N are very firm and suitable for supermarket (customer handling).

# Tomato color

Fruit color parameters presented substantial variation during the storage period (Table 1). Chroma, defined as the purity of a color, was higher in MG tomatoes kept at RT + 1-MCPt (47.0) as well as fruit stored at 4 °C + 1-MCPt (49.2), high values indicate color intensity. Fruit at BR stage stored at RT and 25 °C and 1-MCPt presented the higher chroma values of 42.4 and 43.0, respectively. As ripening developed during storage period, a natural change from green to yellow-red was observed and measured as hue angle (0° = red;  $30^\circ$  = orange;  $60^\circ$  = yellow;  $90^\circ$  = lime and  $120^\circ$  = green). The smaller values were obtained in MG fruit stored at 25 °C + 1-MCPt (57.0) and 25 °C + 1-MCPnt (49.6); on the other hand, MG tomatoes kept at 4 °C tended to be more yellow with hue angles of 85.0 (1-MCPt) and 82.4 (1-MCPnt).

Measures indicated a decrease tendency in lightness when compared with fruit at moment of the harvest (data not shown). Smaller values were obtained in 1-MCPt fruit on all treatments, except MG + 4  $^{\circ}$ C, indicating a beneficial

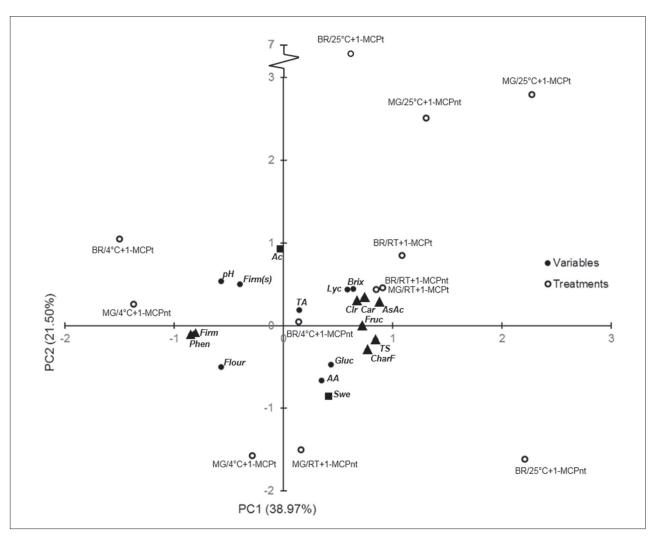


effect of 1-MCP application over maturation. Literature indicates that more advanced ripening stages, when submitted to 1-MCP treatment, tend to respond better, resulting in superior fruit quality, whilst the treatment of green fruit may result in internal and external deformities (Mir *et al.*, 2004). Zhang *et al.* (2009) demonstrated an accentuate decrease in the hue angle until the 16th day after harvest in 'Sebring' tomatoes treated with 1-MCP; the values obtained (41.1°) are similar to those determined in this study. This reduction did not develop in the first days of storage where the hue angle values were still high (87.9°), taking place only after the 6th day. These high values detected in the early harvest are in conformity with the data obtained in this study.

## Tomato phenolic content

Carotenoid contents (Figure 3) varied mainly as a function of temperature; MG tomatoes stored at 25 °C + 1-MCPnt had greater concentrations (25.64  $\mu g \ g^{-1} \ FW$ ) and smaller concentrations were a characteristic of fruit stored at 4 °C (13.56 and 13.68  $\mu g \ g^{-1} \ FW$ ) for 1-MCPt and 1-MCPnt, respec-

tively. Similarly to MG fruit, tomato harvested at BR stage accumulated more carotenoids at higher storage temperatures in absence of 1-MCP, with contents of 18.29 µg g-1 FW at RT and 17.96 µg g-1 FW at 25 °C. With exception of the treatment at 25 °C + 1-MCPnt, the overall  $\beta$ -carotene accumulation was smaller in tomatoes harvested at MG stage, as expected. The concentration of β-carotene was significantly influenced by the application of 1-MCP in MG + 25 °C treatments; 1-MCPt fruit accumulated 14.18  $\mu g$  g-1 FW of  $\beta$ -carotene, while 1-MCPnt accumulated 25.64  $\mu g \, g^{\mbox{\tiny -1}}$  FW. This influence is present, to a smaller extent, on BR tomatoes where 1-MCPt fruit accumulated 13.79 μg g-1 fw of β-carotene and 1-MCPnt accumulated 17.96 µg g-1 FW. Application of 1-MCP is known to delay color development in tomato fruit, however this effect can be restricted to lower temperatures and for a certain storage period. Mostofi et al. (2003) measured the lycopene contents of tomatoes stored at 25 °C; color development was impaired to the 18th day of storage by 1-MCP, after that period the contents are not different from control fruit (not treated with 1-MCP). At lower temperatures, (20 °C and 15 °C)



**FIGURE 5.** Principal component analysis (PCA) bi-plot of treatments and variables measured in tomatoes harvested at mature green (MG) and breaker stages (BR), treated or not with 1-MCP and stored at different temperatures. RT (Room temperature); 1-MCPt (1-MCP treated fruit); 1-MCPnt (1-MCP non-treated fruit).

**PC1** ( $\triangle$ ): *Phen* = Total phenolic compounds; *Firm* = Firmness measured instrumentally; *TS* = Total sugars; *Fruc* = Fructose; Car = Carotene; AsAc = Ascorbic acid; Car = Characteristic flavor; Car = Color in sensorial evaluation.

**PC2** ( $\blacksquare$ ): Ac. = Acidity in sensorial evaluation; Swe = Sweetness in sensorial evaluation. Not correlated to any of principal components ( $\blacksquare$ ): TA = Titratable acidity; Gluc = Glucose; Brix = °Brix; pH; AA = Antioxidant activity; Lyc = Lycopene; Firm(s) = Firmness in sensorial evaluation; Flour = Flouriness.

color development was impaired to the 18th and 24th day of storage. In the present study, BR fruit followed a similar pattern, lycopene contents were smaller in all 1-MCP treatments irrespective of storage temperature; the same was observed for β-carotene contents. As for MG fruit, interestingly, 1-MCPt treatments at 25 °C had greater contents of both lycopene and β-carotene. A pronounced interaction occurred in treatments stored at 25 °C, harvested at MG stage and treated with 1-MCP, where the contents of pigments were approximately 10  $\mu g \ g^{\text{--}1}$  bigger than the average of the other treatments. This could be explained by the differences in storage period (18 days for 1-MCPt and 11 days for 1-MCPnt); at 25 °C terpenoid contents tend to increase until the 12th day, suffering a relative decay in the following days of storage (Mostofi et al., 2003). Thus, a longer storage period of fruit treated with 1-MCP may have allowed a decay in lycopene and  $\beta$ -carotene contents, producing the observed differences.

Mature green tomato had a great increase in lycopene concentrations starting from approximately 3.0  $\mu g \, g^{\text{-}1} \, FW$  at the moment of the harvest (data not shown) to 26.25  $\mu g \, g^{\text{-}1}$  in 25 °C + 1-MCPnt fruit. The application of 1-MCP in this condition led to a decrease in lycopene concentration to nearly a half (13.69  $\mu g \, g^{\text{-}1} \, FW)$  compared to 1-MCPnt fruit. Breaker fruit, similarly to the patterns of carotene contents, accumulated more lycopene than fruit at the MG stage in all treatments, except at 25 °C + 1-MCPnt.

#### Sensorial attributes

Tomato characteristic flavor is of paramount importance for consumer acceptance and is linked to a complex mixture of compounds such as sugars, acids, and a pool of volatiles of approximately 16 molecules that effectively contribute for tomato flavor and odor (Tieman et al., 2012). Breaker fruit stored at 4 °C + 1-MCPt presented a marked flavor alteration, receiving low grades by the panellists (Figure 4). In the same treatment, with the absence of 1-MCP, no characteristic flavor alteration was observed. The flavor alteration is possibly related to the high acidity grades and low sweetness attributed to this treatment. The same patterns were reported by Maul et al. (2000) on tomato stored at 5 °C, resulting in significantly lower sweetness and characteristic flavor and higher acidity compared to tomato stored at 20 °C. On the other hand, MG fruit kept at 4 °C + 1-MCPt were considered the sweetest and less acid, demonstrating a great influence of ripening stage on fruit stored in the same conditions.

Breaker tomatoes stored at RT were best evaluated regarding fruit color, receiving the higher grades, while fruit in MG stage and those kept at 4  $^{\circ}$ C received the lower grades.

## Multivariate analyses

Principal component analysis (PCA) was carried out to cope with the drawbacks of univariate procedures as analyses of variance (ANOVA), characterized by lack of capability to show interrelationship between variables. We performed PCA analyses in order to group and correlate variables obtained by sensorial and physicochemical evaluations, as well as to associate the different treatments with variable or group of variables behavior. This procedure is able to reduce data dimensionality in a simpler analytical sub-space, where two uncorrelated variables (components) are obtained through an orthogonal transformation procedure (Jolliffe, 2002). Another reason for PCA analyses was to reduce possible variable redundancy originated from measurements of TS, glucose and fructose.

Most of the variables are more strongly correlated to

principal component 1 (PC1), which explains 38.97% of the total variability of the experiment, being responsible for most of the variation amongst different treatments. This component presents high coefficients of correlation with the variable ascorbic acid and total sugars; total phenolic content and firmness (instrumental measure) are negatively correlated to this principal component, making it a "non-firm" and "low-phenolic" component. In the positive abscissa axis are the treatments with the best level of ascorbic acid and total sugar and in the negative axis the opposite.

Principal component 2 (PC2), that explains 21.5% of the total variability of the experiment, is strongly correlated to fruit sensorial acidity and negatively correlated to sweetness, both variables attributed by panellists. The sensorial acidity loading score in PC2 is very high (0.94) making of PC2 an "acid component"; as for sweetness, the component score is -0.84, reinforcing the acid character of PC2. The variable titratable acidity does not contribute to the formation of PC2 indicating that the contents of acids are not the only responsible for the perception of acidity by panellists. It is well established that the perception of flavor is a combination of aroma volatiles and compounds responsible for the taste. The aroma of food is perceived by the olfactory system as the volatiles enter the nasal ducts at the back of the throat, named retronasal perception (Meilgaard et al., 2006). The acid flavor is not determined by acids contents alone but also by aroma compounds. Baldwin et al. (1998) found positive correlation between acidity (sourness) perception and the contents of hexanal and negative correlation with acetone. In a similar manner, ascorbic acid is highly correlated to PC1 and does not cluster with variables TA and acidity (panellist scores), instead, this variable increases together with TS, and fructose contents. Sweetness, as perceived by panellists, was not correlated to the contents of fructose.

In relation to the distribution of the treatments in the components, it can be observed that the first component (ascorbic acid/total sugars) clearly separates MG and BR treatments at 25 °C and RT, independent of the application of MCP, on the positive axis and MG and BR treatments at 4 °C, independent of the application of MCP, on the negative axis, confirming the data of the univariate analysis in which the low temperature negatively affects these variables.

Storage temperature seems to account for most of the data variation in the experiment, being strongly correlated to the principal component 1 (PC1), this component contains most of the data variation (38.97%). Treatments at RT showed similar behavior and grouped together with exception of MG/RT + 1-MCPnt; this treatment is characterized by lower contents of ascorbic acid (1.28 mg 100 g-1 FW) and consequent increase in sweetness perception, showed by the correlation of this treatment to the non-acid region of PC2 (-1.5 score). The low ascorbic acid content was also detected by panellists, who attributed a low grade (2) to the acidity attribute. This treatment counterpart (1-MCPt) presented a huge difference in shelf life and achieved complete ripening after 18 days, 7 days longer than 1-MCPnt fruit. As discussed before, the longer shelf life allowed fruit to accumulate more acids thus improving sensorial acceptability. It can be observed that the second component (sensorial acidity/sweetness) separates the MG treatment at 25 °C, independent of the application of 1-MCP, and BR 25 °C with MCP treatment on the positive axis and the MG and BR treatments at 4 °C and RT, independent of the application of MCP, on the intermediate or negative axis.

# Conclusion

The tomato fruits cv. Absoluto harvested in earlier ripening stages (mature green) are not necessarily fated to a poorer sensorial experience, as many reports indicate. For tomatoes stored at room temperature, the factor harvest stage had greater influence in the ripening period, mainly in 1-MCP treated fruits, where mature green fruits took 7 days longer to achieve complete ripening. This increase in storage period allowed a higher accumulation of ascorbic acid resulting in a more balanced flavor. Since household storage is usually made under lower temperatures (4 °C), this data can be readily applied for consumers information in order to obtain fruit with superior sensorial characteristics.

Fruits stored at room temperature did not present an accentuated loss of firmness, presenting values within an acceptable range in which costumers have good fruit acceptance. Fruit stored at 4 °C and treated with 1-MCP were highly affected by harvesting stage regarding the flavor fruit trait. In contrast with previous reports, fruit harvested at breaker stage presented poorer quality attributes such as flavor and sweetness. In the same conditions, mature green fruits were sweeter and less acid.

Our data demonstrate that early harvest of cv. Absoluto is not necessarily detrimental in relation to sensorial quality. Further studies with different maturation inhibitors, concentrations and cultivars/hybrids, as well as different cropping systems, are necessary in order to know if the pattern observed in our study is replicable.

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