Sucralose-mediated effect during post-harvest of summer squash (Cucurbita pepo L.) cv. Ambassador from organic cultivation, using both systems active and passive modified atmosphere

Efecto de la adición de sucralosa en post cosecha en zapallos orgánicos (Cucurbita pepo L.) cv. Ambassador utilizando atmósfera modificada activa y pasiva

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The objectives of this study were to evaluate the behavior of the use of sucralose to 1% in post-harvest fruits of summer squash (Cucurbita pepo L.), cv. Ambassador organically grown and stored under active and passive modified atmosphere (AMA and PMA), from two different pre-harvest conditions, with mulch and without mulch on the incidence in sensory evaluation of attributes. Each trial consisted of 3 treatments: T0 corresponding to the control, packaging under normal atmosphere, T1 packaged in AMA (20% of CO2 injection and 80% N) + sucralose 1% and T2 packaging for PMA + sucralose 1% without injection of gas, the fruits being evaluated in the measurement of gases at 12 and 25 days of refrigerated storage. As for the sensory analysis there was a negative effect of sucralose, being better evaluated treatment T0. Appearance results do not match those of acceptability, but still the treatment of active modified atmosphere (T1) was better than that evaluated with passive modified atmosphere (T2), indicating that the active modified atmosphere may help maintain the appearance of fruit and thus a better acceptability. Therefore, according to the results there was no positive effect of sucralose in the preservation of sensory attributes of summer squash.

Keywords:
Cucurbita pepo L.
Sucralose
Modified atmosphere
Sensory evaluation

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RESUMEN
Los objetivos del presente estudio fueron evaluar el comportamiento de frutos de zapallos italianos (Cucurbita pepo L.) cv. Ambassador, cultivados de forma orgánica almacenados en modificación de atmósfera MAP y SAP con la adición de sucralosa al 1% en postcosecha, provenientes de dos diferentes condiciones de cultivo; con y sin mulch y su incidencia en atributos de evaluación sensorial. Cada ensayo consistió de 3 tratamientos: T0 correspondiente al control, almacenamiento bajo atmósfera normal, T1 envasado en MAP (inyección 20% de CO2 y 80% de N) + sucralosa 1% y T2 envasado en SAP + sucralosa 1% sin inyección de gases, siendo evaluados los atributos sensoriales de los frutos y el contenido de gases de los envases a los 12 y 25 días de almacenamiento refrigerado. Los resultados de la apariencia de los frutos por los panelistas no fueron diferentes a la aceptabilidad, pero la fruta con el tratamiento de atmósfera modificada activa (T1) fue mejor evaluada que aquella con modificación pasiva de atmósfera (T2), indicando que la modificación de atmósfera activa puede ayudar a mantener la apariencia de los frutos y con una mejor aceptabilidad. Sin embargo, por lo estudiado, no existió un efecto positivo de la sucralosa en la conservación de los atributos sensoriales del zapallo italiano.

Palabras claves: Cucurbita pepo L., sucralosa, modificación de atmósfera, evaluación sensorial.

INTRODUCTION

The planted area with summer squash (Cucurbita pepo L.) in south-central Chile, according to the Census of Agriculture and Forestry VII (2007), is 996.3 ha, found mainly in the Metropolitan Region, Valparaíso Region V, and Maule Region VII. Despite being produced throughout the year, both in greenhouses and in the open air, it is a crop of middle importance with most of the production going to supply the domestic market, mainly from October to April (ODEPA, 2010).

The “mulch” or padding is a technique practiced for many years by farmers and involves placing a material on the ground to form a deck, in order to protect the crops and the soil itself from weather erosion and water evaporation. This layer also prevents certain crops from being frozen or damaged by contact with the ground (Escobar, 1990). Sucralose can also be used to...
improve the condition of maturity of fruits, by covering the fruit with a layer that complements the cold storage and thus extends the post-harvest life. Its principle of operation is based on the restriction of the permeability of fruits to those gases involved in the maturity process, acting as a modified atmosphere. Both techniques may contribute to diminish the loss of water in the growing fruits, as well as to maintain the fresh squash quality.

The present study was based on the following hypothesis: The application of 1% sucralose (C<sub>12</sub>H<sub>19</sub>C<sub>12</sub>O<sub>8</sub>) and the use of modified atmosphere in post-harvest evaluation might improve the sensory perception of summer squash cv. Ambassador, organically grown, with and without mulch. Therefore the aim of the study was to evaluate: 1) the use of sucralose in post-harvested summer squash cv. Ambassador, organically grown and stored under modified atmosphere AMA (active modified atmosphere) and PMA (passive modified atmosphere), 2) the carbon dioxide and oxygen concentration in bags with selective permeability containing summer squash cv. Ambassador, and 3) the sensory attributes on summer squash for fresh consumption, such as taste, color, aroma and texture, and acceptability.

MATERIALS AND METHODS

The test was conducted at the Universidad Católica del Maule, San Isidro’s Campus, Los Niches, Curicó, Maule Region, Chile, at geographical location 35° 02’ S, 71° 19’ W (225 meters above sea level).

The climate in this area is lower temperate mesothermal stenothermic semi-arid Mediterranean. Temperatures range from a high in January of 27.5 °C and a minimum in July of 4.1 °C. The frost-free period is 219 days, with an average of 12 frosts a year, still relatively moderate. It registers 1,380 degree days annually and 1,472 hours of chill. Hydric regime has an annual average rainfall of 859 mm, a water deficit of 883 mm and a dry period of 7 months (Santibañez and Uribe, 1993).

The soil of San Isidro’s campus, corresponds to the type of very fine sandy loam, Romeral series is alluvial, presents a flat topography and moderately rapid permeability and well drained (CIREN, 1997).

The fruit of summer squash (Cucurbita pepo L.) cv. Ambassador, which was established by direct seeding on the campus of the University, in an area of 660 m<sup>2</sup> being distributed in four ridges of 110 m long and 90 cm wide. Of these, two implemented ridges were covered with black polyethylene based mulch. For subsequent measurements performed on post-harvest, summer squash cultivation was handled with an organic production system. Fruit harvesting was manually performed during the morning in February to avoid high temperatures. Summer squashes were harvested from the second cutting, being used according to their size, which works as a physiological maturity index, with a homogeneous size of 12 to 15 cm long and with an average weight of 110 g, particularly selecting healthy fruits, with no visible damage, being harvested in separate rows with and without mulch. SURFRUT provided the plastic boxes which had been previously identified.

Washing of the fruits was performed by dipping them for 2 minutes in 4 ppm of hypochlorite solution, waiting an additional 30 minutes to allow for sanitizing effect before drying with absorbent paper towel. There were two trials (A<sub>1</sub> and A<sub>2</sub>), which corresponded to a trial with summer squashes organically grown with and without mulching, respectively.

There were three treatments: a control (T<sub>0</sub>), and two treatments (T<sub>1</sub> and T<sub>2</sub>) for each of the tests, with three replicates:

- Treatment T<sub>0</sub>: control treatment, with packing of fruit in a Styrofoam tray under normal atmosphere.
- Treatment T<sub>1</sub>: AMA plus 1% Sucralose applied on the fruits. A bag, brand San Jorge FF Plum 604, was used. To this bag, a mixture of 20% CO<sub>2</sub> and 80% N<sub>2</sub> was applied by injection with a Tagler machine and then sealed using a pressure of 4 bar.
- Treatment T<sub>2</sub>: PMA plus 1% Sucralose applied on the fruits. A bag, brand San Jorge FF Plum 604 was used. No application of gases was done and it was sealed with a Tagler machine using a pressure of 4 bar.

All treatments were applied equally to the summer squashes, obtained organically with and without mulch. Each package contained 3 summer squashes and the fruits for treatments T<sub>1</sub> and T<sub>2</sub> were immersed in a 1% solution of sucralose for a period of five minutes, and then allowed to drain. This process was applied equally to the summer squashes obtained from non-organical cultivation with and without mulch.

After applying the treatments, the squashes were stored in a cold chamber at an experimental temperature of 7 to 8 °C and 95% relative humidity for a period of 25 days. The bags of each treatment were placed in their respective container (RentaPack plastic boxes).

Two measurements were taken, at 12 and 25 days of storage, as specified in the flow chart (Figure 1). Evaluations, both sensory and instrumental, of treatments T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub> for both assays, with and without mulch, consisted of three repetitions each, in which different parameters were measured at 12 and 25 days of refrigerated storage. The gas measurement was performed at the fruit company Copefrut, in Romeral, south-central Chile. A portable checkpoint (CO<sub>2</sub>/O<sub>2</sub>) PBI sensor was used for determining contents of CO<sub>2</sub> and O<sub>2</sub> in percent for each treatment under AMA or PMA.

Using sensory evaluation guides, both structured and unstructured sensory analysis were performed,
Physiological maturity of summer squash (12 to 15 cm)  
(Basic days, reached 55 to 75 days)

Crop (February)

With mulch/ without mulch

Washing 4 ppm hypochlorite (2 min)

After 3 hours, equilibrium at room temperature

Treatments applied

Immersion in 1% sucralose (5 min, change solution 50% use)

Drying

Packaging (Experimental unit, bag with 3 summer squashes)

T1 AMA                T0                T2 PMA
(Injection, mix of 20% CO2 + 80% N)                        (No application of gas)

Storing in chamber (7 to 8°C, 90 to 95 %H.R., for 25 days)

Measurements (12, 25 days)

Physical and chemical parameters and sensory attributes

Figure 1. Flow chart of the entire process of product application and subsequent summer squash packaging.

which included the participation of 14 trained panelists, who abstained from smoking, eating and drinking one hour before tasting. Furthermore, they also rinsed their mouth between the tasting of each sample, to avoid taste confusion. Panelists expressed their preference marking a cross on a 13 cm line of the unstructured guide, for the intensity of the attributes perceived: flavor, color, aroma and texture (Stone and Sidel, 1993). For evaluation of both appearance and acceptability, a structured-type guide with numeric scale from 1 to 9 was used, in which each panelist after his first impression had to answer how he liked or disliked the squashes typified under the different treatments (Wittig, 2001). A completely randomized design (C.R.D.) with factorial arrangement of 3 x 2 and a confidence level of 95% was used, considering the three factors: T0, T1 and T2, and two measurements at 15 and 25 days of refrigerated storage. For effects of multiple comparisons Tukey test was used, with a confidence level of 95%. The experimental unit contained three fruits in a bag with modified atmosphere.

RESULTS AND DISCUSSION

Oxygen Gas Measurements

Test A1

The percentage of O2 in container with squashes treated with active modified atmosphere (T1) and passive modified atmosphere (T2) with 1% sucralose application from a cultivation with mulch was compared and significant differences were observed (Figure 2).

For the post-harvest treatments, packages using passive modified atmosphere initially gave a higher percentage of O2 compared to canned squash. Howe-
ver, when considering the time spent in storage, a significant decrease in the percentage of $O_2$ was observed in the packages. These results are consistent with another study that found that the modified atmosphere directly influenced the squashes, increasing respiratory rate and decreasing the percentage of $O_2$ in containers similar to those used in this assay (Lucera et al., 2010).

Chakraverty et al. (2003), claim that techniques related to post-harvest such as modified atmosphere could inhibit ethylene action to some degree by reducing respiration rate of the fruit. Squashes are non-climacteric vegetables, therefore rate of respiration are not increased after harvest by response to ethylene biosynthesis. Because of this, it was found that the difference in the stock $O_2$ was significant, this is likely because respiratory rate was significantly different at 12 and 25 days of storage (Chakraverty et al., 2003). Also, according to Chakraverty et al. (2003), it is important to consider that the respiratory rate of summer squashes is 17 to 18 mL CO$_2$ kg$^{-1}$ h$^{-1}$ which is considered high, so that it could be estimated that both the active and passive modified atmospheres could help maintain the normal balance between CO$_2$ and $O_2$ through oxygen levels present in the containers.

**Test A$_2$**

When comparing the percentage of $O_2$ in the packages with squashes treated either with active or passive modified atmosphere, from cultivation without mulch, significant differences were observed (Figure 2). By observing the behavior of fruits without mulch, it could be said that the coincidences in terms of trends in the conditions of pre-harvest, mean that there was apparently an effect of active modified atmosphere on the balance of oxygen in the air inside the fruit container.

This is a consequence of the lower $O_2$ content present, since the percentage decreased between 12 and 25 days of storage, being lower than that for the squashes under passive modified atmosphere. Probably there was a lower respiratory activity with a subsequent decrease in oxygen consumption, which could translate in a lower degree of maturity of the fruit and therefore a better condition to be consumed as a fresh produce.

Lee et al. (1996), claim that the low density bags, as those used in this assay, are highly permeable to $O_2$, which would explain why oxygen concentrations shown by the squashes with passive modified atmosphere treatment (T$_2$) were higher than the contents of CO$_2$. One method to reverse this situation involves choosing a container that includes an exchange of oxygen similar to the product respiration rate, so that an increase in temperature, would provoke an increase in respiration rate and permeability of the film in an equivalent amount (Mir and Beaudry, 2009).

Decreased oxygen levels may also have other effects on the fruit, as for example, inactivating the enzyme reactions to induce color changes of the surface of the fruits, by provoking a reduction of the amount of $O_2$ in the package. The active modified atmosphere techniques can also reduce the discoloration of the fruit surface (Barry-Ryan et al., 2007).

**CO$_2$ gas measurement**

**Test A$_1$**

The squash treated with active modified atmosphere plus 1% sucralose (T$_1$) and passive modified atmosphere plus 1% sucralose (T$_2$), both at 12 and 25 days of storage, showed significant differences in the percentage of CO$_2$ (Figure 3). The squashes stored in active...
Sucralose addition in summer squash under controlled conditions

Figure 3. Percentage of average carbon dioxide in summer squash (Cucurbita pepo L.) cv. Ambassador, subjected to different post-harvest treatments, at 12 and 25 days of storage (A) with mulch and (B) without mulch.

Figure 3. Porcentaje promedio de dióxido de carbono de zapallos (Cucurbita pepo L.) cv. Ambassador sin mulch sometidos a diferentes tratamientos de post cosecha a 12 y 25 días de almacenamiento (A) con mulch y (B) sin mulch.

Modified atmosphere (T₁) had higher percentages of CO₂ compared to those treated with passive modified atmosphere (T₂), being probably lessened the respiratory rate in the first due to increased CO₂ levels during storage (Kader, 2002).

In analyzing the effect of storage time on post-harvest squashes, significant differences were observed, which were linked to a positive trend in the percentage of CO₂ that increased by about 3 percentage points. The creation and maintenance of an optimum atmosphere inside the modified atmosphere packages would depend on the product respiratory rate and the film permeability to CO₂, which is affected by temperature (Kader, 2002).

These authors obtained similar values to those presented here using a container made of organic polymers.

Test A₁

When comparing the percentage of CO₂ in the packages of squashes from a crop without mulch as a condition of pre-harvest and post-harvest treatments (T₁ and T₂), a significant difference was obtained (Figure 3).

If only post-harvest treatments are analyzed, squashes that exhibited higher percentages of CO₂ in the package, were those treated with active modified atmosphere plus 1% sucralose (T₁), coinciding in their tendency with the pre-harvest use of mulch.

In analyzing the effect of storage time on the content of CO₂ in squash package, an increase of nearly 3% was observed after 25 days storage, coinciding with what happened when using mulch as pre-harvest cover.

Sensorial analysis

Acceptability assays A₁ and A₂

Comparing summer squashes subjected to various post-harvest treatments for the entire storage period, significant differences in acceptability were found for both cases of squash cultivated with and without mulch (Figure 4). In analysis of post-harvest treatments for both assays, with and without mulch, a similar trend was observed; panelists evaluated the fruits from the control treatment (T₀) with the highest score, with a result closet score 7.

Squashes treated with active modified atmosphere plus 1% sucralose (T₁) were evaluated with a score 6. The fruits with lowest evaluation (score 4) were those treated with passive modified atmosphere plus 1% sucralose (T₂). Paliyath et al., (2008), state that the acceptability is not a factor that is determined by the conditions of post-harvest, but is a factor essentially genotypic specific to the species being treated and is also closely related to the amount of organic acids and sugars present in the fruit, so that the acceptability may be related to the soluble solids and pH.

Lucera et al. (2010), corroborate the results obtained in this experiment. They stated that the active modified atmosphere treatments (T₁) could help keeping summer squash acceptability, better than the passive modified atmosphere treatments (T₂).

By comparing the evolution of the values of acceptability in time, no significant differences in both trials were observed, although there was a small decline in the acceptability from 12 to 25 days of refrigerated storage. These results were also observed by Lucera et al. (2010), in a study involving squashes, noting a decrease in acceptability with storage time.
By examining the results of the sensorial analysis, there was probably a negative effect of sucralose on the acceptability of the fruits (T₁ and T₂), because the squashes under treatment with sucralose showed lower acceptability than those under the control treatment (T₀).

**Appearance tests, A₁ and A₂**

Appearance is used along the production chain as the primary means of judging the quality of individual units of product. Product appearance is characterized mainly by absence of defects, size, shape and color (Kays, 1997).

When comparing the appearance of squashes from different post-harvest treatments (T₀, T₁, and T₂), after 25 days of refrigerated storage, significant differences were found, particularly in the trial with fruits from a cultivation with mulch (Figure 5).

For all post-harvest treatments, the squashes treated with active modified atmosphere plus 1% sucralose (T₁) were the best evaluated with a score 7. The squash not subjected to any treatment (T₀) were received a score of 6 from the 1-9 scale.

Squashes with least acceptance in terms of appearance were those where passive modified atmosphere plus 1% sucralose (T₂) was applied. According to these results, no significant effect of sucralose on the appearance of the fruit was observed. At the same time, there was no clear response from the panelists regarding the treatment used and evaluated throughout storage.

In analyzing the appearance of the fruit during storage, it was observed that there were no significant differences in the responses of the panelists for either of the two treatments, with values or scores of 7. According to Paliyath et al. (2008), the appearance of the plants should decline as a result of maturation, this did not happen in this study, where appearance remained constant, in the opinion of the panelists. Appearance results did not match those of acceptability, but still the treatment of active modified atmosphere (T₁) was better than that evaluated with passive modified atmosphere (T₂), indicating that the active modified atmosphere may help maintaining the appearance of the fruit and thus a better acceptability.

**Color assays, A₁ and A₂**

Color of squash from cultivation with mulch from the different post-harvest treatments (T₀, T₁, and T₂) showed no significant differences throughout the storage period. However, significant differences in color were found for those squashes in cultivation without mulch (Figure 6).

Fruits from cultivation without mulch, subjected to the three treatments (T₀, T₁, and T₂) and subsequent cooking, and presented for sensorial analysis in the form of slices, did not show significant differences in the green color of their skin and pulp. After 25 days of storage, darker shades of skin and flesh were observed (and fruits of treatments T₀, T₁, and T₂ received a score of 6 points on a scale of 1-13).

Comparing the results of sensorial analysis against those obtained for color by instrumental determination, as described previously, no significant correlation was found. Although darker shades in squashes receiving treatments (T₁ and T₂) were detected by sensorial analysis, as compared to those that did not undergo any
Sucralose addition in summer squash under controlled conditions

Figure 5. Average appearance of summer squash (*Cucurbita pepo* L.) cv. Ambassador, subjected to different post-harvest treatments, at 12 and 25 days of storage. Figure A: Fruits from a culture with mulch. B: Fruits from a culture without mulch.

Figure 6. Average color of summer squash (*Cucurbita pepo* L.) cv. Ambassador, treated with different post-harvest treatments, at 12 and 25 days of storage. Figure A: Fruits from a culture with mulch. B: Fruits from a culture without mulch.

Post-harvest application ($T_0$), the color parameter "b" of instrumental determination showed darker fruits only for treatment $T_2$. On the contrary Lucera *et al.* (2010), reported that sensorial analysis revealed darker color in squashes treated with passive modified atmosphere over those treated with active modified atmosphere.

**Olfactory assays, $A_1$ and $A_2$**

Squashes from different post-harvest treatments ($T_o$, $T_1$, and $T_2$), and different pre-harvest conditions, with and without mulch, were subjected to olfactory sensorial tests. In the fruits from pre-harvest condition with mulch, no significant differences in the responses were found, whereas in fruits from cultivation without mulch, significant differences were detected (Figure 7), yielding values close to strong aromas, for those fruits grown without mulch and values similar to soft aromas for those from assays with mulch.

When comparing only the post-harvest treatments ($T_o$, $T_1$, and $T_2$), the strongest flavors were detected in squash of control treatment ($T_o$). The squashes
with lower values for aroma were those to which the
treatment of active modified atmosphere with added
sucralose (T1) was applied. Furthermore, the applica-
tion of a film with sucralose could reduce the respi-
ration rate and prevented to some extent gas exchange
from the fruit, thus decreasing the ripening process.
This would lead to maintain the characteristic aroma
of squash (Kader, 2002).

In assessing the performance of squashes during post-
harvest storage, no significant difference was observed
between 12 and 25 days of storage for both assays with
and without mulch, since sensorial analysis resulted in a
similar aroma perception in the fruits analyzed.

**Texture assays, A1 and A2**

Sensorial analysis of the squashes in all three
treatments (T0, T1 and T2) and from cultivation with
mulch, showed no significant differences in texture
attribute. Significant differences in the response were
found for fruits grown without mulch (Figure 8).

Among the three treatments applied to the fruits, the
control treatment (T0) received the best score for texture
(score 6 on a 1-13 scale), indicating a rather crunchy
texture. Therefore, squashes from passive modified at-
mosphere treatment plus 1% sucralose had higher tex-
ture values, compared to squashes from active modified
atmosphere treatment plus 1% sucralose. Moreover,
it was observed that the application of sucralose may
improve texture, giving the fruits values closest to the
 crunchiness (score 4 on the 1-13 scale). Storage time
cauied a significant increase in the score value for the
texture. From 12 to 25 days of storage score value for
texture increased to values typical for a crispy product.

**Flavor assays, A1 and A2**

The squashes from the assays with mulch (A1), sub-
jected to different postharvest treatments (T0, T1 and
T2) did not show any significant difference in flavor,
as compared to those from pre-harvest without mulch
(A2), where significant differences in the taste of the
fruit were observed (Figure 9).

Analyzing only the post-harvest treatments (T0, T1
and T2) for the assay without mulch, significant diffe-
rences in flavor were observed, wherein squashes from
the control treatments (T0) showed the highest values
with respect to the attribute flavor. Intensity in flavor
of the tested squashes diminished from treatments T1
to T2. Probably, sucralose had no effect on flavor attrib-
ute, since the fruits from the control treatments were
those that obtained score values denoting a more in-
tense flavor compared to the other two treatments.
This may be because squashes underwent cooking for
1 minute, which probably allowed added sucralose to
drain away from the fruit.

**CONCLUSIONS**

Treatments that best retained the level of gas in
containers with treated squash, were those from
treatment Tp, with slight increase in percentage of CO2
and slight decrease in percentage of O2.

The results of sensorial analysis showed that sucra-
lose had a negative effect on acceptability, appear-
ance, color, flavor, aroma and texture, since treatments
where sucralose was administered (T1 and T2) were
rated with lower score values compared to the control
treatment.
Figure 8. Average texture of summer squash (Cucurbita pepo L.) cv. Ambassador, subjected to different post-harvest treatments, at 12 and 25 days of storage. Figure A: Fruits from a culture with mulch. B: Fruits from a culture without mulch.


Figure 9. Average flavour of summer squash (Cucurbita pepo L.) cv. Ambassador, subjected to different post-harvest treatments, at 12 and 25 days of storage. Figure A: Fruits from a culture with mulch. B: Fruits from a culture without mulch.


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