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Modified Atmosphere Packaging of Nopal (Prickly Pear Cactus Stems, Opuntia spp.)

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Abstract

Prickly pear cactus stems (nopal or nopalito) are widely consumed in Mexico for their nutritional and health benefits, and also exported to some other countries. The effect of passive or semi-active modified atmosphere packaging (MAP) on the physico-chemical and microbiological characteristics of nopalitos was determined during storage at 5°C and 85% RH for up to 35 days. In semi-active MAP we have injected elevated pressures of CO₂ (20, 40 or 80 kPa) in the packages immediately after sealing. Passive atmospheres (where no CO₂ was added) were created with respiration gases and film permeability. Semi-active MAP with initial pressures of 40 or 80 kPa CO₂ increased the losses in texture, weight, chlorophyll content, dietary fiber content and color. Passive MAP, which developed an atmosphere of up to 8.9 kPa O₂ and up to 7 kPa CO₂, and semi-active MAP with an initial 20 kPa CO₂ significantly decreased losses in all the quality parameters analyzed and in microbial population (total aerobic mesophiles, moulds and yeast counts), but slightly increased the total anaerobic mesophiles counts. The microorganisms identified were not pathogenic. Therefore, fresh prickly pear cactus stems can be stored for up to 32 days in MAP with CO₂ pressures ≤20 kPa without significant losses in quality, nor any significant increases in microbial population.

INTRODUCTION

Nopal or nopalitos, the young cladodes of prickly pear cactus stems (Opuntia spp.) are low in calories, high in fiber, are traditionally consumed as a vegetable in Mexico and are exported to some other countries (Guevara and Yahia, 2000, 2003, 2005). Nopal is commonly consumed as a broiled, blended or made into a juice. It is used in various pharmaceutical applications for its therapeutic, dermatological and medical properties. Experimental evidence demonstrated that the ingestion of some species of nopal (Opuntia streptacantha, O. ficus-indica) could decrease both blood glucose levels and blood lipid levels in patients with non-insulin dependent diabetes mellitus (NIDDM) (Frati et al., 1983). They are marketed either as wholes or in slices. The stems are very perishable, being very sensitive to water loss, darkening and decay (Guevara et al., 2001, 2003). Storage life of nopalitos is one day at room temperature and up to 6 days in refrigeration. The objective of this work was to determine the effects of passive and semi-active MAP (with initial pressures of 20, 40 and 80 kPa CO₂), and holding at 5°C for up to 35 days, on the storage life and quality of prickly pear cactus stems.

MATERIALS AND METHODS

Young cladodes (Opuntia ficus-indica cv. Milpa Alta) were obtained from a commercial plantation in Milpa Alta, Mexico. The cladodes (about 15 cm long) were precooled in chlorinated water (100 ppm) at 4°C for 15 min, and the cut zones were immersed in ascorbic at 100 ppm. Cladodes were then left to dry for about 30 min in regular air and classified into 5 lots of 180 cladodes each. One lot was left without packaging in polymeric bags, packed in a carton box in the same way that are commonly marketed and was considered as control. The other cladodes were loosely packed in Cryovac RS425 bags (2 stems of about 200 g/bag). Cryovac RS425 bags had dimensions
of 30 x 20 cm and a volume of 2 liters. The film had a thickness of 0.032 mm, and gas transmission rates were 4,000–6,000 ml O₂, m⁻², day⁻¹, atm⁻¹, and 18,000–20,000 ml CO₂, m⁻², day⁻¹, atm⁻¹. Water vapor transmission was 0.5–0.8 g H₂O. 100 m⁻², 24h⁻¹, atm⁻¹. One lot was packed in these bags, sealed and considered as the passive MAP (without adding CO₂). The third, fourth and fifth lots were placed in the same bags and different pressures of CO₂ (20, 40 and 80 kPa, respectively) were added in each bag immediately after sealing (Yahia, 2004; Yahia et al., 2004; Yahia and Gonzalez, 1998; Yahia and Rivera, 1992). These were considered as the semi-active MAP treatments. Packaged and non-packaged cladodes were held at 5°C and 80% RH for up to 35 days in the dark.

In-package atmosphere (O₂ and CO₂) was measured initially and each 7 days, using a Nitec Model GA-20 portable O₂/CO₂ analyzer. The O₂ sensor is an electrochemical fuel cell with a range of 0–100 kPa and accuracy of ±0.5 kPa of full scale. The CO₂ sensor is a non-dispersing infrared sensor with a range of 0–100 kPa and accuracy of ±0.2 kPa of full scale. Gas samples were taken by a syringe from the inside of the package through a rubber septum glued on a surface of the package film.

Overall quality was evaluated on the basis of a subjective scoring system (9 = excellent quality, 7 = good, 5 = intermediate, 3 = low, and 1 = very low). Surface color of the stems was measured with a portable Minolta CM-2002 chromameter. The maximum force (N) to rupture pulp tissue was determined with a stable micro system TA-HD texture analyzer. Dietary fiber content was determined using the neutral detergent reagent method (Guevara et al., 2001, 2003). Chlorophyllase activity was determined by measuring the formation of chlorophyllides (Fernandez et al., 1992). Protein content was measured according to the dye binding method of Bradford (1976) using serum albumin for the standard curve. One unit of enzyme activity was defined as the formation of one nmol of chlorophyllide/min/mg protein. Chlorophyll content was determined according to Mencarelli and Saltveit (1988) with some modifications.

Microbiological counts (total aerobic mesophiles, total anaerobic mesophiles and yeast and molds) were carried out using established methods on 10 g homogenized samples that were macerated in 90 ml of sterile peptone water (pH 7.4) (Sneath et al., 1986). Plate count was performed by preparing appropriate sample dilution.

RESULTS AND DISCUSSION

The in-package atmosphere changed rapidly (Fig. 1), probably as result of the high respiration rate of the cactus stems. O₂ pressure in the passive MA decreased and reached about 8 kPa after 35 days in storage (Fig. 1), while the CO₂ pressure (Fig. 1a) increased to about 7.5 kPa in the same period of time. In the semi-active 20 kPa CO₂ MA (Fig. 1b), the O₂ pressure decreased to about 13 kPa, and the CO₂ pressure decreased to about 13.5 kPa after 35 days in storage. On the other hand the CO₂ pressure in 40 kPa and 80 kPa CO₂ semi-active MAP (Fig. 1), decreased to about 15 kPa and 25 kPa, respectively, and the O₂ pressure slightly increased to about 15 kPa, at the end of the storage period. The CO₂ pressures created in the 40 and 80 kPa semi-active MAP were higher than the minimum tolerated levels for many fresh horticultural crops (Yahia, 1998). However, it decreased very sharply in the package, which was the reason for not causing major negative effects.

Texture decreased in all cladodes, but the decrease was faster in those that were kept in 40 and 80 kPa CO₂ semi-active MAP and cladodes that were maintained without packaging, and the least texture loss was observed in the passive MAP and in 20 kPa CO₂ semi-active MAP (Fig. 2a). A relation similar to texture was found in the changes of dietary fibers content. The highest loss in dietary fibers was observed in cladodes maintained in 40 and 80 kPa CO₂ MAP and in cladodes that were maintained without packaging, and the lowest loss was observed in the cladodes kept in passive MAP and in 20 kPa CO₂ MAP (Fig. 2b). It is possible that the loss in texture is caused by losses in fiber resulting in the softening of the cladodes. The retention in texture of cladodes maintained in passive MAP or in 20 kPa CO₂ MAP might be due to the positive effects of MA with intermediate CO₂ pressures.

Weight loss was least in cladodes that were packaged in passive MAP and in 20
kPa CO₂ MAP, and the rest of the conditions showed a high weight loss of more than 25% (Fig. 3a). There was a close relation between weight loss and the overall quality (Fig. 3b). Cladodes that were not packaged lost their brilliant green color appearance and became dull as the storage period increased. Quality was highest in cladodes that were packaged in 20 kPa CO₂ MAP, followed by those packaged passive MAP. ≤20 kPa CO₂ can decrease the rates of respiration and transpiration, resulting in low weight loss.

The L* value (Fig. 4a) slightly increased in cladodes maintained in semi-active MAP with initial 40 and 80 kPa CO₂ and in those maintained without packaging, for up to 25 days in storage. The lowest L* values were observed in cladodes packaged in passive MAP and in 20 kPa CO₂ MAP. The a* value (Fig. 4b) slightly increased in all conditions as a result of the loss in green color; however, the increase was slower in cladodes packaged in semi-active MAP with initial 40 and 80 kPa, and in cladodes that were not packaged. The b* value (Fig. 4c) increased, and the highest increase was observed in cladodes packaged in 80 kPa CO₂, followed by those maintained in 40 kPa CO₂, and by those kept without packaging until 25 d in storage, after which a decrease was observed. Darkening was present in cladodes packaged in 40 and 80 kPa CO₂ MAP, especially during the last period of the study, which might be due to enzymatic oxidation. External color of control cladodes that were not held in MAP changed from brilliant green to dull green.

The decrease in chlorophyll content (total, a and b) (Fig. 5) was the least in cladodes that were maintained in passive MAP and in 20 kPa CO₂ MAP, followed by those packaged in 40 and 80 kPa CO₂ MAP, and in those kept without packaging. Total chlorophyll and chlorophyll a of cladodes kept in passive MAP and in 20 kPa CO₂ MAP decreased about 23% and 35%, respectively, while elevated CO₂ pressures (40 and 80 kPa) caused a decrease of about 53 and 63%, respectively (Figs. 5a, b). Chlorophyll b content decreased about 57%, 58%, 37%, 80% and 99% for in cladodes that were not packaged, packaged in passive MAP, or packaged in 20, 40 or 80 kPa CO₂ MAP, respectively after 35 days in storage (Fig. 5c). Chlorophyll a was higher than chlorophyll b at all stages, however the degradation velocity of the two was similar.

Chlorophyllase activity increased in all cladodes as the storage period increased (Fig. 5d). The lowest activity was present in cladodes packaged in passive MAP and in 20 kPa CO₂ MAP, and the highest activity was found in cladodes packaged in 40 and 80 kPa CO₂ MAP, and in cladodes kept without packaging. The highest chlorophyllase activity present in treatments with elevated CO₂ pressures (semi-active MAP with initial 40 and 80 kPa) is probably due to the stress caused by CO₂ injury. There is a clear relation between chlorophyll degradation and chlorophyllase activity. Conditions that resulted in the highest chlorophyll degradation also had the highest rate of chlorophyllase activity. Chlorophyllase can be considered either responsible or not for chlorophyll degradation. However, the data support a role for chlorophyllase in chlorophyll degradation in prickly pear cactus stems.

Microbial population started to increase after 15–20 days of storage (Fig. 6). The least increase in aerobic mesophiles (AeM) population was in cladodes that were kept in 20 kPa CO₂ MAP followed by those maintained in 40 kPa CO₂ MAP, and the highest increase was in cladodes maintained in 80 kPa CO₂ MAP and in cladodes that were maintained without packaging (Fig. 6a). AeM population reached up to 2.2 x 10⁷ CFU g⁻¹ after 35d in storage. Anaerobic mesophiles (AnM) (Fig. 6b) were highest in cladodes that were held in 80 kPa CO₂ MAP and least in those held without packaging and in those held in 20 kPa CO₂ MAP. The highest AnM count was 2.1 x 10⁷ CFU g⁻¹. The highest increase in moulds and yeasts (MY) (Fig. 6c) was found in cladodes that were held in semi-active MAP with an initial of 40 kPa CO₂, followed by those maintained without packaging, while the least MY population was in cladodes maintained in 20 and 80 kPa CO₂ MAP. The highest MY population reached up to 5 x 10⁴ CFU g⁻¹. Isolation of single colonies was performed by randomly selecting the square root of the total number of colonies counted on each plate. Identification was conducted according to Bergey’s manual (Sneath et al., 1986). On the basis of the macro and microscopic characteristics of
AeM bacteria isolated, and the biochemical test done we identified bacteria of the genus *Leuconostoc*, *Pseudomonas*, *Microccocus* and *Bacillus*. The AnM were identified the same way as the AeM, and the bacteria found were of the *Ruminococcus* genus. In the case of MY we took in consideration the reproduction and hyphae form and macroscopic and selective growth, and on the basis of that we have identified molds from the genera *Absidia*, *Cladosporium* and *Penicillium* and the *Pichia* yeast, which produces necrosis of tissue. Microbial counts were low compared to those encountered in other fresh products. The effect of CA on micro-organisms is temperature dependent, with the inhibitory effect being less at higher temperatures. The bacteriostatic effect of CO₂ is not fully understood, but it could be due to lowering of the pH of growth media, the exclusion of O₂ by replacement of CO₂ or the acidification effect of CO₂ on the organism directly.

CONCLUSIONS

It is possible to extend the shelf life of prickly pear cactus stems. This can be achieved by generating an atmosphere with O₂ levels of up to 8 KPa and CO₂ levels of up to 7 KPa in passive MAP, or up to 20 KPa in semi-active MAP. CO₂ pressures between 7 and 20 KPa decreased the loss in color, texture, and fiber content, and decreased the chlorophyllase activity, and the use of MAP decreased the microbial flora on the stems. These benefits are due to atmosphere modification and not to increased humidity in the atmosphere (Guevara et al., 2001). On the other hand, elevated CO₂ pressures (≥40 kPa) caused injury in cladodes in comparison with non-packaged cactus stems. There were no significant differences between prickly pear cactus stems packaged in passive MAP or in 20 kPa packages. The relative limit of tolerance of prickly pear cactus stems to CO₂ was 20 kPa. The storage life of the cactus stems can be up to 32 d in passive MAP or in semi-active 20 kPa CO₂ MAP. Bacteria of the genus *Leuconostoc*, *Bacillus*, *Pseudomonas*, *Micrococcus* and *Ruminococcus* were identified in the microflora of *Opuntia ficus-indica* cactus in MAP. The moulds isolated were of the genus *Absidia*, *Cladosporium*, *Penicillium*, in addition to the yeast *Pichia*. No pathogenic micro-organisms were identified in the cactus stems.

LITERATURE CITED


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Figures

25
20
15
10
20 kPa CO2

80 kPa CO2

40 kPa CO2

Partial pressure (kPa)

20
10
5
20
10
5
20
10
5

Days at 5°C

Fig. 1. Changes in the in-package O2 (—O—) and CO2 (—●—) concentrations in passive and semi-active (20, 40 and 80 kPa CO2) modified atmosphere packaged prickly pear cactus stems stored at 5°C for up to 35 days.
Fig. 2. Changes in the texture (N) and crude fiber (%) in passive and semi-active modified atmosphere packaged prickly pear cactus stems stored at 5°C for up to 35 days. Control (●—●), passive MAP (○—○), 20% CO₂ (■—■), 40% CO₂ (▲—▲), 80% CO₂ (□—□).
Fig. 3. Changes in weight loss (%) and overall quality scores in passive and semi-active modified atmosphere packaged prickly pear cactus stems stored at 5°C for up to 35 days. Histogram bars are, from left to right, control (■), passive MAP (■), 20% CO₂ (■), 40% CO₂ (■), 80% CO₂ (■).
Fig. 4. Changes in the color (L*, a* and b* values) in passive and semi-active modified atmosphere packaged prickly pear cactus stems stored at 5°C for up to 35 days. Control (---), passive MAP (---), 20% CO₂ (---), 40% CO₂ (---), 80% CO₂ (----).
Fig. 5. Changes in chlorophyll content (total, a, b) and chlorophyllase activity in passive and semi-active modified atmosphere packaged prickly pear cactus stems stored at 5°C for up to 35 days. Control (●—●), passive MAP (○—○), 20% CO₂ (▼—▼), 40% CO₂ (▼—▼), 80% CO₂ (■—■).
Fig. 6. Changes in total microbial counts (aerobic mesorphiles, anaerobic mesorphiles, yeast and moulds) in passive and semi-active modified atmosphere packaged prickly pear cactus stems stored at 5°C for up to 35 days. Symbols are: control (——), passive MAP (——), 20% CO₂ (→), 40% CO₂ (←), 80% CO₂ (■).