

Biodegradable PLA (polylactic acid) hinged trays keep quality of fresh-cut and cooked spinach

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Abstract This work examines the effects of packaging using two different polymeric trays with hinged lids, polyethylene terephthalate (PET) and polylactic acid (PLA), on fresh-cut and cooked spinach (*Spinacia oleracea*). Samples were stored in a cold room for 16 days at 4 °C. Chemical (total pigments, total polyphenols, ascorbic acid, antioxidant activity), physical (water activity), technological (colour evaluation), sensorial (aroma, visual appearance and water accumulation) and microbial (total aerobic mesophilic and psychrotrophic counts) parameters were tested. Both polymeric trays maintained the overall quality of fresh spinach for 6 days but spinach stored in PLA trays maintained its flavour longer. A significant increase in total polyphenols, antiradical activity, total carotenoids as well as a decrease in ascorbic acid in fresh spinach was observed in the first 3 days of storage in both samples. Unfortunately, the PLA package accumulated condensed water. The total microbial load of fresh-cut spinach reached about 6.3–7.3 log CFU g⁻¹ within 8 days. Cooked spinach packed in PLA and PET polymeric hinged trays showed the same behaviour as fresh spinach in terms of quality and shelf life. In conclusion, PLA plastic hinged trays can be used for packaging fresh-cut and cooked cut spinach, but the problem of condensed water must be solved.

Keywords *Spinacia oleracea* · Packaging · Processing · Antioxidants constituents

Introduction

Vegetables play a significant role in the human diet and provide vitamins, fibers—and minerals essential for human health and growth. For this reason, in the last decade, their consumption increased in all the socioeconomic groups of people living in rural or urban areas (Kumar et al. 2004).

In Italy, in 2010, minimally processed products were leaders in food consumption growth, with an increase in purchases and a decrease in selling price, because they need no preparation. Spinach is a crop of high economic interest in Italy, with a total production of about 90,000 t in 2010. Sorting, washing, peeling, and slicing or shredding are all operations necessary for producing fresh-cut food. Generally, cutting destroys cell walls and membranes, reducing the shelf life and quality of ready-to-use products; commercial value of green leafy vegetables may be greatly reduced due to high rates of respiration and transpiration (Nobile et al. 2006). Leafy vegetables are highly susceptible to mechanical damage, as well as to bacterial and mould growth, which drastically reduce shelf life (Cantwell and Kasmire 2002). These phenomena may result in the development of strong unpleasant odours, decay, discolouration, and tissue softening (Watada and Qi 1999). Pre-packaged mixed salads have a high potential for contamination by *Listeria monocytogenes* (due to extensive handling during preparation) or for cross-contamination from the environment. The lack of any heating step prior to the consumption of such products places the emphasis on high-quality ingredients, hygienic manufacturing, appropriate shelf life and correct storage for maintaining product safety (Little et al. 2007). For this reason sanitizing operations for fresh-cut products become an integral part of the production process and the goal is to ensure the absence of pathogenic microorganisms and to minimize the presence of non-pathogens (Ferrari et al. 2010; Bartoloni et al. 2012).

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Temperature control also helps to maintain product quality because it affects the respiration rate of fresh-cut products: the difference in O₂ consumption rate and CO₂ evolution rate increased gradually with the increase in temperature (10–20 °C), with greater water vapour production (Botondi et al. 1996; Kaur et al. 2011a).

Packaging technology is an available method for reducing postharvest deteriorative processes on fresh products. Currently, many types of plastic films are available for fruit and vegetables; it is important to select an appropriate packaging film because a bad material shortens the shelf life of this kind of food. Generally speaking, polyethylene (PE) or polypropylene (PP) bags and polyvinylchloride (PVC) trays are used. European Union regulation n. 1580/2007 introduces the use of eco-friendly packages for fruit and vegetables: polyethylene terephthalate (PET), which is recyclable, and polylactic acid (PLA), which is compostable in accordance with EN 13432:2002 (UNI 2002) and derived from renewable sources; it has been considered one of the solutions for packaging materials (Rhim et al. 2007). Both PET and oriented PLA can be used as fresh food service containers (Auras et al. 2005). Beyond the great environmental advantages of biopolymers, the actual cost of these materials is a great obstacle to their use but the EU policy in terms of environmental protection with incentives for companies using biodegradable materials, opens the possibility of reducing the cost of this material.

Fresh-cut baby spinach has a very high respiration rate, and thus high oxygen permeability films or perforated ones are required to avoid the risk of anoxic concentration in the package (Gorny 1997; Wooster 1998). Piagentini and Güemes (2002) demonstrated that the type of packaging film affected off-odour development, while having no effect on visual and sensory attributes; the off-odour score of fresh-cut spinach packaged in PP film decreased faster than the off-odour score of samples packaged in low-density polyethylene (LDPE) bags. Kaur et al. (2011b) observed that after 4 days of storage for fresh-cut spinach, retention of chlorophyll, β -carotene, polyphenolic content and ascorbic acid was better in LDPE packages than in polypropylene (PP) packages with macroperforations. Perforated polyethylene packages combined with super atmospheric O₂ treatments were advantageous in reducing aerobic mesophilic growth and eliminating the possibility of post-processing contamination in minimally processed baby spinach (Allende et al. 2004). Bottino et al. (2009) found no differences in nutritional content and visual performance between fresh-cut and intact spinach, both stored in PET hinged containers for 162 h at 4 °C. Yommi et al. (2011) evaluated leaf water status, weight loss and colour retention of bunched spinach packaged in LDPE and oriented polypropylene (OPP) bags stored at 2 °C for 25 days. They found better physical and physiological properties for spinach packaged in OPP bags.

The aim of this work was to evaluate the quality of fresh-cut and cooked spinach packed in two different clamshell trays (PET or PLA) with hinged lids.

Material and methods

Plant material and sanitation

Fresh spinach produced by a farm in Central Italy was used in this study. After harvest, the plant material was stored in a cold room for rapid cooling in order to remove field and respiration heats. Spoiled or damaged leaves were detached by a clean cut at the neck with a sharp blade previously immersed in a 5 % sodium hypochlorite commercial solution. The fresh spinach was then washed with a sanitizer solution of 300 ppm, “Tsunami 100”, constituted by 15.2 % peroxyacetic acid and 11.2 % hydrogen peroxide (Tsunami™, Ecolab, Mendota Heights, MN) for 10 min. Afterward the spinach were superficially dried with airflow for 30 min.

The cooked spinach was boiled at 100 °C for 5 min and then superficially dried and cooled for 30 min before packaging.

Packaging

Fresh-cut spinach and cooked spinach (approx. 100 g) were packed using two types of trays (118×172×65 mm) with hinged lids as used commercially (clamshell package not sealed): one is a recyclable PET polymer used for the commercial packaging of fruits and vegetables and the other is a new PLA polymer produced by renewable sources, 100 % biodegradable and compostable (CoopBox Group SpA, Reggio Emilia, Italy). Packages were closed with a hinged lid, not tightly sealed, which permitted gas exchange, and were stored in a cold room at 4 °C in the dark for 16 days. All analyses were performed immediately at 0 day (harvest), 3, 6, 8, 10, 14 and 16 days of storage for fresh spinach and at 6 and 10 day for cooked spinach. Three packages were used for each sampling time.

Water activity (aw)

Water activity was monitored by placing a round piece of leaf, 3 cm diameter, inside an AquaLab series 3 (Decagon Devices Inc., Pullman, WA, USA) instrument, which measures water activity based on dew point. The AquaLab carried out a highly accurate reading in less than 5 min, with an accuracy of ± 0.003 aw.

Colour evaluation

Surface colour of spinach was measured using a Minolta CM-2600d colourimeter (Minolta CO., LTD, Japan). Four colour

measurements were performed, two on the upper side and two on the lower side of the leaves. Colour parameters (L^* , a^* , b^*) measured in the CIELAB colour space were recorded and the results were expressed as Hue angle ($H^\circ = \arctan b^*/a^*$).

Quantitative analysis of pigments

Quantitative determination of pigments (chlorophylls and carotenoids) was made through spectrophotometric analysis. 2.5 g of spinach were homogenized with distilled water and a 100 % acetone solution (4 mL H₂O + 20 ml acetone) using Ultra Turrax (Jankle & Kunkel IKA-Labortechnik Ultra Turrax T25, Germany). The homogenized solution was allowed to stand for 30 min in an ice bath and was then centrifuged at 13,600g for 15 min. 2 ml of supernatant was taken from each sample for absorbance determination at wavelength: 470 nm (carotenoids), 663.2 nm and 646.8 nm (chlorophyll a and b) through spectrophotometer (Perkin Elmer Inc. Lambda 25 UV/VIS Spectrometer, USA) using 80 % acetone (v/v solvent) as standard. The Lichtenthaler and Wellburn formulas (1983) were used for chlorophyll and carotenoid quantification. The formula equation used is (for acetone 80 %): $C_a = 12.21A_{663} - 2.81A_{646}$; $C_b = 20.13A_{646} - 5.03A_{663}$ and $C_{(x+c)} = (1000A_{470} - 3.27C_a - 104C_b) 229^{-1}$. The data were expressed as $\mu\text{g mg}^{-1}$ fresh spinach.

Determination of total polyphenols

Polyphenol extracts (three replicates for each package) were obtained through maceration of frozen spinach samples (solid/liquid ratio 1:15) in a hydroalcoholic mixture of methanol (50:50v/v), for 24 h at room temperature under constant stirring. Extraction was carried out in closed bottles. The next day, the sample was rinsed adding 5 ml of methanol water solution (50:50v/v). Extracts obtained (20 ml) were centrifuged at 2200 g for 5 min; consequently supernatant was removed and filtered through an 8 μm filter (Millipore S.p.a., France). Total polyphenols were determined using the Folin-Ciocalteu method (Singleton and Rossi 1965), which involves reduction of reagent by polyphenolic compounds with formation of a blue complex. Data were expressed as gallic acid equivalents (mg GAE 100 g⁻¹ fresh spinach). The equation of standard curve used is: $y = 118.44x$. Spectrophotometric readings were taken using a spectrophotometer (Perkin Elmer Inc. Lambda 25 UV/VIS Spectrometer, USA), which measures the absorbance of blue complex at 725 nm, using a methanol/water solution (50:50v/v) as a comparison.

Free-radical scavenging assay

The radical scavenging ability or hydrogen donating of the extract was monitored using the stable free radical DPP, following the method described by Contini et al. (2008),

modifying the procedure shown earlier by Brand-Williams et al. (1995). Different dilutions of crude extract or antioxidant (in 0.4 ml of methanol) were mixed in a 1 cm disposable cuvette with 3 ml of freshly prepared methanolic solution of DPPH (65 μM). Control test was prepared with 0.4 ml of pure methanol; its initial absorbance was between 0.716 and 0.720. The cuvette was capped and left to stand in the dark at room temperature for 180 min (time required to reach the steady state). At this time, the decrease in absorbance was measured at 515 nm against a blank of pure methanol, with a Perkin–Elmer Lambda 25 UV/vis. The percentage of remaining DPPH (%DPPD_{rem}) was calculated as follows:

$$\%DPPH_{rem} = \frac{DPPH_s}{DPPH_c} * 100$$

where DPPH_s was the DPPH concentration in presence of sample and DPPH_c was the DPPH concentration of the control, at $t = 180$ min. The radical scavenging was calculated as inverse of EC₅₀ (efficient concentration), which is the amount of pure extract necessary to reduce the initial 2,2-diphenyl-1-picrylhydrazyl (DPPH) concentration by 50 %. Results were expressed as mg of fresh weight mg⁻¹ of DPPH.

Ascorbic acid determination

Ascorbic acid was measured on fresh leaves through enzymatic bioanalysis (R-Biopharm Italia Srl), a colourimetric method that allows L-ascorbic acid determination in food. Data were reported as mg of ascorbic acid 100 g⁻¹ of fresh spinach.

Sensory quality evaluation

An unofficial sensory evaluation of the samples was performed by five people: lab members and the spinach grower and processor. Aroma (odour score) was assessed by using the procedure by Carvalho and Clemente (2004): 6 = no off-odour, 4 = very light off-odour, 3 = medium off-odour and 1 = strong off-odour; visual appearance was evaluated on a 10-point scale, 10: very pleasant to 0: not pleasant. Water accumulation severity inside the packages was evaluated on a 10-point scale; 0: leaves completely wet and water accumulation, 1: leaves and film moderately wet, 3: leaves moderately wet, 5: leaves and film slightly wet, 7: leaves slightly wet and 10: no water accumulation. During scoring, the intermediate values were also given to the samples depending on their perceived characteristics.

Microbiological analyses

About 10 g of spinach from each tray were homogenised in a Stomacher (mod. 4153-50, International PBI, Milan, Italy) for

2 min with 90 ml of peptone water. Serial dilutions (1:9) of each homogenised sample with peptone water were made in the same diluent and surface-spread in duplicate. Total aerobic mesophilic and psychrotrophic viable counts were determined using a plate count agar (PCA, Merck - Germany); plates were incubated at 30 °C for 48 h and 7 °C for 10 days, respectively.

Statistical analysis

Trials were carried out three times, with three packages analysed at each sampling time. Analysis of variance (ANOVA) was performed and significance was evaluated for $p < 0.05$. Mean values were compared using least significant difference (LSD) at the 0.05 probability level ($p < 0.05$) and significant differences are indicated on the figures and tables with letters. Calculations were performed with the statistical software Graphpad Prism 3.05 (San Diego, CA, USA).

Results and discussion

Fresh-cut spinach storage

Beside the high relative humidity inside the packages, the PLA-packaged spinach and PET-packaged one lost 1 and 3 % of weight, respectively. This difference can be attributed to package closure: both packages had hinged lids but the adherence of the lid over the lips of the trays was slightly different, which could have affected the maintenance of relative humidity inside the package. During the processing operations, which entail a washing step, hydration of samples recovered freshness, brightness and crispiness.

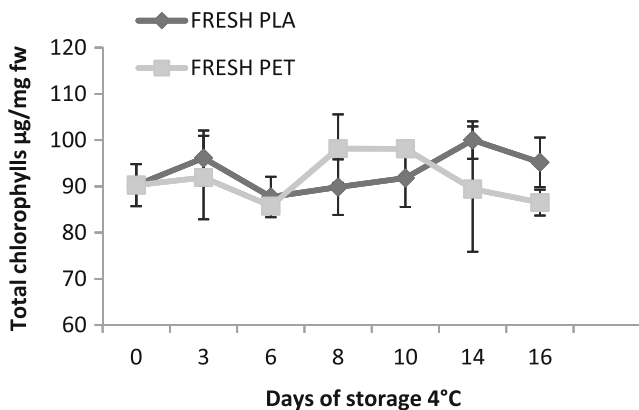


Fig. 1 Total chlorophylls of fresh-cut spinach packaged in PET or PLA trays. Data are the mean of three package analyses. Vertical bars show the standard error. Means followed by the same letter are not statistically different ($p < 0.05$). Letters are not reported here because there was no significant difference between samples in all sampling times

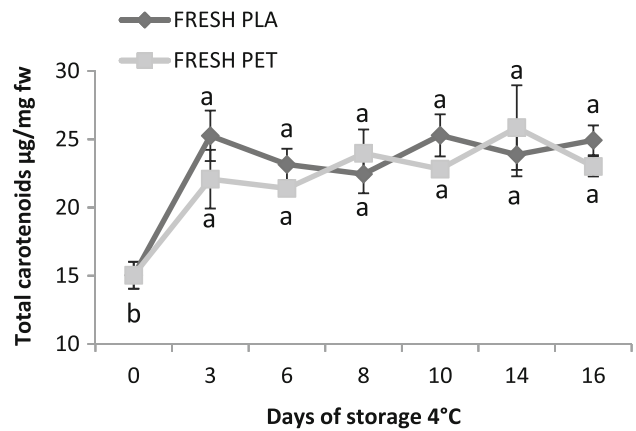


Fig. 2 Total carotenoids of fresh-cut spinach packaged in PET or PLA trays. Data are the mean of three package analyses. Vertical bars show the standard error. Means followed by the same letter are not statistically different ($p < 0.05$)

The shelf life of fresh-cut products is strongly affected by sensory attributes such as odour, visual appearance and water accumulation on the product surface or in the package.

Concerning visual appearance, it is necessary to distinguish between the visual quality of the fresh-cut spinach and the visual quality of the package as a whole. At each sampling time, no significant differences in visual score were found between spinach stored in PTA or PLA (data not shown). After 8 days of storage, the spinach were still in good shape (score between 7 and 8). Later, yellow areas appeared on the surface of the leaves; these increased in intensity and size during storage. In contrast, the visual appearance of the package showed a substantial difference between the two plastic trays; specifically, the PLA trays exhibited condensed water on the internal surface of the lid, even after 1 day of being closed, which compromised product visibility (data not shown). The difference between the two packages was due to the presence of an antifog layer in PET trays which was absent in PLA one. No off-odour was perceived until the sixth

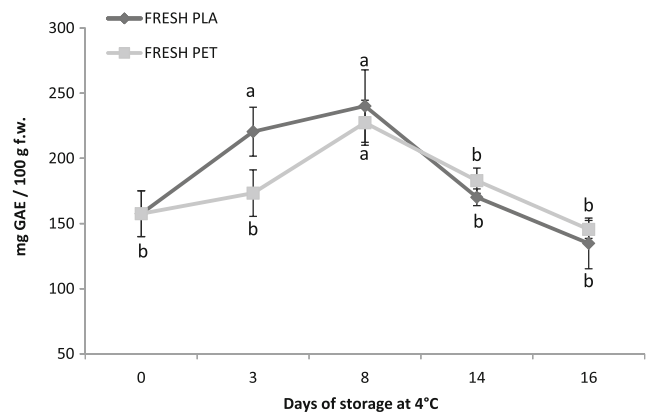
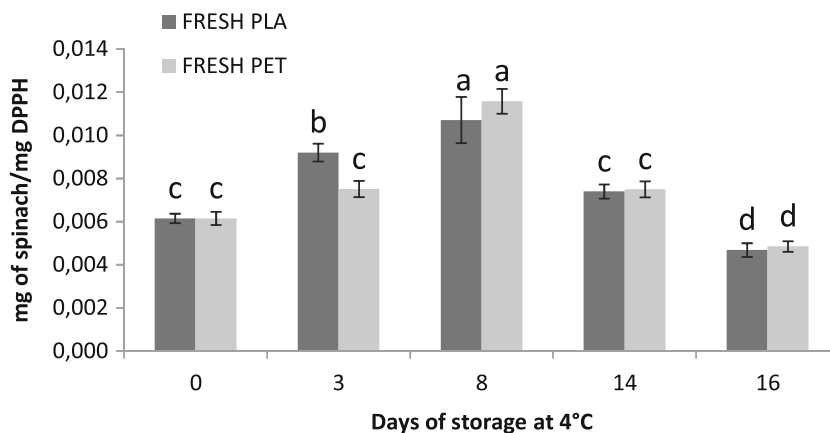


Fig. 3 Polyphenolic content of fresh-cut spinach packaged in PET or PLA trays. Data are the mean of three package analyses. Vertical bars show the standard error. Means followed by the same letter are not statistically different ($p < 0.05$)

Fig. 4 Antiradical scavenging of fresh-cut spinach packaged in PET or PLA trays. Data are the mean of three package analyses. Vertical bars show the standard error. Means followed by the same letter are not statistically different ($p < 0.05$)



day in both PLA and PET samples (data not shown). From the eighth day of storage at 4 °C, the spinach began to lose its characteristic smell, with abnormal and unpleasant odours whose intensity increased during storage. Medina et al. (2012) detected off-odours in baby spinach packaged in perforated and non-perforated bags exposed to high RH (water content after processing in water 91.9 %) at 12 days: regardless the RH, off-odours were scored as severe in all non-perforated packages while slight off odours were detected in perforated packages.

Water activity (a_w) was 0.973 throughout the storage period without significant difference between the samples (data not shown).

The surface colour of the spinach was not affected by polymer packaging and the hue angle values range between 115 and 120 during the entire storage time (data not shown). This is a very important result because consumers purchase fresh-cut produce based on its visual appearance, so colour is extremely important (Kader 2002; Ferrante et al. 2004). One of the symptoms of senescence in harvested leafy vegetables is loss of greenness with the degradation of chlorophyll. Quantitative changes in chlorophyll, degradation products of chlorophyll and the hydrolyzing enzymes have been monitored in spinach (Yamauchi and Watada 1991). To confirm the maintenance of the green colour of the spinach in our study, total chlorophyll did not change during storage time, with no difference between the samples (Fig. 1) as chlorophyll a and b did not change (data not shown). In contrast, total carotenoids increased significantly from 15 to 22–25 $\mu\text{g mg}^{-1}$ fw in the first 3 days in both samples and

remained at this level for the entire storage time (Fig. 2). Total polyphenols of fresh-cut spinach packed in PLA and PET increased from 160 to 230–240 mg GAE 100 g^{-1} fw until day 8 and then decreased to the initial values (Fig. 3). During the first 3 days of storage, extracts obtained from PLA samples had a higher polyphenolic content but, successively, the loss of polyphenols content was similar in the two samples. The antiradical scavenging activity followed the same pattern as total polyphenols (Fig. 4). The significantly higher value of polyphenols in PLA-packaged spinach in the first 3 days may have been due to a higher stress response, but the reason is unclear. In contrast with polyphenols and carotenoids, a sharp decline in ascorbic acid content was observed, especially in PET samples, during the first 3 days of storage (Table 1). As the gas concentration inside the package was similar to the normal atmosphere, the observed increase in polyphenols and carotenoids in both samples could be due to a stress condition as a consequence of the whole treatment, cutting and washing in a disinfectant solution sold as Tsunami. This disinfectant compound is formed by hydrogen peroxide and peroxyacetic acid; thus, its action is primarily oxidative and then acidifying. High H_2O_2 production rates are normally balanced by very efficient antioxidant systems. Abiotic stresses such as dehydration, low and high temperatures, and excess irradiation can disturb this balance, such that increased H_2O_2 initiates signalling responses that include enzyme activation, gene expression, programmed cell death (PCD) and cellular damage (Neill et al. 2002). In our case, the high H_2O_2 concentration used in the washing step of the spinach likely disturbed the above-mentioned balance; there is growing

Table 1 Ascorbic acid content (mg of ascorbic acid 100 g^{-1} fw) of fresh-cut spinach packaged in PLA or PET trays

	Day 0	Day 3	Day 8	Day 14	Day 16
Fresh PLA	21.88±1.96a	8.84±1.76b	5.20±1.04cd	1.46±0.51f	0.5±0.44g
Fresh PET	21.88±1.96a	3.58±0.71e	4.01±0.80de	1.40±0.49f	0.3±0.27g

Data are the mean of 3 analyses from 3 different sets of spinach leaves. Means separation was performed by applying LSD test. Values with different letters were significantly different ($p < 0.05$)

evidence that ROS participates systematically in signalling pathways and stress responses (Fujita et al. 2006). In addition, it is now well-established that secondary metabolites play a key role in the adaptation of plants to environmental constraints and this positive role may be attributed, for most of them (mainly polyphenolic compounds and carotenoids), to their antioxidant properties (Foyer and Noctor 2005). In conclusion, the initial increase in polyphenols and carotenoids in packaged fresh-cut spinach might be due to this oxidative stress effect, which, in turn, causes a rapid decline in ascorbic acid concentration because it is used by hydrogen peroxide for the conversion to water (Neill et al. 2002). The later decline in polyphenolic compounds is the natural consequence of the oxidation of these compounds and the inability for new synthesis due to the antioxidant metabolic imbalance and the senescence metabolism.

Furthermore, ascorbic acid recorded at the beginning of the study was lower than the values listed in the bibliography due to the high solubility of ascorbic acid, which was probably lost during washing and, above all, because the analyses were carried out on frozen spinach. Gil et al. (1999) observed that the initial fresh-cut spinach contained ascorbic acid (AA) as a predominant form of vitamin C. However, after 3 days of storage, a decrease in AA to one-half of the initial value was noted in both air and MAP, followed by a higher reduction after 7 days of storage.

Total mesophilic bacteria, initially equal to 5.2 log CFU g⁻¹ for both investigated samples, increased during storage, following the same trend (Table 2A). In particular, the microbial cell load of spinach reached about 6.9–7.3 log CFU g⁻¹ within 8 days and, at the end of the storage period, these values increased, reaching 7.7–8.1 log CFU g⁻¹ both for PLA and for PET. Psychrotrophic bacteria (Table 2B) showed a similar microbial growth trend. In this case also the viable cell concentration of fresh-cut spinach reached about 5.9–6.3 log CFU g⁻¹ within 8 days; this value rose further, reaching 7.8 (PET) and 8.1 (PLA) log CFU g⁻¹ at 16 days, without significant differences between PLA or PET packages. Babic and Watada (1996) reported that low O₂, rather than high CO₂, seemed to be the limiting factor for the growth of aerobic microorganisms on spinach at 5 °C but not at 10 °C.

Cooked-cut spinach storage

A significant drop in the visual appearance of the cooked spinach was observed on day 8 but without differences between the samples (data not shown). As with the fresh spinach, for the cooked spinach the accumulation of condensed water on the internal surface of the PLA lid compromised clear visibility of the leaves (Table 3A).

No off-odour was detected until the sixth day in both spinach samples packaged in PET and PLA. PLA samples were scored 3.5 vs 2.5 of PET samples on day 8, by indicating a

Table 2 Mesophilic and psychrophilic bacteria counts (log CFU g⁻¹) of fresh-cut (A, B) and cooked (C, D) spinach packaged in PLA or PET trays

A	Day 0	Day 8	Day 16
Fresh PLA	5.2±0.4a	6.9±0.5b	7.7±0.6bc
Fresh PET	5.2±0.4a	7.3±0.3b	8.1±0.7c
B	Day 0	Day 8	Day 16
Fresh PLA	5.0±0.3a	6.3±0.5b	8.1±0.4c
Fresh PET	5.0±0.3a	5.9±0.4b	7.8±0.7c
C	Day 0	Day 6	Day 10
Cooked PLA	2.9±0.2a	4.3±0.5b	7.8±0.3c
Cooked PET	2.9±0.2a	4.7±0.4b	8.0±0.5c
D	Day 0	Day 6	Day 10
Cooked PLA	2.7±0.5a	4.5±0.3b	7.7±0.4c
Cooked PET	2.7±0.5a	5.1±0.4b	8.1±0.5c

Data are the mean of 3 analyses from 3 different sets of spinach leaves. Means separation was performed by applying LSD test. Values with different letters were significantly different (*p*≤0.05)

longer maintenance of the typical aroma of the cooked spinach, likely due to the better closure system, as has been postulated for the reduced weight loss (data not shown). As expected, with the progression of the storage days at 4 °C, the spinach began to lose its characteristic flavour, and on the tenth day, it smelled unpleasant. No significant differences were found in free water content (a_w) during the storage period and between the samples (data not shown). According to Lunati (2001), a storage period not exceeding 7 days is recommended because the hygienic and health properties deteriorate and qualitative decay leads to abnormal colours and smells that affect marketability. Surface colour, measured as hue angle of cooked spinach, was not affected by packaging (data not shown). Total chlorophyll content decreased in both samples, especially in PLA trays, but there was no significant difference among the sampling times and between the two samples due to large variability in the replication values (Table 3B). Chlorophyll a and b confirmed the total

Table 3 Water accumulation (A), total chlorophylls (B) and total carotenoids (C) of cooked spinach packaged in PLA or PET trays

A	Day 0	Day 6	Day 10
Cooked PLA	9.0±0.4a	8.5±0.3a	8.0±0.2a
Cooked PET	9.0±0.4a	7.0±0.2b	6.0±0.2c
B	Day 0	Day 6	Day 10
Cooked PLA	105.57±5.93a	79.38±5.21b	83.86±8.18b
Cooked PET	105.57±5.93a	95.84±7.34ab	95.54±8.83ab
C	Day 0	Day 6	Day 10
Cooked PLA	19.49±2.04a	13.92±2.56b	13.83±5.49ab
Cooked PET	19.49±2.04a	18.73±2.71a	18.24±5.87a

Data are the mean of 3 analyses or observations from 3 different sets of spinach leaves. Means separation was performed by applying LSD test. Values with different letters were significantly different (*p*≤0.05)

chlorophyll content (data not shown). Even the total carotenoid pattern was similar to that of total chlorophyll with no significant difference due to the great variability among replications (Table 3C).

Total mesophilic bacteria in the cooked spinach packaged in PET or in PLA trays increased slightly during storage, following the same trend. In particular, the mesophilic microbial cell load of the cooked spinach (Table 2C) reached 4.3–4.7 log CFU g⁻¹ within 6 days, reaching 7.8–8.0 log CFU g⁻¹ at the end of the storage period (10 days). Psychrotrophic bacteria of the cooked spinach (Table 2D) were equal to 4.5–5.1 log CFU g⁻¹ within 6 days, without significant differences between spinach packaged in PLA and spinach packed in PET. In this case also the viable cell concentration reached 7.7–8.1 log CFU g⁻¹ at the end of the trial. Psychrotrophic and mesophilic bacteria showed a similar microbial growth trend even for cooked spinach stored in PLA and PET.

Conclusions

Six to seven days is the maximum cold storage time for fresh or cooked cut spinach both in PET and in PLA trays with hinged lids. At this time, the overall quality and antioxidant capacity of the spinach leaves is good even though ascorbic acid loss occurs in the fresh spinach. Because of the biodegradability and compostability of PLA package, this result must be seen positively to promote the use of PLA instead of PET in retail distribution of spinach, fresh or cooked. Unfortunately, the PLA package showed undesirable water condensation on the internal surface of the lid.

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