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THE EFFECT OF PACKAGING AND PRE-PACKAGING TREATMENTS ON QUALITY OF FRESH-CUT SPINACH DURING COLD STORAGE

Rabie, S. M. H. and Asrar Y. I. Mohamed
Food Technology Res. Institute, Agric. Research Center, Giza, Egypt.

ABSTRACT

Fresh cut spinach leaves (*Spinacia oleracea L.*) can be used for preparing many types of delicious dishes or eaten raw along with green salads because of its high nutritional value. Different pre-packaging dipping treatments and local packaging materials were used to extend the shelf life of this highly perishable product. The dipping solutions for shredded spinach leaves (2 cm in width) were: water (control), ascorbic acid (0.2%) and potassium permanganate (0.02%). After dipping, drained samples were then packaged in two polypropylene (PP) pouches (12 × 20 cm) of different thickness (22, and 30 µ) and in polystyrene (PS) trays over-wrapped with a clear stretch polyvinylidene chloride (PVdC) film. Packaged samples were kept refrigerated at 4±0.5 °C and 85±3 % R.H. for nine days.

Water loss of packaged shredded spinach leaves, was minimal for samples packaged in PP-22µ and PP-30µ pouches, whereas those packaged in PS trays showed higher weight loss. The water treated samples showed the least shelf life. Other treated samples showed lower microbial counts initially and continued lower during storage. Samples treated with ascorbic acid were the best followed by those treated with potassium permanganate. Deterioration in color (yellowing) was fast notable for the water dipped samples, whereas ascorbic acid treated samples had higher panelists' scores for color during storage. Fresh cut spinach leaves packaged in PS trays showed about 9 days shelf life, compared with 6 days for those packaged in PP pouches. It is recommended to package ascorbic acid treated fresh cut spinach leaves in PS-tray covered with stretch PVdC overwrap than using PP pouches which, unless perforated, may lead to development of in-package anaerobic atmosphere.

INTRODUCTION

Spinach (*Spinacia olera*cea L.), among other fresh leafy greens, is an important source of nutrients having current health concerns. Spinach is rich in total carotenoids, antioxidant capacity, folate and vitamin C (Cao, *et al*., 1996; Holden *et al.,* 1999; Pandrangi and LABorde, 2004). Fresh cuts, however, have short shelf life because of their inherent sensitivity and quick quality deterioration. In addition, many of outbreaks have been linked to spinach consumption. Good processing practice and proper packaging are needed to maintain the added value, quality, microbial and consumer safety, as well as to extend the shelf life of this highly perishable product.

Two main factors contribute to the short shelf life of packaged fresh cut spinach; cell senescence and microbial activity. Cell senescence is triggered by ethylene evolution resulting from bruising, mechanical injuries, and excessive trimming leading to discoloration (yellowing in spinach) which reflects degradation of chlorophyll and revelation of carotenoid pigments (Heaton and Marangoni, (1996). This process is accompanied by an increase in respiratory activity, elevation in CO_2 production, and depletion of the indigenous antioxidants including vitamin C (Yamauchi and Watada, 1991 and 1998; Gil *et al*., 1999 and Bergquist *et al.,* 2005). This process may be delayed by proper temperature management after harvest and applying modified storage atmosphere of reduced O_2 and/or up to 10% CO_2 (Lee and Kader, 2002; Techavuthiporn, *et al*., 2006).

Methods of preparation of minimally processed fresh produce affect nutrient retention. Tosun and Yücecan (2007) studied vitamin C level and the factors affecting it before and during home processing and freezing and during freeze storage of some fresh vegetables including spinach. Vitamin C content of fresh spinach samples was 89.5± 5.86 mg/100g. Root cutting and washing resulted in 17.5% loss in the initial vitamin C content. The amount of vitamin C decreased to two-third after blanching and freezing and to one-third after 6 month of frozen storage. Ma *et al*., (2006) reported that the decrease in chlorophyll and soluble protein contents and hue angle value in leafy vegetables can be significantly delayed by using 1-methylcyclopene (which is an ethylene binding inhibitor and has been used to prolong the shelf life of many fresh produce) at a level up to 0.5µl/L.

Beis *et al.,* (2002) determined the dry matter, soluble solids, nitrates, oxalates, pH and titratable acidity in spinach leaf parts at different plant age. The authors found significant variation among leaves of various ages with respect to these measurements except for the soluble solids content. Pandrangi and LABorde, (2004) studied the effect of packaging and storage temperature on nutrient retention, weight loss, microbial population, enzyme activity, and visual quality of commercially packaged fresh spinach. The authors reported a shelf life of 4, 6, and 8 days for packaged spinach stored at 20, 10, and 4 °C, respectively. Weight loss, chlorophyll degradation, color difference, carotenoid losses and microbial populations increased during storage and the higher the temperature the faster was the increase. The authors found also that vitamin and quality changes were unaffected by the presence or absence of packaging. The package had 2.5 perforations (1-mm diam.) per cm^2.

Microbiological quality of fresh produce is a concern not only from a food safety perspective, but also because of resultant losses due to decreased product shelf-life. An estimated 30% of produce is lost due to microbial spoilage between the time of harvest and consumption (Beuchat, 1992). Francis*, et al*., (1999), in their review on microbiological safety of minimally processed vegetables, stated that the fresh nature of these products and the mild processing techniques in addition to the following storage conditions, present health hazard vehicles posing a potential safety problem. This is due to the fact that “ready-to-use” vegetables retain much of their indigenous microflora – including possible pathogens - after processing.

The effect of washing treatments on microbiological quality of leafy vegetables such as lettuce serviced at university restaurants has been of great concern (Soriano *et al.,* 2000). The authors reported that sodium hypochlorite or potassium permanganate solutions when used in washing procedure, the aerobic microorganisms were reduced by more than two log units, and the total coliforms by at least one log. El-Sayed *et al.,* (1997) stated that prevention of human fascioliasis could depend on clearing of the leafy salads from the metacercariae. The authors found that washing in running water alone for 10 minutes detached only 50% of metacercariae. Potassium permanganate at 24 mg/L level detached all metacercariae after 10 minutes exposure. Vegetable leaves were not softened and maintained fresh.

Several studies examined the overall microbial quality in fresh and fresh cut vegetables starting from the field, during processing, and storage till final consumption (Ruiz *et al*., 1987, Pingulkar *et al.,* 2001). Using the electron microscope, Babic *et at*., (1996) found that microorganisms were infecting the internal palisade parenchyma where the cuticle was injured. Verlinden and Nicolaï (2000) stated that microbial loads can be minimized by implementing HACCP programs into the production process. The authors also recommended a suitable application of modified atmosphere packaging (MAP) in order to retard the high respiration rate of the product and minimize moisture loss and microbial growth.

Allende *et al.,* (2004) studied the effect of package material and atmosphere on the microbial and quality changes of minimally processed baby spinach leaves. They found that adding super atmospheric O_2 to the packages alleviated tissue injury in addition to reducing microbial growth and was beneficial in maintaining quality of fresh-cut baby spinach. Verlinden and Nicolaï, (2000) reported that very low oxygen concentrations inside the package that lead to fermentative metabolism should be avoided. Such anaerobic atmosphere may preserve a fresh appearance of the product, but off-odors and off-taste render it unsuitable by the consumer.

MATERIALS AND METHODS

Materials:

Spinach Source: Fresh sound **Spinach** (*Spinacia oleracea* L.) leaves of Balady variety was obtained from Horticultural Research Institute, Agriculture Research Center, Giza, season 2006 .

Package materials: Pouches (150x200 mm) were made of bi-axial oriented polypropylene (BOPP) of 22µ thickness (PP_22) and 30µ thickness (PP_30), and polystyrene (PS) trays covered with polyvinylidene chloride (PVdC) stretch overwrap. The trays and the wrap were bought from the local market at Giza, whereas the BOPP materials were donated by Egy. Wrap; a local manufacturer of polypropylene film, 6 October city, Giza .

Chemicals: All chemicals materials used were food grade and were bought from the local market (Al-Gomhyria Medical and Chemical Company, El-Sawah, Cairo).

Methods:

Sample preparation: Prior to packaging, spinach leaves were sorted and the good sound clean healthy ones were selected, divided into groups. Groups were randomly assigned to each of the treatment-combinations. Leaves were chopped at the width of 2 cm.

Dipping and packaging treatments:

Chopped spinach leaves were dipped for 5 min in one of the following dipping solutions; ascorbic acid solution (0.2%), potassium permanganate (0.02%), and water (control). After dipping, excess solutions were left to drain for 5 min. Two hundred grams of drained spinach leaves from each dip treatment were packaged in PP_30 and PP_22 pouches and PS trays covered with polyvinylidene chloride (PVdC) stretch film over wrap. Packaged samples were stored refrigerated at 4°C and 85% R.H. for nine days. Random samples were withdrawn regularly during storage for inspection and to run weight- loss , total soluble solids % (TSS), pH , microbial and sensory evaluations.

Loss in Weight (L.W.): Changes in weight of packaged spinach samples were monitored during storage, and calculated (average of three replicates) according to the following equation:

% L.W = (Initial weight - weight of spinach at sampling date) X 100/ Initial weight of spinach.

pH value of spinach leaves: The pH value of the sap-expelled by pressing of spinach tissues was measured by the use bench top pH-meter [Jenway Model 3510 pH/mV/Temperature Meter, Gransmore Green Felsted, Dunmow Essex CM6 3LB, England].

Total soluble solids (TSS)% : The percentage of total soluble solids (TSS) of spinach leaves sap was measured by a refractometer [Abbe Refractometer, Leica Mark II, Germany] (A.O.A.C, 2000). One ºBrix is equivalent to approximately 1 g of soluble solids per 100 g. of a sample solution.

Microbial enumeration:

The microbial contents of spinach samples were determined according to the methods described in the DIFCO manual (DIFCO, 1984). Acidified potato dextrose agar and nutrient agar were used to enumerate yeast & mold and total microbial counts, respectively. Other total count plates were incubated under anaerobic conditions in order to enumerate anaerobic bacteria. Duplicated plates of these cultures were enumerated and expressed as colony forming units (CFU/ g sample)

Sensory evaluation: Samples were evaluated by a group of trained panelists recruited from the Food Technology Research Institute. Color, smell, texture and wiltness attributes were evaluated for the degree of likeness and given a point from 5 on an opened scale. Where 1 is unlike most, and 5 is extremely liked (Meilgaard, *et al.,* 1999).

Statistical analysis.: Statistical analysis was conducted using the SPSS Statistical Software Package v.11.5 (Pallant, 2001). Comparisons among the main treatment means were made using L.S.D test at (P = 0.05) (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Physicochemical evaluation:

Data in Table (1) summarize the analysis of variance of the weight loss, total soluble solids (TSS) and pH value of packaged fresh cut spinach leaves during nine days of cold storage. The mean values of weight loss, TSS and pH values of minimally processed spinach leaves during storage are presented and depicted in Figure (1).

Weight loss:

Weight loss (Table 1) was significantly affected by the three main factors; package materials, dipping solutions, and storage duration (***P*** ≥ 0.001, 0.05, and 0.01; respectively). The two-way interaction PKGxTRT was also significant (***P*** ≥ 0.05) which indicates that the effect of packaging on weight loss depends on the type of dipping solution and vice versa. The loss increased as storage duration was increased. It was greater within first few days than by the end of the storage period. Comparison among the overall means of the main treatments indicated that the mean value of weight loss of the 3d day be significantly lower than the other two storage durations. The mean value of weight loss of samples treated with the potassium permanganate was significantly lower than those of samples treated with water or ascorbic acid. These results were in agreement with those reported by Pandrangi and LABorde, (2004).

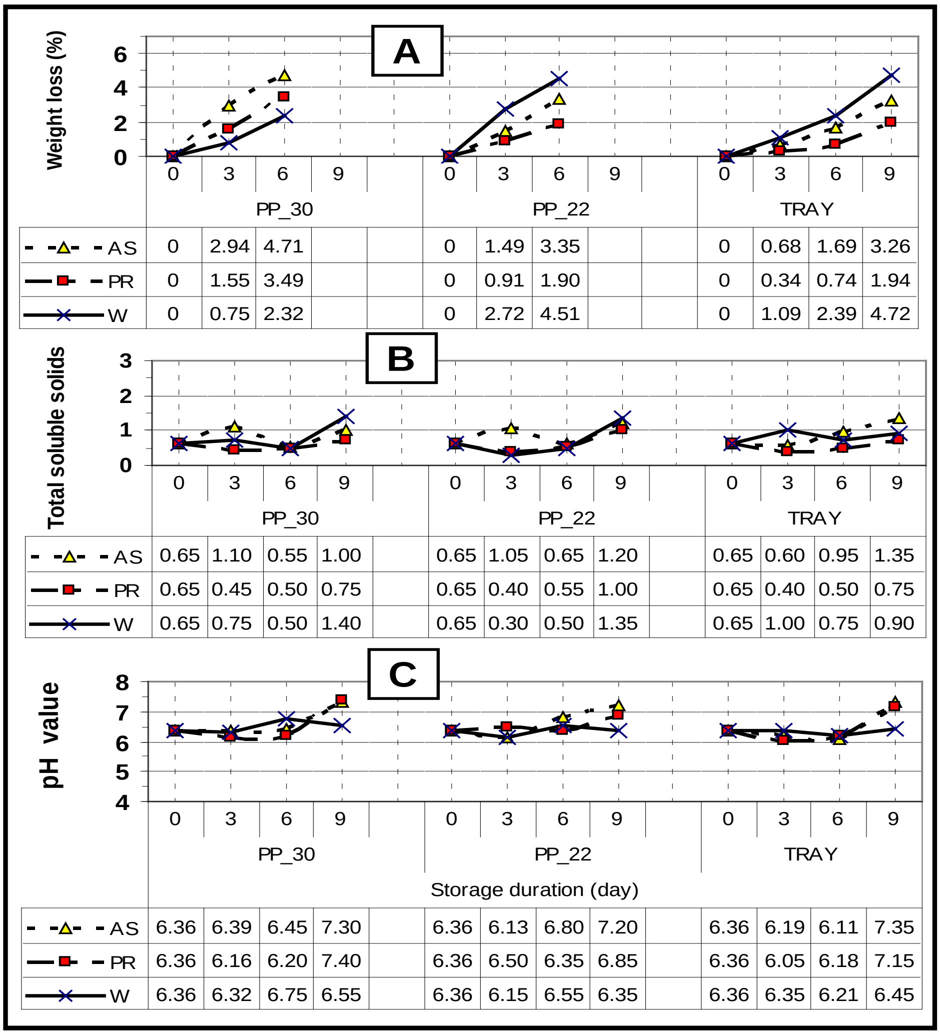
Table (1): Analysis of variance mean squares estimates for weight loss, total soluble solids (TSS %), and pH value of packaged fresh-cut spinach leaves during nine days of storage at 4°C and 85% R.H. .

Source of variance[1]	**Weight loss %**		d.f	**TSS (%)**	**pH value**
	d.f	**Mean squares**		**Mean squares**	**Mean squares**
Packaging materials (PKG)	2	7.924 **	2	0.003 NS	0.124 NS
Dipping solutions (TRT)	2	5.641 *	2	0.576 **	0.278 *
Storage duration (DAYS[2]**)**	2	18.141 **	2	1.176 **	2.501 **
PKG x TRT	4	4.158 *	4	0.042 NS	0.018 NS
PKG x DAYS	2	0.595 NS	4	0.089 NS	0.139 NS
TRT x DAYS	4	0.513 NS	4	0.066 NS	0.512 **
PKG x TRT x DAYS	4	0.142 NS	8	0.131 NS	0.081 NS
Error	21	1.312	27	0.086	0.070
$LSD_{0.05}$ =		**0.693**		**0.201**	**0.181**

[1] PKG x TRT, PKG x DAYS, and TRT x DAYS denote the two-way interactions between each pairs of the main factors; Packaging materials (PKG), Dipping solutions (TRT) and Storage duration (DAYS), whereas PKG x TRT x DAYS denotes the three-way interaction among them.

[2] Days factor (storage durations) for weight loss has one degree of freedom (d.f) less because data at zero time were all zeros and were excluded from the analysis.

***,** denote significant at P level 0.05 and 0.01; respectively. NS= not significant at P=0.05.**



A – Weight loss (%)

	PP_30 0	3	6	9	PP_22 0	3	6	9	TRAY 0	3	6	9
AS	0	2.94	4.71		0	1.49	3.35		0	0.68	1.69	3.26
PR	0	1.55	3.49		0	0.91	1.90		0	0.34	0.74	1.94
W	0	0.75	2.32		0	2.72	4.51		0	1.09	2.39	4.72

B – Total soluble solids

	PP_30 0	3	6	9	PP_22 0	3	6	9	TRAY 0	3	6	9
AS	0.65	1.10	0.55	1.00	0.65	1.05	0.65	1.20	0.65	0.60	0.95	1.35
PR	0.65	0.45	0.50	0.75	0.65	0.40	0.55	1.00	0.65	0.40	0.50	0.75
W	0.65	0.75	0.50	1.40	0.65	0.30	0.50	1.35	0.65	1.00	0.75	0.90

C – pH value

	PP_30 0	3	6	9	PP_22 0	3	6	9	TRAY 0	3	6	9
AS	6.36	6.39	6.45	7.30	6.36	6.13	6.80	7.20	6.36	6.19	6.11	7.35
PR	6.36	6.16	6.20	7.40	6.36	6.50	6.35	6.85	6.36	6.05	6.18	7.15
W	6.36	6.32	6.75	6.55	6.36	6.15	6.55	6.35	6.36	6.35	6.21	6.45

Figure (1): Changes in weight loss (A), total soluble solids (B), and pH value (C) of packaged fresh cut spinach leaves during nine days of cold storage at 4 °C and 85% R.H.

Packaging materials: PP_30, PP_22 = Polypropylene pouches 30 μ and 22 μ, respectively, and TRAY= Polystyrene tray with stretch film over wrap.
Dipping solutions: As = Ascorbic acid (0.2%), Pr = Potassium permanganate (0.02%), and W = Water (Control).

Total soluble solids (TSS) %:

Data in Table (1) show that the effect of dipping solutions and storage duration on total soluble solids % (TSS) of fresh cut spinach leaves were significant (***P*** ≥ 0.001). The effects of the packaging materials and the interactions were not significant (***P*** = 0.05). The overall mean value for TSS of samples treated with ascorbic acid was significantly higher (***P*** ≥ 0.05) than that of samples treated with potassium permanganate, whereas the control showed an intermediate value that was not significantly different from any of them. The overall mean value of TSS for the 9th day was significantly higher than that on the 3d and 6th days of storage.

Data in Figure (1) show that the initial value for TSS was 0.65 % and ranged between 0.3 – 1.4 % during the nine days of storage. It can also be seen from Figure (1) that a shallow change in the TSS was observed at the beginning of storage, followed by a continuous increase by the end of nine days of storage. The value of TSS reflects the concentration of soluble constituents (sugars, organic acids, ..etc.) in the available cell sap. Such concentration represents the metabolic activity of the fresh produce (respiration, enzymes ..etc.) and that of the harboring microorganism. These results are in agreement with those reported by Pandrangi and LABorde, (2004).

pH value:

The analysis of variance (Table 1) showed that the effect of dipping treatment (TRT) and storage duration (DAYS) as well as the TRTxDAYS interaction were significant (($P \geq 0.05$, 0.01, and 0.01, respectively). This indicates that the effect of any of the treatments depends on duration of the storage and vice versa. The pH value at the beginning of the experiment was 6.36 and ranged between 6.11 and 7.40 during the storage. Comparison among the overall means of the dipping solutions showed significantly higher value for ascorbic acid treated sample than those of the other treatments and that of the 9th day was also higher than those of the 3d and 6th days of storage.

It can be seen from Figure (1) that water treated samples packaged in PP_22 and PP_30 showed a higher increase in pH value at the beginning of storage followed by a continuous decline towards the end of storage. On the contrary , water treated sample packaged in trays and other samples treated with ascorbic acid or potassium permanganate showed negligible changes in pH value at the beginning of storage followed by a continuous increase following the 6th day of storage. The increase in pH value for during storage may reflect depletion of indigenous organic acids.

Microbiological evaluation:

Analysis of variance for microbial counts; total microbial count, yeast and mold count, and anaerobic bacterial counts were summarized in Table (2). The mean values are presented and depicted in Figure (2).

Total microbial count (TC):

Data in Table (2) show that all the main factors; package materials, dipping solutions, and storage duration had significant effects ($P \geq 0.01$) on total count. In addition, the two- and three-way interactions showed also significant effects on microbial counts. These means that the effect of any of the main factors on microbial count depends on the value/level of the other two factors. In this case comparisons among the overall means of the main factors are not valid.

Table (2): Analysis of variance mean squares estimates for total count (TC), yeast and mold count (Y&xM), anaerobes count (AN) of packaged fresh-cut spinach leaves during nine days of **storage at 4°C and 85% R.H.**

Source of variance[1]	d.f	Mean squares		
		TC x 10^3	**Y&M** x 10^3	**AN** x 10^3
Packaging materials (PKG)	**2**	485.08 **	254.78 **	3.44 NS
Dipping solutions (TRT)	**2**	227.19 **	60.08 **	7.19 *
Storage duration (DAYS)	**2**	2183.44 **	69.25 **	5.36 NS
PKG x TRT	**4**	314.99 **	12.74 NS	4.43 NS
PKG x DAYS	**4**	81.44 **	93.15 **	6.60 *
TRT x DAYS	**4**	50.56 *	26.33 **	1.85 NS
PKG x TRT x DAYS	**8**	44.72 *	19.42 **	1.96 NS
Error	**81**	20.56	5.65	2.19
$LSD_{0.05}$ (CFU/g) **=**		2.13	1.12	0.70

[1] PKG x TRT, PKG x DAYS, and TRT x DAYS denote the two-way interactions between each pairs of the main factors; Packaging materials (PKG), Dipping solutions (TRT) and Storage duration (DAYS), whereas PKG x TRT x DAYS denotes the three-way interaction among them.

***,** denote significant at *P* level 0.05 and 0.01; respectively. NS = not significant at *P* = 0.05.**

Data in Figure (2) show that TC increased continuously during the nine days of storage. The TC value was 11.60 x10^3 CFU/g sample at the beginning of the experiment and reached the range of 25.25 - 49.25 x10^3 CFU/g sample by the day 9th. It can be seen from Figure (2) that TC counts increased by increasing storage duration. When PS trays were the packaging material, there was no difference among dipping treatments with respect to TC during storage. For samples packaged in PP_22 and PP_30, those treated with water recorded higher values than the other two treatments. For water treated samples, those packaged in PP_22 showed higher TC on the 6th and 9th days of storage than those packaged in other types of packaging. Those samples treated with ascorbic acid and packaged in PP_30 recorded lower values than the others at the 9th day of storage.

These values were lower than those reported by Ruiz *et al.* (1987) and Pingulkar *et al*., (2001) for total aerobic counts in leafy vegetables. According to Piagentini and Guemes (2002), samples in Figure (2) had total counts within the acceptable limit stated by the French regulations for fresh cut vegetables (i.e. less than 5x10^7). Odumeru *et al*., (1997) reported that ready-to-use vegetables showed up to a 1-log decrease in aerobic colony counts after processing. Those counts increased to preprocessing levels after 4 days of storage at both 4 and 10 ^oC.

Yeast and mold counts (Y&M):

The main factors involved in the experiment showed significant effect on yeast and mold counts Table (2). The interaction PKGxTRT showed insignificant effect whereas other two- and the three- way interactions were significant. The yeast and mold count was 6.80 x10^3 CFU/g sample at the beginning of the experiment and reached the range of 9.25 - 15.00 x10^3 CFU/g sample by the 9th day (Fig.2). It should be noted that permeability of the three types of packages for O_2, CO_2 and water vapor are in the following order: PS trays >> PP_22 pouches > PP_30 pouches. Yeast and molds are aerobic microorganisms. It is expected fro the PS tray to allow enough gas exchange with the outer environment more than the PP pouches. The atmosphere inside PP pouches is expected to have more CO_2 and far less oxygen than the PS tray does. Therefore, less yeast and mold counts is expected for samples packaged in PP materials. The decrease in aerobic counts at the beginning of storage followed by an increase to the preprocessing levels within 4 days of storage was reported by

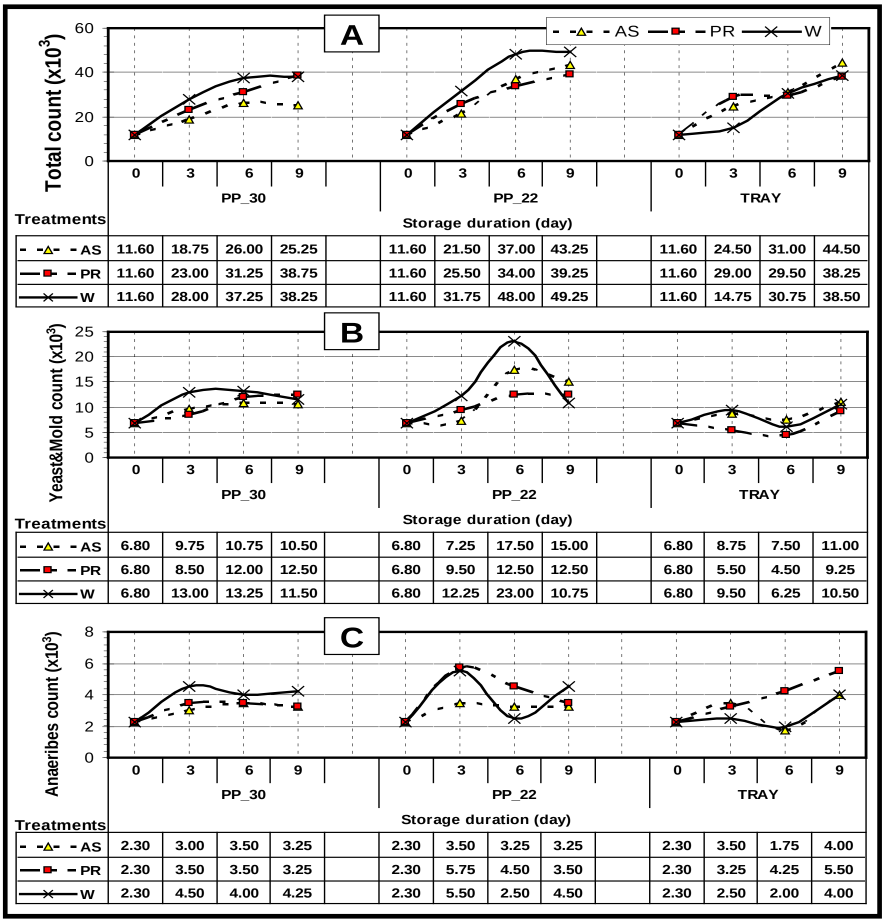


Figure (2). Total microbial (A), yeast and mold (B) and anaerobes (C) counts of fresh cut spinach leaves during nine days of cold storage at 4 C and 85% R.H.

Packaging materials: PP_30, PP_22 = Polypropylene pouches 30 μ and 22 μ, respectively, and TRAY= Polystyrene tray with stretch film over wrap.

Dipping solutions: As = Ascorbic acid (0.2%), Pr = Potassium permanganate (0.02%), and W = Water (Control).

However, the values for yeast and mold counts in Figure (2) are within the range reported by Pingulkar *et al.,* (2001) for yeast-mold counts in washed leafy vegetables (10^2-10^5 cfu/g).

Anaerobic bacterial count (AN):

Data in Table (2) showed that only the dipping treatment to have a significant effect on anaerobic bacterial count (***P*** ≥ 0.05). In addition, the PKGxDAY interaction was also significant (***P*** ≥ 0.05) which indicated that the effect of packaging material on the AN depends on the storage duration of evaluation and vice versa. The anaerobic bacterial count was 2.30 $x10^3$ CFU/g sample at the beginning of the experiment and reached the range of 3.25 - 5.50 $x10^3$ CFU/g sample by the day 9th. It can be seen from Figure (2) that the anaerobic bacterial count to increase during the first 3 days for all treatments, then either leveled off or increased.

This results can be explained in the light changes in gas composition of package atmosphere which depends mainly on produce respiratory activity and package permeability to O_2 and CO_2 gases. Within an impermeable package, depletion of oxygen and accumulation of respiratory CO_2 will develop anaerobic conditions resulting in anaerobic respiration of the packaged samples and development of off-odor and off-taste (Verlinden and Nicolai, 2000). Growth of anaerobes under such conditions will depend – among other factors - upon level of available nutrients and other competing microorganism. It should be noted here that non of the samples particularly those packaged in PS trays developed such conditions.

Sensory evaluation:

Data summarized in Table (3) present the analyses of variance of panelists' scores for sensory attributes of packaged fresh cut spinach leaves during nine days of cold storage. The mean values of these attributes are summarized and depicted in Figure (3).

Table (3): Analysis of variance mean squares estimates for sensory attributes of packaged fresh-cut spinach leaves during nine days of cold storage at 4°C and 85% R.H.

Source of variance[1]	d.f	Mean squares			
		Color	Smell	Texture	Wiltness
Packaging materials (PKG)	**2**	2.389 **	3.500 **	1.352 **	5.556 **
Dipping solutions (TRT)	**2**	4.955 **	4.226 **	2.276 **	4.560 **
Storage duration (DAYS[2])	**2**	2.833 **	4.759 **	2.046 **	3.796 **
PKG x TRT	**4**	0.056 NS	0.056 NS	0.046 NS	0.056 NS
PKG x DAYS	**2**	0.056 NS	0.056 NS	0.019 NS	0.296 NS
TRT x DAYS	**4**	0.056 NS	0.065 NS	0.102 NS	0.130 NS
PKG x TRT x DAYS	**4**	0.111 NS	0.111 NS	0.046 NS	0.074 NS
Error	**42**	0.286	0.254	0.270	0.302
$LSD_{0.05}$ =		**0.29**	**0.28**	**0.29**	**0.30**

[1] PKG x TRT, PKG x DAYS, and TRT x DAYS denote the two-way interactions between each pairs of the main factors; Packaging materials (PKG), Dipping solutions (TRT) and Storage duration (DAYS), whereas PKG x TRT x DAYS denotes the three-way interaction among them.

[1] [1]Days factor (storage duration) for weight loss has one degree of freedom (d.f) less because data at zero time were all fives and were excluded from the analysis.

***, ** denote significant at *P* level 0.05 and 0.01; respectively. NS= not significant at *P* level 0.05.**

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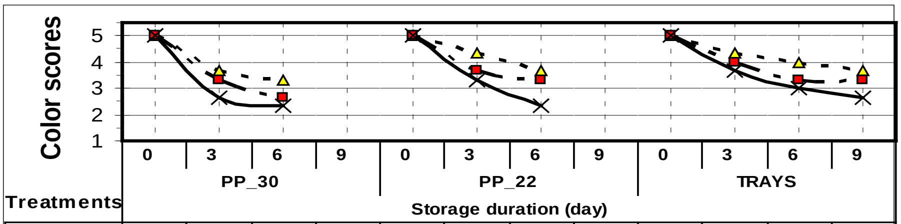
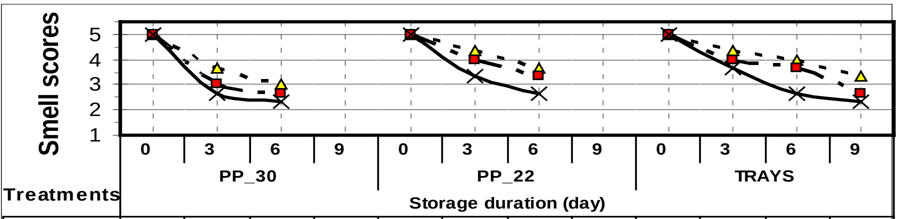
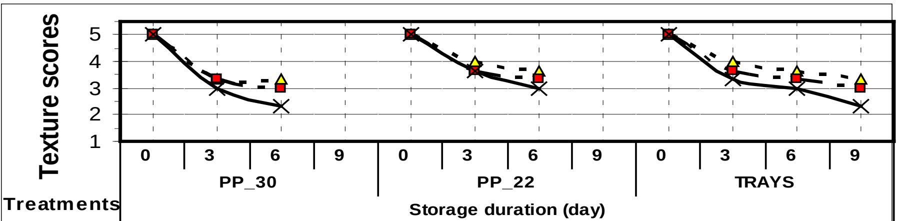


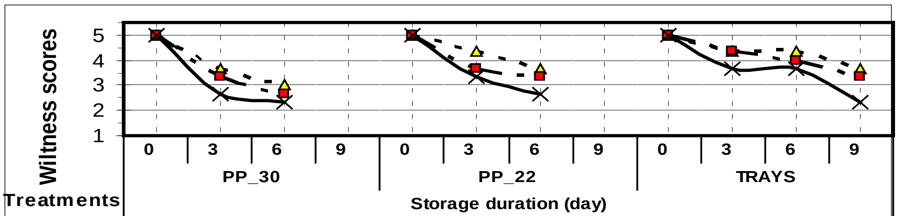
Treatments	PP_30 0	PP_30 3	PP_30 6	PP_30 9	PP_22 0	PP_22 3	PP_22 6	PP_22 9	TRAYS 0	TRAYS 3	TRAYS 6	TRAYS 9
AS	5.00	3.67	3.33		5.00	4.33	3.67		5.00	4.33	4.00	3.67
PR	5.00	3.33	2.67		5.00	3.67	3.33		5.00	4.00	3.33	3.33
W	5.00	2.67	2.33		5.00	3.33	2.33		5.00	3.67	3.00	2.67



Treatments	PP_30 0	PP_30 3	PP_30 6	PP_30 9	PP_22 0	PP_22 3	PP_22 6	PP_22 9	TRAYS 0	TRAYS 3	TRAYS 6	TRAYS 9
AS	5.00	3.67	3.00		5.00	4.33	3.67		5.00	4.33	4.00	3.33
PR	5.00	3.00	2.67		5.00	4.00	3.33		5.00	4.00	3.67	2.67
W	5.00	2.67	2.33		5.00	3.33	2.67		5.00	3.67	2.67	2.33



Treatments	PP_30 0	PP_30 3	PP_30 6	PP_30 9	PP_22 0	PP_22 3	PP_22 6	PP_22 9	TRAYS 0	TRAYS 3	TRAYS 6	TRAYS 9
AS	5.00	3.33	3.33		5.00	4.00	3.67		5.00	4.00	3.67	3.33
PR	5.00	3.33	3.00		5.00	3.67	3.33		5.00	3.67	3.33	3.00
W	5.00	3.00	2.33		5.00	3.67	3.00		5.00	3.33	3.00	2.33



Treatments	PP_30 0	PP_30 3	PP_30 6	PP_30 9	PP_22 0	PP_22 3	PP_22 6	PP_22 9	TRAYS 0	TRAYS 3	TRAYS 6	TRAYS 9
AS	5.00	3.67	3.00		5.00	4.33	3.67		5.00	4.33	4.33	3.67
PR	5.00	3.33	2.67		5.00	3.67	3.33		5.00	4.33	4.00	3.33
W	5.00	2.67	2.33		5.00	3.33	2.67		5.00	3.67	3.67	2.33

Figure (3). Changes in panelists' scores of sensory attributes of packaged fresh cut spinach leaves during nine days of cold storage at 4 °C and 85% R.H.

Packaging materials: PP_30, PP_22 = Polypropylene pouches 30 μ and 22 μ, respectively, and TRAY= Polystyrene tray with stretch film over wrap.

Dipping solutions: As = Ascorbic acid (0.2%), Pr = Potassium permanganate (0.02%), and W= Water (Control).

Data in Table (3) for all the attributes showed a significant effects (***P*** $\geq$ 0.01) of the main factors; packaging, dipping, and storage on panelists' scores. The two- and three- way interactions were not significant (***P*** = 0.05). In such case comparison among the overall means of the main factors is possible. It can be noticed from Figure (3) that all attributes recorded a decline in panelists' score during nine days of storage. Samples recorded less than 3 were considered questionable and rejected by the panelists.

Panelists' scores for color decreased continuously during storage (Figure 3). Comparison among the overall means of panelists' scores for color attribute showed significant variation for samples treated with the three dipping treatments; samples treated with ascorbic acid recorded the highest scores, whereas those treated with water showed the least scores. Also, samples packaged in trays or PP_22 showed similar overall means for color attributes that were significantly better than those packed in PP_30. The overall means for panelists scores of color attributes of samples on the 3d day were significantly higher than those the 6th and 9th days of storage. Scores equal to or above 3 was considered unobjectionable. All samples treated with ascorbic acid were acceptable at the 6th day of storage but those packaged in PP_22 and PP_30 were rejected after then.

The characteristic green color of vegetables is attributed to domination of the chlorophyll pigment, which may degrade to undesirable grey-brown compounds such as pheophorbide and pheophytin (Heaton and Marangoni, 1996). This degradation is mediated by acid and the enzyme chlorophyllase. Pheophorbide can be further metabolized to colorless compounds in metabolically active tissue. Yellowing of the fresh cut spinach leaves reflected degradation of the chlorophyll and dominance of the carotenoid pigments. Pandrangi and LABorde (2004) reported shelf life of 8 days for whole spinach leaves stored at 4 °C in a perforated package. Samples packaged in trays showed acceptable panelists' scores for color attribute till the 9th day of storage except those treated with water was rejected by the panelists after the 6th day of storage (Figure 3).

Smell attribute:

Data in Table (3) also show the significant effects of the main three factors on panelists' scores for smell attribute of the fresh cut spinach leaves during storage. Comparisons among overall means for packaging materials showed an overall lower score of smell attribute for samples packaged in PP_30 than those packaged in PP_22 or PS trays. Off odor may result from development of anaerobic conditions which lead to anaerobic respiration of the fresh produce inside the package. Comparisons among the overall means for dipping treatment showed that samples treated with ascorbic acid to receive the highest scores by the panelists, followed by those treated with potassium permanganate, whereas those dipped in water showed the lowest scores for smell attribute. Scores of smell attribute for the 6th and 9th days of storage did not vary significantly but were significantly lower than that of the 3d day of storage. When PS trays were the package material, samples treated with ascorbic acid remained acceptable for smell attribute till the 9th day of storage (Figure 3). Samples treated with permanganate was rejected at the 6th day of storage where as those treated with water was rejected after the 3d day of storage. Development of off odor indicated the presence of anaerobic conditions within the package atmosphere and reflects anaerobic respiration of the fresh cut spinach leaves (Verlinden and Nicolai, 2000 and Allende *et al*., 2004).

Texture attribute:

The three main factors showed significant effects on panelists' scores for texture attribute (Table 3). Samples packaged in PP22 showed significantly higher overall means for texture to those of samples packaged in PP30 but both were of similar magnitude to the scores of samples packaged in PS trays. Similarly, samples treated with ascorbic acid showed significantly higher value for texture attribute than those treated with water (the control), where as those treated with potassium permanganate recorded intermediate values (Figure 3). The overall means for texture scores of samples on the 6^{th} and 9^{th} days were similar but were significantly lower than those of the samples on the 3d day of storage . All samples treated with ascorbic acid or potassium permanganate and packaged in PS trays remained acceptable for texture attribute till the 9^{th} day of storage, whereas those treated with water showed fast decline in panelists' scores for texture after the 6^{th} day of storage (Figure 3).

Wiltness attribute:

The overall mean of wiltness attribute for samples packaged in PP_22 was similar to that of samples packaged in trays and both were significantly better than that of samples packaged in PP_30. Also The overall mean of wiltness attribute for samples treated with ascorbic acid was similar to that of samples treated with potassium permanganate and both were significantly better than that of the control (water treated samples). Also, the overall means of wiltness attribute for samples on the 6^{th} and 9^{th} days were not significantly different but were significantly lower than those of samples on the 3d day.

Samples packaged in PS trays and treated with ascorbic acid or potassium permanganate showed similar acceptable scores till the 9^{th} day of storage whereas those treated with water recorded unacceptable scores on the 9^{th} day of storage (Figure 3). These results is in agreement with Mangaraj *et al.,* (2009) who indicated that respiration of the packaged produce and permeation of package materials affect package atmosphere during storage. Successful MAP influence metabolism of the packaged product, improves moisture retention, minimizes the activity of decay-causing organisms and thereby, increases product shelf life.

CONCLUSION AND RECOMMENDATION

It can be concluded that samples packaged in PS trays and treated with ascorbic acid or potassium permanganate showed acceptable panelists' scores till the 9^{th} day of storage whereas those treated with water deteriorates quickly after at the 6^{th} day of storage. The PS trays were the best packaging material to preserve quality of the fresh cut spinach leaves followed by the PP pouches of 22µ particularly when samples were treated with ascorbic acid or potassium permanganate. PP pouches of 30µ was the worst package and water was the worst dipping treatments. It is recommended to use trays as the package material and ascorbic acid as a dipping treatment to preserve quality of the minimally processed spinach leaves for 9 days. Using PP pouches of 22µ may limit the shelf life to 6 days only.

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تأثير نوع العبوة ومعاملات ما قبل التعبئة على صفات الجودة للسبانخ محدودة التجهيز أثناء التخزين المبرد

سمير محمد حسين ربيع و أسرار يس إبراهيم محمد
معهد بحوث تكنولوجيا الأغذية - مركز البحوث الزراعية بالجيزة

يمكن استخدام أوراق السبانخ الطازجة محدودة التجهيز في إعداد العديد من الأطباق الشهية أو تناولها نيئة ضمن السلطات الخضراء وذلك لقيمتها الغذائية العالية ، ونظراً لحساسيتها للتلف والفساد فان فترة حفظها تكون عادة قصيرة فقد تم دراسة تأثير المعاملة بمحاليل غمر وهى الماء و برمنجانات البوتاسيوم بتركيز 0.02%، وحامض الاسكوربيك بتركيز0.2% و ثلاثة أنواع من مواد التعبئة والتغليف وهى: حوافظ pouches من البولي بروبيلين polypropylene (12 x 20 سنتيمتر) بسمك مختلف 22 ميكرون و30 ميكرون ، وكذلك أطباق من البولي استيارين polystyrene (PS) trays مغطاة over-wrapped بأغشية stretch film من مادة الكلوريد عديد الفنيليدين PVdC على صفات الجودة لأوراق السبانخ الطازجة محدود التجهيز للصنف البلدي. تم غمر شرائح الأ وراق (بعد تقطيعها بسمك 2 سم) في محاليل الغمر المذكورة وتعبئتها في العبوات تحت الدراسة والتخزين على درجة 4± 0.5 درجة مئوية ورطوبة نسبية 85±3 % وتم متابعة وتقييم التغير في صفات الجودة على مدي تسعة أيام أثناء التخزين بسحب عينات على فترات زمنية و تقدير: الفقد في الوزن ، المواد الصلبة الكلية ، pH ، الحمل الميكروبي ، والصفات الحسية.

أظهرت النتائج أن الفقد في الوزن بالنسبة لشرائح أوراق السبانخ المعبأة في عبوات البولي بروبلين بسمك 30 ميكرون و22 ميكرون كان محدودا، بينما أعطت العينات المعبأة في عبوات البولي استيارين فقداً أعلى في الوزن. و أعطت العينات المعاملة بالماء (كنترول) أقل فترة حفظ ، بينما أعطت باقي المعاملات تحت الدراسة أقل حمل ميكروبي أثناء التخزين. وأوضحت الدراسة أن المعاملة بحامض الاسكوربيك بتركيز 0.2% كانت الأفضل يليها المعاملة ببرمنجانات البوتاسيوم بتركيز 0.02%، ولوحظ تدهور اللون (اصفرار) مع العينات المعاملة بالماء ، بينما العينات المعاملة بحامض الاسكوربيك أعطت أعلى قيم للتقييم الحسي إثناء التخزين. ويوصي تعبئة أوراق السبانخ محدودة التجهيز بعد المعاملة بحمض الاسكوربيك باستخدام أغشية الإسترتش stretch PVdC over wrap مع أطباق البولي إستيارين (بقيت صالحة لمدة 9 أيام) عن استخدام أكياس البولي بروبيلين (بقيت صالحة لمدة 6 أيام). وكذلك يوصي باستخدام حوافظ البولي بروبيلين المثقبة عند تعبئة الخضار الطازج ذو معدل التنفس العالي لتجنب حدوث التنفس اللاهوائي ونمو الميكروبات اللاهوائية.

قام بتحكيم البحث

أ. د/ ممدوح محمد أحمد ربيع	**كلية الزراعة – جامعة المنصورة**
أ. د/ عبد الباسط عبد العزيز سلامه	**كلية الزراعة – جامعة كفر الشيخ**