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Expanding the Shelf-Life of Ready to use Shredded Icebergs by Using Oxidation - Reduction Potential controlled Release of Various Chlorine Concentrations in Washing Water

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ABSTRACT

Ready to use (RTU) shredded iceberg lettuce is a perishable product with a short shelf-life. Expanding the shelf life to seven days of this product as an economic need for the producers is the aim of this research. Fresh samples were harvested from the field and prepared according to the standard protocol of minimal processing fresh produce (receiving, removing dribs and pre-washing, washing, de-watering, packaging, and storing). Several concentrations of sodium hypochlorite were precisely optimized by oxidative/ reductive Potential (OPR) automated dozing pump under controlled neutral pH and cold temperature during washing regimen. Samples were vacuumed packed and kept tightly sealed at 4 °C for further analysis. Aerobic plate count and coliform as well as sensory evaluation were performed. Results showed that at a chlorine dose level less than 700 mV ORP, the shredded lettuce deteriorated due to elevated microbial load as well as from a sensory point of view. The level of 700 and 800 mV ORP extended shelf life for 7 days. Washing with chlorine levels higher than 800 mV showed alteration in sensory attributes although the microbial load was under control.

Results of this study show that the automated ORP chlorine dozing "in form of sodium hypochlorite" at 700 mV (50 ppm) and 800 mV (100 ppm) within controlled pH and temperature, followed by storing the vacuumed well-sealed packs at 4 °C, were the best measures taken to increase the shelf life of the shredded iceberg lettuce from 5 to 7 days. Furthermore, there was no significant difference between the uses of an ORP reading of 700 mV vs. 800 mV of chlorination. However, it is preferable to use the 700 mV ORP reading because it will release less chlorine in the washing water, hence, it will be healthier and more economical.

Keywords: Ready to Use fresh produce, Chlorination, lettuce (class Iceberg), Shelf-life, Minimal Processed Food.

INTRODUCTION

All varieties of lettuce are considered leafy vegetables which define as all vegetables of a leafy nature and of which the leaf and core are intended to be consumed raw (FAO)/WHO 2008). Among lettuce varieties there is the iceberg lettuce called (Lactuca sativa L) Iceberg lettuce consists of very soft and squishy leaves with more than 95% moisture content; it can be used in salads or as a garnishing item as a minimally processed fresh-cut and ready-to-use RTU form (Kank, *et al.*, 2007).

Ready to use (RTU) fresh produce is defined as the fresh-cut products that have been trimmed and/or peeled and/or cut into a usable item which is then packaged to offer consumers nutritional, conveniently ready to use the product while still maintaining its freshness (Lamikanra, 2002). Stepwise operations of trimming, core removal, slicing in shreds, washing, drying, packaging, and stored chilled are the well-identified activities in preparation of RTU fresh shredded iceberg lettuce and it is well known as a standard protocol for preparation and handling the RTU produce (King, *et al*., 1991; Riva, *et al.,* 2001; Kank, *et al.,* 2007).

RTU produce has limited shelf stability (Riva, *et al*., 2001; Kim, *et al*., 2005). While processing the leafy vegetables through cutting the surface area will be increased, this will lead the plant cells to be disrupted causing an increase in the respiration rates and production of phenolic acids and polyphenol oxidase which will lead to browning reactions as well (Saltveit, 2003; Riva, *et al*., 2001). Tissue disruption will cause not only a browning reaction but also the development of volatiles like alcohols, aldehydes, terpenes, esters, and acids (Belitz *et al*., 2004) accordingly, off-flavor of the product will be initiated (Saltveit, 2003; Couture, *et al*., 1993). Both the browning and the flavor-off are considered limiting factors that affect iceberg lettuce quality and shelf-life (Heimdal, 1995). On the other hand, fewer processing treatments lead to an accelerated deterioration of the produce (Ma, *et al*., 2017) due to the presence of microorganisms such as Pseudomonas spp., Erwinia spp., and lactic acid bacteria (Garcìa-Gimno and Zurera-Cosano, 1997). Studies showed that the presence of the aforementioned microorganisms was higher in the shredded lettuce compared to the intact produce which adversely affected the safety and quality of the product (Francis, 2013; Han *et al.* 2000; Takeuchi and Frank 2001; Seo and Frank 1999;). In addition, it is known that the high water activity and neutral tissue pH, will facilitate the microbial growth of shredded lettuce (Francis, 2013). Kang, *et al*., (2007) reported that a bacterial count found in minimally processed lettuce was too high compared with the control, it was detected around 10^5 CFU/g in shredded minimally processed lettuce compared to the range of 105 - 10^7 CFU/g in unprocessed/ untreated lettuce.

The consumer safety issue is an important field of concern. Based on FAO/ WHO report of microbiological hazard assessment (2008) leafy vegetables voiced the greatest attention in terms of microbiological hazards, as they have been associated with many outbreaks, additionally, they reported that the multiple activities of post-harvest may significantly contribute to exaggerating the incidence of the foodborne pathogen (Gil *et al*., 2009; FAO/ WHO 2008). The fresh produce may contaminate with foodborne pathogens such as E. coli and Listeria monocytogenes may in the field or at any point in the produce supply chain (Ferguson *et al*., 2013; Luo *et al*., 2010; Cooley, *et al*., 2006), causing dramatic produce associated-outbreaks (Abadias, *et al*., 2012; Buchholz, et al., 2011). The contamination may occur before, during, after harvesting and processing (Cooley et al., 2006).

Many management solutions were also proposed, among these, the chemical disinfectants such as chlorine (Jeddi et al., 2014; Francis, 2013). Washing with chlorinated water followed by appropriate cooling storage conditions proved to reduce the microorganisms' populations significantly (Francis, 2013).

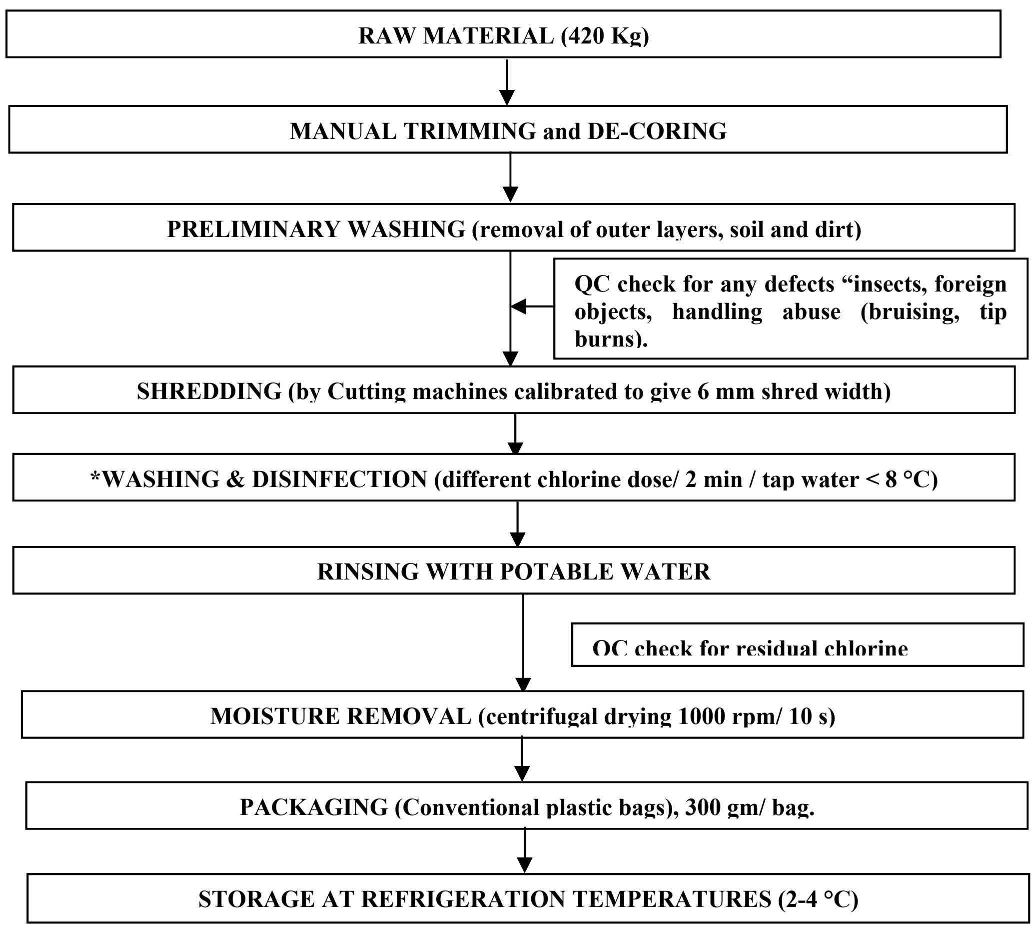
Chlorination may be introduced in several forms such as chlorine gas, sodium or calcium hypochlorite, or chlorine dioxide. It is considered highly effective and more economic than other methods such as ultraviolet irradiation or ozone (Pan & Nakano 2014). Other protocols were suggesting a sequential treatment where a combination of temperature and chlorine was adopted (Ono, *et al*., 2005). It has been reported that the typical concentration range of chlorine of 50–200 ppm and a contact time of 1–2 min was used for disinfection of fruits and vegetables in the food industry (Parnell *et al*., 2005). It is so important to optimize the level of chlorine during the period of contacting the chlorinated water with the product, thus oxidation-reduction potential (ORP) is one of the proposed systems used to optimize the chlorine release and enhance the performance of injections at proper timing (Sulsow, 2000).

Expanding the shelf life of the RTU iceberg is considered to be both a consumer's and a producer's desire. Currently, our local market introduces five days as an average period of shelf life of the fresh produce. Therefore, this research aimed to study the possibility of extending the shelf life of conventionally packed RTU shredded iceberg lettuce to seven days using chlorinated washing water under specified environmental conditions and optimized by an automated oxidation-reduction potential system to maintain the quality of RTU shredded iceberg.

Materials and Methods

Fresh produce: 420 Kg of iceberg lettuce was harvested early morning in the fall cycle from a private farm in Middle Ghore/ Jordan, and the whole quantity was processed immediately. This farm complies with the requirements of Global MacDonald's GAP and exclusively marketed its fresh produce after minimally processing techniques to one unique fast-food restaurant (MacDonald's/ Jordan).

Preparation of the RTU shredded iceberg cuts: the whole quantity of harvested iceberg lettuce was entered into the preparation room and treated automatically following a generic standard procedure of washing in chlorinated water before being de-watered and packaged into bags (Ferguson *et al*., 2013) with some modifications. At the chlorination step each 30 Kg was treated separately as per the following protocol, and each treatment was performed twice.



***@this step each 30 Kg was entered separately and continued automatically to the end of the process**

Primary washing includes cleaning with running potable water which is changed to prevent any cross-contamination that may encounter the sanitation or reduce the efficiency of washing in the next step. It has been reported that cross-contamination even for a short term in the pre-washing step may reduce the efficiency of the chlorinated water washing process (López-Gálvez *et al*., 2010).

Chlorine stock (Sodium Hypochlorite 12%) was purchased locally from Jordan National Chlorine Industries Co. LTD.

Treatment: Oxidation-Reduction Potential (ORP) used to control the chlorine injection as it is reported that using ORP in the combination of pH will ideally control and optimize the automated system and enhance the performance of injections in proper timing (Sulsow, 2000). The following Oxidation-Reduction Potential (ORP) of chlorine were used: 400, 500, 600, 700, 800, and 900 mV via calibrated dozing metering pump equipped with ORP Sensor (Prominent, model BT4A1005NPB900VA000D00- Germany). Each procedure was handled in duplicate.

Dozing parameters: to assure the effective sanitizing effect of chlorine, the pH parameter is controlled at 7.0 for all treatments. The pH is optimized to be at almost 7 during washing with chlorine water, as the OPR is correlated with pH (Bartz *et al*., 2001), on other hand, hypochlorite efficiency is limited by pH and temperature (Pan & Nakano 2014; Suslow, 2001), accordingly, those parameters were controlled. Ambient and water Temperature is kept < 8 °C during the treatment. The free chlorine against the ORP was as shown in Table 1.

At the end of the final washing process, residual chlorine was tested by colorimetric reaction with N, N-diethyl-1,4-phenylenediamine (DPD) using Lovibond AF112K testing kit, to ensure that the free residual chlorine does not exceed that of the potable water (< 0.5 ppm).

After spin-drying using a conventional centrifuge to dewater the shredded iceberg, the samples were packed tightly under vacuum; each in a 300 g/ tightly sealed polyethylene bag. Before filling, a sample of bags was selected to test the sterility using ATP sanitation Monitoring System "AccuPoint®- Neogen- P/ N 9602- USA"

Samples stored at ≤ 4 C. The refrigerated temperature was chosen based on Francis, (2013) findings, where he has shown that the storage temperature must be kept ≤ 4 OC, while storing at around 8 OC may have a negative impact in terms of pathogen survival and growth.

Table 1: Reading setting on auto dosing unit testing-Total Chlorine vs. ORP*

Instrument Set Reading	Chlorine Reading mV (ORP)	Total Chlorine
900	897	>200 ppm
800	808	100 ppm
700	705	50 ppm
600	607	20 ppm
500	502	10
400	418	<10

* At pH reading =7:0. The variation in pH readings was 6.9-7.2. The pH electrode was calibrated at two points of standard pH 7 and pH 10 buffer.

Analyses: At each storage time (day 1, 2, 3, 4, 5, 6, 7 days) aerobic plate count APC, total coliform, and Overall Visual Quality (OVQ) were performed in duplicate for each test and the result was the average of the reading of each test.

Aerobic plate counts (APC) were enumerated using Plate Count Agar, incubated at 35 ± 0.5 °C for 48 hours (Maturin and Peeler, 2001 (BAM)). APC was expressed as CFU/ g of fresh weight. Total coliform was enumerated using a solid medium technique, where violet red bile agar was used, and the incubation was continued at 35 ± 0.5 °C for 24 hours, followed by confirmation of coliform using brilliant green 2% lactose broth (35 ± 0.5 °C for 48 hours) according to Feng *et al*., 2020 (BAM) results expressed in CFU/g of fresh produce. OVQ for each sample was subjected to overall visual quality evaluation by a sensory panel. The evaluations were performed immediately after lettuce removal from storage conditions. A panel of 8 experienced judges ages 30 to 55 years old, who are members of the Jordanian Society for Sensory Evaluation of Food, or having sensory evaluation experience in leafy vegetables and other food commodities, after they were exposed to a short training course in lettuce quality evaluation. Three-digit code samples were presented one at a time, in random order, to the judges who made independent judgments. The judges were asked to evaluate OVQ based on leaf characteristics as the following: shade and uniformity of color (C), brightness (B), texture (T), presence or absence of defects including the off-flavor (D), and finally the overall acceptability (O). A nine-point scoring scale was used, in which 9 stood for excellent quality and 1 for very poor quality; the limit of acceptance was 5, according to Peryam and Pilgrim, 1957), for each treatment, the reading was the average of duplicate samples. The test was performed in the isolated well- aerated room, designated for the sensory evaluation test.

Samples were tested daily for a week. Those samples that proved to exceed the standard criteria limits of APC, TC, and OVQ were excluded and the test was aborted.

Statistical Analysis

Statistical analysis of data obtained in this study was performed using SAS statistical program (SASI, 2002). Analysis of Variance (ANOVA) was carried out using the General linear model procedure to elucidate the effect of chlorine level and days of cold storage on the various microbial and quality attributes. Means were separated using the Least Significant Difference test (LSD) to compare significance at a 5% level of probability ($P \leq 0.05$). Descriptive scoring was given for sensory evaluation upon the hedonic scale judgement; the score was the median of the panelist readings.

Results and Discussion

This study aimed to maintain the quality of shredded iceberg for seven days, accordingly, we adopt the OPR automated system to control and optimize the correct level of chlorine during the washing step. Different OPR- readings were studied, under neutral pH, and cold temperature.

APC Results for shredded lettuce at different chlorine treatment

The purpose of this experiment was to assess the microbial load in terms of aerobic plate count APC after the treatment with different dose levels of chlorine. Table (2) represents the results of aerobic plate count (APC) of the packed shredded iceberg at different chlorine concentrations for each day of the seven days storage interval. The standard cut-off point for the APC is $<10^6$ CFU/ g (Centre for Food Safety 2014).

The APC for raw cut produce of shredded iceberg lettuce was 6.5-9.0 X 10^5 CFU/g. One log decrement was noted directly after treatment with all chlorine levels, as shown in Table (2). A study of the effect of chlorination of pre-washing water at a level of 200 ppm on shelf-life of RTU iceberg lettuce conducted by Baur *et al*., (2005) showed a reduction of 1.63 logs in the aerobic bacterial count. The reduction in one log was noticed after washing at all levels of coloring, but the progress in bacterial load proceeded during the shelf-life period under the specific conditions of this research.

The APC of shredded iceberg lettuce started to exceed the fifth log on the third day for those treated with 400 mV chlorine, accordingly the test was terminated at that point for treatments of 400 mV as they were technically proved to exceed the cut-off standard limits (unfit for human consumption).

On the sixth day, the test was concluded for those samples treated with 500 mV chlorine, where the APC started to exceed the fifth log for that treatment. Samples treated at the level of 600, 700, 800, and 900 mV were proved to maintain the APC values at the fourth log for seven days. Furthermore, there were no significant differences between the APC results of those treated with the levels of 600-900 mV.

Table 2: APC Results

Raw	9.0 X 10^5	7.5 X 10^5	7.5 X 10^5	6.5 X 10^5	7.8 X 10^5	7.9 X 10^5
ORP Setting						
	400 mV/L	**500 mV/L**	**600 mV/L**	**700 mV/L**	**800 mV/L**	**900 mV/L**
Day1	a8.5 X 10^4	a6.2 X 10^4	a4.5 X 10^4	a2.5 X 10^4	a2.0 X 10^4	a1.8 X 10^4
Day2	Ab1.2 X 10^5	a7.1 X 10^4	a5.2 X 10^4	a3.1 X 10^4	a2.8 X 10^4	a2.2 X 10^4
Day3	Stop	a7.8 X 10^4	a5.8 X 10^4	a3.9 X 10^4	a3.6 X 10^4	a2.6 X 10^4
Day4		a8.3 X 10^4	a6.3 X 10^4	a4.5 X 10^4	a4.2 X 10^4	a3.5 X 10^4
Day5		a9.2 X 10^4	a6.9 X 10^4	a5.1 X 10^4	a4.8 X 10^4	a4.2 X 10^4
Day6		Ab1.2 X 10^5	a7.5 X 10^4	a6.8 X 10^4	a5.5 X 10^4	a5.8 X 10^4
Day7		Stop	a8.9 X 10^4	a7.5 X 10^4	a7.0 X 10^4	a6.9 X 10^4

All figures were average of two readings. A small letter in the superscript at the right side of the number indicates significant differences (P ≤0.05) between days (in the same column). A capital letter in the superscript at the right side of the number indicates significant differences (P ≤0.05) between treatments (in the same raw). When the log reached 5, the treatment for that level was stopped.

The total aerobic plate count for the pre-treatment was around 6.5-9.0 X 10^5 CFU/g (5.0 log CFU/g), these figures aligned with the study published by meeting report of FAO/ WHO (2008) on microbiological hazards in fresh leafy vegetables and herbs, the reported result was 5.2 Log CFU/ g for the pre-wash step of selected leafy items. Moreover, Kang *et al* (2007) had reported that the count of bacterial population of the minimally processed shredded iceberg was around the fifth log CFU/ g. In their study on leafy vegetables and herbs of domestic and Mexican origin, Johnston *et al.* (2006) reported that the aerobic bacterial load (APC) was 4.0 - 7.9 log10 CFU/g. Our results were aligned to those of results published by the FAO/ WHO study (2008), Kang *et al* (2007), and Johnston *et al.,* (2006). On other hand, a study in Nigeria market for assessing the bacteriological quality of some ready to eat vegetables as retailed and consumed revealed that the total aerobic plate count for lettuce at sales ranged between 2.0 x 10^7 to 5.7 x 10^8 CFU/g (seventh to eighth logs), (Abdullahi and Abdulkareem, 2010). Kaneko *et al*, (1999) had revealed that the bacterial populations in unprocessed icebergs were at a range of 10^5- 10^6 CFU/ g. The differences in our outcomes compared to those results may attribute to conditions of transportations and sale places, and the period between harvesting and provision into the market, while the lettuce samples of our current research were harvested and tested directly.

A significant reduction in the bacterial population in terms of APC was seen in all groups on the first day, flowed by a significant decrease in the microbial load in the treatment group using an ORP level of 400 mV. The starting load was 5 logs then dropped after the treatment to four CFU/ g logs, then elevated in the second day to 1.3 X 10^5 CFU/ g. Accordingly, the experiment assigned for that treatment level was terminated on the third day. Referring to table (1), the dose set at 400 mV represents the concertation of chlorine less than 10 ppm. We can reach the conclusion that less than 10 ppm level of chlorine was inadequate to maintain the microbial load to the accepted range even at controlled conditions during post-harvesting, pre-preparation, preparation, and controlled packaging ends with cold storage.

For those treated with 500 mV chlorine, the bacterial load was controlled until day five, wherein on day six the bacterial load increased significantly one log, the experiment then was terminated at that point. 500 mV represents 10 ppm chlorine concentration (Table 1), meaning that this concentration is insufficient to control the bacterial load in terms of APC for a week at cold storage.

Samples treated with 600, 700, 800, and 900 mV showed a controlled APC load (sustained at log 4) until the end of the week, indicating that washing at a level ≥ 20 ppm will control the bacterial load in terms of APC under well packed and a storage condition of ≤ 4 °C.

Baur *et al*., (2005) suggest 200 ppm of chlorine concentration for washing iceberg, while Sulsow (2001, 2000) recommended 100 ppm chlorine concentration for washing iceberg, yet, the results obtained by this research showed that 20 ppm chlorinated washing under the specified conditions of this research as a minimum can lead to control the APC at the log 4 for 7 days. It has been reported that the addition of chlorine as sanitizer may be questioned because of the possible formation of organic chlorinated compounds with potential carcinogenic effect (Gil *et al*., 2009; Rico *et al.,* 2007), accordingly, if we control the bacterial load at the least concentration of chlorine this would be useful in term of the safety of the RTU produce.

Total Coliform Results for shredded lettuce at different chlorine treatment

The purpose of determining the TC was to assess the sanitary quality of shredded lettuce at different chlorine treatments. It is well known that coliform test traditionally has been used as an indicator test for assessing the hygiene condition and contamination after processing.

Table (3) represents the results of the total coliform of the packed shredded iceberg at different chlorine concentrations with a seven days interval. The standard cut-off point for the total coliform as a hygiene indicator organism is $<10^2$ CFU/ g (Centre for Food Safety 2014). The experiment was terminated when the level exceeds 100 CFU/ g.

The total coliform for raw cut produce of shredded iceberg lettuce was 1.2 -1.5 X 10^3 CFU/g. Johnston et al. (2006) reported that the overall quality in terms of total coliform in random samples of domestic and Mexican leafy produce included herbs was less than 1.0 log10 to 4.5 log10 CFU/g; moreover, it has been reported that total coliform in minimally processed vegetables ranged from 0.7 to 6 log10 CFU/g (Jeddi *et al.,* 2014; Fröder *et al.,* 2007). It is documented that the total coliform was reduced around 1 log10 during the minimal processing of lettuce (Fröder *et al*., 2007).

One log decrement was noted directly after treatment with 400 mV chlorine, meaning that washing the shredded iceberg lettuce at this level of chlorine was not sufficient to reduce the TC to an acceptable limit, accordingly, the TC test was aborted on day 2. The ineffective dose of chlorine at that level may explain the recommended high doses of chlorine 50- 200 ppm (Rico *et al*., 2007; Parnell *et al*., 2005; Sulsow, 2000).

Significant two logs decrement was noticed at levels of 500-700 mV chlorine, and three logs reduction was noted at 800 and 900 mV (Table 3). An experiment conducted by Allende et al., (2008) to evaluate washing systems and commercial sanitizers, proved that after washing with 100 ppm chlorine water, the deduction in 2.2 logs in total coliform was recorded.

The total coliform started to exceed 100 CFU/g on day 4 for those samples treated with 500 mV. At the level of 600 mV, the total coliform started to exceed 100 CFU/g on day 7. The total coliform is controlled at a level of < 100 CFU/ g when samples are treated with 700-900 mV/L until day 7. There were no significant differences in total coliform at those mentioned doses. We can conclude that to control the level of TC < 100 CFU/ g of shredded iceberg lettuce should be washed at a level of 700 mV (50 ppm as indicated in Table 1) and more under the tight conditions of this research where packed samples must be kept under a cooling chain and stored at ≤ 4 °C.

Table (3): Coliform Results

Raw	1.5 X 10^3	1.3 X 10^3	1.2 X 10^3	1.2 X 10^3	1.3 X 10^3	1.25 X 10^3
ORP Setting						
	400 mV/L	**500 mV/L**	**600 mV/L**	**700 mV/L**	**800 mV/L**	**900 mV/L**
Day1	A1.2 X 10^2	Bb7.0 X 10^1	Bb5.2 X 10^1	Ba1.1 X 10^1	Ca8	Ca6
Day2	Stop	b8.5 X 10^1	b6.1 X 10^1	a1.9 X 10^1	b1.2 X 10^1	b1.1 X 10^1
Day3		b9.2 X 10^1	b6.8 X 10^1	a2.8 X 10^1	b1.8 X 10^1	b1.8 X 10^1
Day4		Aa1.2 X 10^2	Bb7.3 X 10^1	Ba3.6 X 10^1	Bb2.2 X 10^1	Bb2.1 X 10^1
Day5		Stop	b8.1 X 10^1	a4.7 X 10^1	b3.2 X 10^1	b3.2 X 10^1
Day6			b9.9 X 10^1	a5.5 X 10^1	b4.2 X 10^1	b4.5 X 10^1
Day7			a1.2 X 10^2	a7.4 X 10^1	b5.5 X 10^1	b5.2 X 10^1

All figures were average of two readings. A small letter in the superscript at the right side of the number indicates significant differences ($P \leq 0.05$) between days (in the same column). A capital letter in the superscript at the right side of the number indicates significant differences ($P \leq 0.05$) between treatments (in the same raw). When the log reaches 2, the treatment for that level was stopped.

Overall Visual Quality (OVQ) Results for shredded lettuce at different chlorine treatment

Sensory evaluation of appearance, color, flavor, and texture attributes, is one of the quality parameters that the overall quality of fresh produce is assessed for (Ma *et al.*, 2017). In this experiment, researchers agreed on considering a midpoint score (5) as a marginal accepted score. The results of OVQ are presented in table (4).

Table 4: Overall Visual Quality (OVQ) Results for shredded lettuce at different chlorine treatment

	400 mV					**500 mV**					**600 mV**					**700 mV**					**800 mV**					**900 mV**				
	C	**B**	**T**	**D**	**O**	**C**	**B**	**T**	**D**	**O**	**C**	**B**	**T**	**D**	**O**	**C**	**B**	**T**	**D**	**O**	**C**	**B**	**T**	**D**	**O**	**C**	**B**	**T**	**D**	
Day1	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9
Day2	7	7	7	8	7	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9
Day3	4	4	5	5	5	8	8	8	9	8	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9
Day4	/	/	/	/	/	6	6	7	7	6	8	8	8	8	8	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9
Day5	/	/	/	/	/	5	4	4	5	5	8	8	8	8	8	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9
Day6	/	/	/	/	/	/	/	/	/	/	7	7	8	8	7	8	8	8	9	8	8	8	8	9	8	7	8	8	8	8
Day7	/	/	/	/	/	/	/	/	/	/	7	7	7	7	7	8	8	8	9	8	8	8	8	9	8	7	7	7	7	7

Shade and uniformity of color (C), brightness (B), texture (T), and presence or absence of defects(D). Overall acceptability (O). A 9-1 scoring scale was used; where 9 stood for excellent quality and 1 for very poor quality; the limit of acceptance was 5.

The purpose of this research is to keep the quality of the shredded iceberg within specified shelf life (7 days) through which the microbial growth is controlled, the browning process is minimized, and the formation of off-odors which is developed during natural respiration of the product is retarded. Bearing in mind that the formation of off-odor can be a limiting factor affecting the quality attributes of shredded lettuce (Heimdal *et al*., 1995).

The Colour of the shredded iceberg is attributed as light green with no yellowing. Appearance must be predominantly 6 mm (1/4 inch) bright edges widths, where no welting or soft pale shreds appeared. Regarding the texture (using both fingers and mouth), shreds must be crisp, firm, fibrous to the bite or when pressed by fingers. Characteristic clean, mild, iceberg lettuce flavor and aroma including the absence of off-odor off- taste, and other defects (such as rots) were included under the attribute of absence/ presence of defects (D).

As shown in table (4), on day 3, a marked detrimental effect was recorded at a dose of 400 mV. This is not surprising as the APC and TC were higher than the acceptance range, and it has been recorded that as much the contamination is, as much the deterioration is encountered. Taking into consideration the above results of APC and TC in addition to those of sensory evaluation, it was clear that washing at a very low concentration of chlorine (400 mV <10 ppm) is not effective in extending the shelf life of RTU shredded iceberg; even if all the conditions are controlled during postharvest, preparation, pre-washing, washing, dewatering, packaging, and refrigerated storage at ≤ 4 °C.

At dose 500 mV, shredded lettuce scored 5 as overall acceptability on day 5. The experiment was stopped at that level. With compiling those results of the quality microbial analysis, we guarantee that washing at a level of 10 ppm of chlorine will be ineffective to maintain and extend the quality of shredded RTU iceberg even under the controlled conditions of this experiment including the cold chain conditions.

At levels of 600 to 900 mV, the sensory scores were higher than the acceptable value >5. The highest scores were noticed at levels of 700, and 800. At a dosing level of 900 mV, the quality attributes regarding the brightness, color, and texture were slightly reduced on days 6 and 7. It has been reported that high levels of chlorine may alter the taste and odor of fresh produce (Mahajan *et al.,* 2014). Our results were aligned with those reported by Mahajan *et al*., (2014).

A study by Allende *et al.,* (2008) to evaluate the overall sensory attributes of the iceberg after different washing systems and commercial sanitizers noticed that washing at 100 ppm chlorinated water will keep the OVQ within the acceptable limits after 8 days of storage.

According to Baur *et al.,* 2005, the pre-washing procedures delayed the progress of browning at cut edges and retarded the deterioration in the OVQ (Baur *et al*., 2005). Moreover, Fukumoto *et al*., (2002) stated that when washing with chlorinated water at low temperature the browning reaction will be delayed, additionally, the accumulation of phenolics will reduce as well. Nevertheless, in their review article, Rico *et al.,* 2007 reported that all the available washing and sanitizing strategies and techniques cannot guarantee the microbial load quality of RTU vegetables without compromising their sensorial quality (Rico *et al.,* 2007).

Conclusion

This research aims to optimize the chlorine levels by using ORP dozing pump during the washing step of shredded iceberg lettuce to increase the shelf-life up to one week. This is would be an unsophisticated economically affordable protocol for the producers of the RTU fresh produce. The process parameters were controlled at neutral pH, cold surrounding environment, chilled water, dewatering system, and the well-vacuumed packed shredded iceberg bags, which were stored at ≤ 4 °C. Results showed that at dosing levels of 700 and 800 mV, the bacterial load in terms of APC, and TC was within the acceptable limits. Moreover, the OVQ sensory tests were also at the highest scores at these dosing levels during a week of storage. No significant differences were observed between both 700 and 800 mV in terms of APC, TC, OVQ. With considering the economic feasibility, and based on minimizing the safety risk of chlorine, we recommend the dose of 700 mV rather than 800 mV.

From both health and economic point of view, we recommend the use of an ORP reading of 700 mV, which represents a 50 ppm of chlorine concentration under controlled ORP automated system to control the bacterial load and the quality, attribute for shredded iceberg and extend its shelf life from five to seven storage days.

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تمديد مدة صلاحية الخس (صنف الايسبرج) المعدّ للاستخدام وذلك باستعمال تقنية مضبوطة لمستويات متنوعة من الكلور تعتمد على جهد الأكسدة والاختزال

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الملخص

الخس المشرح (صنف ايسبرج) المعد للاستعمال (RTU) هو منتج قابل للتلف مع مدة صلاحية قصيرة، وتمديد مدة الصلاحية إلى سبعة أيام كمتطلب سوق من المنتجين هو الهدف من هذا البحث، وتم حصاد عينات من الحقل، وتم إعدادها مباشرة وفقاً للبروتوكول القياسي المعتمد للمنتجات الطازجة (استقبال المحصول، ثم مرحلة التنظيف والغسل الأولي، فالغسل النهائي، تنشيف الماء، التعبئة والحفظ). وقد استخدمت عدة تراكيز من هيبوكلوريت الصوديوم مضبوطة من خلال مضخة وحساس لفرق الجهد بالاكسدة والاختزال في ظروف معتدلة من الاس الهيدروجيني ودرجة حرارة التبريد أثناء عملية الغسل، ولقد تم تعبئة العينات بأكياس مفرغة من الهواء والاحتفاظ بها في 4 درجة مئوية لمزيد من التحليل لاحقاً.

وتم إجراء عد الطبق الهوائي والعد الكلي للكوليفوم وكذلك تم إجراء التقييم الحسي للعينات، وأظهرت النتائج أنه في مستوى الكلور أقل من 700 (ORP) ميليفولت، تدهورت صلاحية الخس المشرح بسبب الحمل الميكروبي المرتفع، وأظهرت النتائج أن الصفات الحسية قد تأثرت سلبياً بشكل واضح، وبينما على مستوى تركيز 700 و 800 ميليفولت من الكلور استطعنا رفع مدة الصلاحية إلى 7 أيام، وتبين من هذا البحث أنه في حال كان مستوى الكلور أعلى من 800 ميليفولت فأنه، وعلى الرغم من أن الحمل الميكروبي تحت السيطرة، فقد لوحظ تغييراً سلبياً في السمات الحسية للخس المشرح المعد للاستعمال.

وتظهر نتائج هذه الدراسة أن الاحتفاظ بكلا المستويين من الكلور "هيبوكلوريت الصوديوم" على 700 ميليفولت (50 جزء في المليون) و800 ميلفولت (100 جزء بالمليون) وأس هيدروجيني معتدل في ظروف منضبطة في التبريد ثم التعبئة في أكياس مفرغة وخزنها على درجة حرارة 4 درجة مئوية، هي أفضل طريقة لضمان أطول مدة صلاحية تصل إلى 7 أيام، إذْ لم يكن هناك تدهور في الصفات الحسية (لا تلون ولا تلف) مع السيطرة على مستوى مقبول من الحمل الميكروبي، بيد أنه ولغايات الجدوى الاقتصادية، وضمان أمان المنتج من ازدياد تركيز الكلور فإننا ننصح بمستوى 700 ميلفولت مضبوط آلياً.

الكلمات الدالة: جاهزة استخدام المنتجات الطازجة، الكلورة، الخس (صنف الايسبيرج)، مدة الصلاحية، الحد الادنى من المواد الغذائية المصنعة.