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Trends in Minimal Processing of Lettuce

Manpreet Kaur[1*], Sangram S. Dhumal[2] and Manish Kumar[1]

[1]Division of Vegetable Sciences, ICAR-Indian Agricultural Research Institute, New Delhi, India

[2]Department of Horticulture, College of Agriculture (Kolhapur),
Mahatma Phule Krishi Vidyapeeth, Rahuri, India

*Corresponding Author E-mail: mkaur89096@gmail.com

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ABSTRACT

Lettuce is an expanding salad vegetable for supermarket in India and abroad. It has been emerged as a valued salad alternative in minimally processed food products. Minimal processing gives an additional value to lettuce in terms of convenience and time saving. Mechanical damage to the cells during processing, however, is a major limitation to shelf life of minimally processed lettuce which enhances the microbial growth, enzymatic browning and moisture loss. Different minimal processing techniques such as use of post harvest chemicals, irradiations and modified atmosphere packagings (MAP) have been employed for different category of lettuce. These techniques can be adopted either individually or in combination which extends the shelf life of the products through suppression of respiration, ripening and subsequent senescence and microbial activity. MAP inhibits the ethylene induced effects, balances the O_2 and CO_2 concentrations and reduces the moisture loss due to the moisture barrier properties of the packaging. This review paper aims to supply information about different approaches being employed to extend shelf-life and quality of minimally processed lettuce. A brief description of the MAP technology is also given.

Key words: *Lettuce, Minimal processing, Shelf-life, Quality, Enzymatic browning*

INTRODUCTION

The minimal processing techniques have emerged to meet the challenges of replacing traditional methods of preservation by retaining nutritional and sensory quality[1]. Demand for minimally processed fruits and vegetables has increased rapidly in Europe and USA and in developing countries like India, demand is expected to represent 25% of the total food market [2]. Lettuce is an expanding salad category for supermarket in minimally processed food sector. India is the 3rd largest producer of lettuce with a production of 1.08 lakh MT followed by China and USA[3]. Among the three main type *viz*; leaf, head and cos or romaine, leaf lettuce is commonly grown in India and used as minimal processed form. It perceived as a maintained and high valued product since it provides naturally maintained quality with the true effects of freshness, high content of water, vitamins, minerals, fibers and low energetic food. It is highly requested commodity by salad bars and fast food centers in India and abroad.

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The marketing period of lettuce is limited to about 3 to 4 days for whole leaves tissues. In the processing of vegetables like lettuce, physiological deterioration, biochemical changes and microbial degradation is at faster rate[4] which lowers the shelf life due to disruption of tissue and cell integrity and thus require special attention for quality and safety[5]. Storage temperature is the single most important factor affecting spoilage of fresh-cut products. However, numbers of many other preservation techniques are currently being used by the fresh-cut industry such as antioxidants, chlorines and modified atmosphere packaging (MAP)[6] different packaging material and cold storage, MAP, controlled atmospheric storage, UV-C illumination, ozone bubbling as antimicrobial and antibrowning agents, calcium as firming agent and allicin as antimicrobial agent, which could be a solution to delay quality losses and extend shelf life and freshness of minimally processed lettuce[7, 8,9,10]. The new alternative processing and preservation techniques to improve the quality and shelf life of fresh-cut fruits and vegetables are reviewed below in this chapter.

PROCESSING TECHNIQUES AND METHODS

Post-harvest chemicals

The minimal processing induces surface damage increasing the tissue respiration which leads to biochemical deterioration of the fresh-cut fruits and vegetables[11]. Many chemical treatments which include antioxidant like ascorbic acid, citric acid and surfactants like sodium hypochlorite (NaOCl) and chlorinated water showing broad biocidal effectiveness due to high antimicrobial action have been tested in ready to eat food products in recent years. Chlorine-based chemicals, especially liquid chlorine and hypochlorite, are most widely used at the level of 50-200 ppm free chlorine and contact time less than 5 minutes as a sanitizers for decontaminating fresh produce[12, 13].

Washing with chlorinated water has been traditionally applied to decontaminate vegetables Chlorine dioxide (ClO_2) is accepted for use in washing fruits and vegetables and many studies have demonstrated its antimicrobialactivity[14, 15]. Chlorine dioxide (3 ppm) and ozone (0.34 ppm) treatments alone or in combination to Iceberg lettuce effectively influenced the microbial population (1.17 log cfu/g) after 6 hrs with increase in phenolic compound[16]. Chlorine dioxide (60mgL^{-1}/10 min), per acetic acid (100mgL^{-1} for15 min) and ozonated water (1.2mgL^{-1}/ min) are effective alternative sanitizers to sodium hypochlorite (150mgL^{-1} for 15 min as they promoted reduction of 2.5, 1.1, and 0.7 log cycle respectively on count of microbial load of minimally processed product and increased shelf life by 6 days as compared to sodium hypochlorite treatment which had 12 days shelf life[17]. The maximum reduction of more than 3 log cfug^{-1}and 2 log cfug^{-1} were observed after washing with acidified sodium chlorite at concentration of 1gL^{-1} and 0.25 to 0.5gL^{-1} respectively[8]. In contrast, Lopez-Galvez *et al*.[18] evaluated the efficacy of chlorine dioxide (3mgL^{-1}) and sodium hypochlorite (100mgL^{-1}) sanitation solution on prevention of cross contamination of fresh-cut lettuce by *E. coli* and found no significant reduction in *E. coli* content in both inoculated and cross contaminated lettuce. However, sanitizers used inactivated most *E. coli* cells that passed from inoculated product to wash water.

Calcium lactate has been reported to be a good alternative to calcium chloride because it avoids the bitterness or off flavors associated with this salt[19]. Calcium lactate was tested as fresh-cut lettuce and carrots sanitizer and compared with chlorine[20]. The use of calcium-based treatments presented a further advantage; in some cases the final product can significantly increase the calcium content[21], which might enhance the appreciation of these products, due to the fact that the awareness of consumers on the benefits of calcium is relatively high. The shelf- life of fruits and vegetables can be extended by calcium treatments by maintaining the cell wall integrity and firmness by cross-linking with cell wall and middle lamina pectin to form calcium pectate[22] The wide use of calcium lactate for delicate fruit and products having high senescence index is reported by Luna-Guzman and Barett[19] in fresh-cut cantaloupes and Anino *et al.*[21] in apples. Rico *et al*.[7] observed reduction in enzyme activity(~300 units g^{-1}), higher oxygen decline(~6%) and reduced enzymatic browning in ozone (1mgL^{-1}) treated fresh-cut lettuce and recorded maximum polyphenol oxidase activity with best quality markers in fresh-cut lettuce treated with calcium lactate (15gL^{-1}) after 10 days of storage at 50^0C. The calcium lactate at high temperature (50^0C) recorded higher value of crispness coefficient and maintained qualities upto storage period of 12 days[20].

The use of ozonated water has been applied to fresh-cut vegetables for sanitation purposes reducing microbial populations and extending the shelf-life of some of these products[23.] Many researchers,[24, 25] have opined that ozone treatment have a beneficial effect in extension of storage life of different commodities such as lettuce, broccoli, cucumber, apples, grapes, oranges, pears, raspberries and strawberries by reduction of microbial populations and by oxidation of ethylene. Grass *et al*.[26] reported ozone as a strong antimicrobial agent with high reactivity, penetrability and had spontaneous decomposition to a non-toxic product. Modified atmosphere packed fresh-cut green asparagus (*Asparagus officinalis*) treated with aqueous ozone treatment (1mgL^{-1}) at 3^0C for 25 days inhibited the enzymatic activities including phenylalanine ammonia lyase (PAL), superoxide dismutase (SOD), ascorbate peroxidase (APX) and glutathione reductase (GR)[27].

Immersion solution (2gL^{-1} citric acid, 1 gL^{-1} calcium chloride and 250gL^{-1} garlic extract) is the best mixed immersion solution as they controlled enzymatic browning, chlorophyllase activity and had minimum weight loss up to 9th day of storage of fresh-cut lettuce. However microorganism growth was not significantly controlled[28,29] for the first time reported that natural additive, allicin (0.2 and 1%) could be used in prolonging the shelf life of fresh lettuce var. Angustana Irish. Fresh-cut lettuce treated with 1% allicin and stored at 4^0C with >90% relative humidity retained its color and maintained visual quality without impairing flavor with maximum antioxidant activity after 6 days of storage. Allicin (1%) treatment lowered total viable count by 2.52 log cfu/g and yeast and mold count by 1.59 log/g as compared to control with prevention of tissue browning. 5% citric acid or 100% sugar syrup dipping treatment inhibit the browning and polyphenol oxidation in fresh-cut lettuce with good sensorial attributes up to 12 days when stored at 0^0C and 95% relative humidity[30].

UV-C Irradiations

Low dose gamma irradiation is very effective in reducing bacterial, parasitic, and protozoan pathogens in raw foods. Irradiation was approved by the FDA for use on fruits and vegetables at a maximum level of 1.0 kGy[31]. The use of Ultraviolet- C (254 nm, UV-C) on minimally processed lettuce, showed that this technique can reduce deterioration of the produce by effectively reducing microbial populations (Fan *et al.*)[32]. Fresh-like quality of minimally processed lettuce was retained better during storage when irradiated with 0.5kGy dose as compared to irradiation levels of 0.81 and 1.1kGy Fan *et al*.[33] reported that the fresh cut iceberg lettuce samples treated with warm water (47^0C) and irradiated at 0.5 and 1 kGy had best sensory quality without significant loss in texture, vitamin C, or total antioxidants. Fan and Sokorai[34] found that the overall visual appearance was best for fresh-cut iceberg lettuce irradiated at 1 or 2 kGy. However, lettuce samples irradiated at 4 kGy were much less firm than non irradiated samples after 3 and 7 days of storage at 3°C. Further, in 2003 they reported that Irradiation at 1, 2, 3 and 4 kGy reduced the ascorbic acid (AA) of fresh-cut lettuce but did not affect the total ascorbic acid content (TAA) [33]. Furthermore, total ascorbic acid levels decreased during storage for both non irradiated and irradiated lettuce.

Shredded iceberg lettuce irradiated at 0.55 kGy dose reduced inoculated *E. coli* 0157: H7 population by5 log without causing adverse effects on sensory attributes. Low to medium dose irradiation of shredded iceberg lettuce could improve safety and microbiological shelf-life of lettuce for retail or food service[35] Niemira *et al*.[36] opined that low irradiation doses (0.3 or 0.6 kGy) and modified atmosphere packaging could be combined to reduce the level of *L. monocytogenes* in minimally processed endive, thereby serving to protect the consumers. Also the exposure of minimally processed (shredded) ice-berg lettuce to 0.7 kGy gamma irradiation reduced the population of *Salmonella spp*. by 4 logs and *E. coli* by 6.8 logs without impairing the sensory attributes as reported by Goularte *et al.*[37].

Modified atmosphere packaging on Lettuce

Modified atmosphere packaging, a novel technology has been extensively used to achieve safety and/or to prolong the shelf life of fruit and vegetables products. MAP replaces original atmosphere gas partial pressure with nobles gases, such as helium(He), argon (Ar), xenon (Xe), nitrous oxide (N_2O) or super atmospheric oxygen (O_2) and carbon dioxide (CO_2)[38] MAP could be adopted for different type of raw or processed fruits and vegetables produce such as lettuce, broccoli, spinach, melon, apple, cucumber, carrot and some other, either individually or in combination which extends the shelf life of the products through suppression of respiration, ripening and subsequent senescence, microbial activity due to diversity of effect by low O_2, elevated CO_2, by inhibition of ethylene induced effects and by reduction of moisture loss due to the moisture barrier properties of the packaging film[39]. Fresh vegetables continue to respire, consuming oxygen and producing carbon dioxide and water vapor. Very successful applications of MAP are reported for fresh-cut pineapple[40], kiwifruit[41] and Honeydew[42]. Beneficial modified atmosphere within fresh-cut fruits and vegetables packages are attained by correctly choosing packaging material that will provide the appropriate levels of oxygen and carbon dioxide into packets[43]. Delaying in the packaging of fresh-cut Romaine lettuce for 12 hrs at 5^0C in ambient air before packaging decreased the fermentative volatile production, off-odor development and alleviating CO_2 injury and increased browning[44].

Mattos *et al.*[45] studied the effects of two active modified atmosphere packaging (5% O_2, 5% CO_2 and 2% O_2, 10% CO_2-balance N_2) and one passive modified atmosphere as control on quality attributes and physiological responses of fresh-cut crisp head lettuce stored at 5^0C and 85% relative humidity for 14 days. They observed no significant difference among treatments for chlorophyll degradation. Mechanical stresses induced by processing increased CO_2 and ethylene evolution and O_2 consumption. Polyphenol oxidase and peroxidase activity recorded slight increase on processing but reduced over storage period. The active modified atmosphere has no significant effect on quality attributes and physiological behavior of fresh-cut lettuce for storage period of 14 days. Fresh-cut Romaine lettuce packaged in 60 µm thick LDPE (Low Density Poly and MDPE (medium-density polyethylene) film bags and stored at 0^0C had very low quality deterioration in terms of firmness, color, ascorbic acid and sensory evaluation concerns with reduced browning. Hamza *et al.*[46] opined that modified atmosphere of 10% CO_2 plus 1% O_2 could maintain the fresh like quality of minimally processed Romaine lettuce stored at 4^0C upto 16 days. The increase CO_2 content in the package atmosphere to 10% or more reduced enzymatic browning due to wounding and improved visual quality. After packaging the shredded lettuce in sealed bag of monooriented polypropylene (OPP) film and in polyethylene (PE) trays overwrapped with a multilayer polyolefin (RD 106) or plasticized polyvinyl chloride (PVC) film; Pirovani *et al.*[47] reported that modified atmosphere within the package changed from normal air composition to about 1.5% O_2 and 12% CO_2 for OPP bags and to about 17-18% O_2 and 1-2% CO_2 for PVC-PE trays and RD106-PE trays after 8 days of storage. The visual sensory quality of the product was retained better in OPP bags than in the other ones. Gloria Lopez *et al.*[45]

assessed quality changes in packaged commercial salad products stored at 5^0C upto 20 days. They opined that browning of cut lettuce pieces was minimal and the quality of commercial salads decreased with time. The concentrations of O_2 and CO_2 in package were at 0.2-1.5% and 5-30% respectively. They further stated that the concentrations of fermentative volatiles compound like ethanol and acetaldehyde in lettuce tissues were significantly correlated with off odour and aroma score with decrease in sugar content from 12% to 20%. Castaner *et al*.[49] stored white, green and red section of Lollo Rosso lettuce in perforated or non-perforated PE film with active atmosphere modification at 5^0C for 7 or 14 days. The quality of white and green tissues was best maintained in modified atmosphere storage whilst red tissue was better preserved in air. Segall and Scanlon[50] stored minimally Romaine lettuce under three modified atmosphere viz. 3% O_2 combined with either 6%, 10%, or 14% CO_2 for 20 days. They found that O_2 consumption rate deceased with increased CO_2 levels. The O_2 levels in packaging equilibrated at 7% to 11%. They further observed that MAP delayed the development of tissue discoloration but had no effect on microbial growth. Out of three levels of CO_2 tried, 10% CO_2 was found to be effective and thereby increased shelf life by 50%. Hemidal *et al.*[51] observed that use of an atmosphere of 80 kPa O_2 plus 20 kPa CO_2 delayed browning of shredded lettuce under MAP for 10 days as compared to air. MAP Treatments of 6kPa O_2+7kPa CO_2 in oriented polypropylene (OPP) and polyethylene-perforated bags improved the sensory quality, avoided the loss of green colour, decreased the development of pithiness and retarded the growth of microorganisms in celery sticks stored at 4^0C for 15 days than compared to the control (air)[52] . In another experiment, Gil *et al*.[53] observed no effect on total flavonoid content of Swiss chard spinach beets leaves (cv. Green) packed with 7%O_2+10%CO_2 upto 8 days of storage, although increased flavonoid extraction during cooking in boiling water was noted. However, 50% decrease in ascorbic acid content over the initial, especially in MAP-stored Swiss chard, after 8 days of cold storage was noted. Peiser *et al*.[54] observed that food service garden salad packages (containing commercially processed iceberg lettuce, carrot and red cabbage) obtained from 5 California processors had 0.2-1.0% O_2 and 5-20% CO_2 after over 15 days of storage at 5^0C.Acetaldehyde and ethanol developed in the tissues at concentrations of 2-22 µl/kg and 50-1500 µl/kg, respectively, by the 'Best if used by Date' (BIUD). Packages from all processors were above the limit of salability for overall visual quality by the BIUD, although off odors were detected.

Two plastic films maintained the nitrous oxide concentration within the bags, with some slight differences in O_2 and CO_2 concentration when packaged fresh in two different non-micro perforated plastic film bags (polypropylene+polyamide, PP+PA and polypropylene+polyethylene tetraphthalate, PP+PET) with three modified atmospheres (97%N_2O+3%O_2; 50%N_2O+47%N_2 and 97%N_2+3%O_2). They found that that the two plastic films maintained the nitrous oxide concentration within the bags, with some slight differences in O_2 and CO_2 concentration. No significant differences were observed in texture, colour and sensory quality (off odour, off-flavor, texture and overall acceptability) of lettuce. All treatments delayed microbial growth (mesophiles, yeast and mold, *psychrophiles* and *enterobacteria*) and compared to control in air, reduced weight loss in lettuce. MAP treatments maintained very good quality and freshness of 'Iceberg' lettuce for 4 days. Slight increase in off odour and off-flavor observed during storage were below the moderately acceptable limit even after 9 days[55]. A high variability in growth of *E. coli* O157:H7 under low temperature conditions with maximum growth rates (log cfu/day) corresponded to 0.14 (95% Cl: 0.06-0.31), 0.55(95% Cl: 0.17-1.20) and 1.43 (95% Cl: 0.82-2.15) for 8, 13 and 16^0C respectively[56]. Sharma *et al.*[57] reported that leafy green lettuce packaged with commercial modified atmosphere conditions in gas permeable film with N_2 and stored at 4^0C decreased *E. coli* O157:H7 populations on lettuce. However, storage of leaf lettuce under near ambient air atmospheric conditions promoted higher expression levels of O157:H7 virulence factors in lettuce. A combination of high O_2 (75kPa) and low CO_2 (15kPa) in packaging of fresh-cut lettuce caused reduction in the aerobic mesophilic bacterial count, avoiding anaerobic fermentation, and delayed color change up to 10 days for fresh-cut lettuce stored at 7^0C[10]. Further they observed that number of psychrotrophic bacteria, *Coliform* and *Lactic acid bacteria* (LAB) were influenced by packaging of fresh-cut lettuce in PP bags with MAP (3kPa O_2 and 5kPa CO_2)[58]. No growth of microorganisms in the fresh-cut salad mixes containing lettuce, packaged in a low or a high barrier film with an initial oxygen level of 95kPa and stored at 40C was reported. Gil *et al.*[59] found no browning and no increase in the amount of soluble phenyl propaniods in the mid ribs of minimally processed Lollo Rosso lettuce during storage in 2-3% O_2 and 12-14% CO_2 at 5^0C.

On investigated the effect of chemical sanitizers (100ppm chlorine dioxide, or 100 ppm sodium hypochlorite) alone or in combination with packaging methods (air, vacuum, N_2 and CO_2 gas packaging) on inactivating *E. coli* O157:H7 in spinach at $7±2^0C$, Lee *et al.*[60] found that treatment with chlorine dioxide and sodium hypochlorite significantly decreased levels of *E. coli* O157:H7 by 2.6 and 1.1 log cfu/g respectively. In addition to this, significant difference of about 3-4 log cfu/g of *E. coli* O157:H7 populations between samples packed in air and other packaging methods following treatment with chemical sanitizers was observed after 7 days of storage at 7±20C. Radziejewska-Kubzdela *et al*.[61] reported that modified atmosphere with the content of 5% or 10% CO_2, 2% O_2 and balanced N_2 in the packaging of celeriac flakes during storage at 4 and 15^0C for 12 days, inhibited growth of mesophilic, psychrophilic and coliform bacteria. Also in cut Chinese cabbage, modified atmosphere packaging (MAP) enhanced the reduction of total aerobic and coliform bacteria in cut chinese cabbage, irradiated at dose up to 2kGy with air CO_2 or CO_2 /N_2 packaging during refrigerated storage for 3 weeks. Izumi *et al.*[62] monitored the physiological and quality of spinach (cv. Sunbest) leaves during storage in air and controlled atmospheres (0.5, 1 and 2% O_2) at 0^0C (for 28 days), 10^0C (for 9 days) and 20^0C (for 5 days) and observed that low-O_2 atmospheres did not affect the development of decay, or populations of aerobic mesophilic and lactic acid bacteria at any of the storage temperatures tested. However, Barriga *et al.*[63] found no effect of controlled atmosphere (3%O_2, 3%O_2+5%CO_2 or 3%O_2+10%CO_2) on the studied microbiological population in fresh-cut lettuce samples. However, a 3% O_2 + 10% CO_2 atmosphere maintained acceptable visual quality of lettuce, without appreciably affecting microbial environment for 12 days at 4^0C.). Koseki and Itoh[64] concluded that 100% N_2 gas packaging delayed degradation of fresh-cut lettuce and cabbage stored at 1, 5 and 10^0C for 5 days but did not significantly affect the growth of microbial population of total aerobic bacteria (TAB), coliform bacteria and psychotrophic bacteria in or on fresh-cut lettuce and cabbage. They also found that microbial growth in or on the fresh-cut lettuce and cabbage was inhibited at 1^0C storage regardless of packaged atmospheric conditions.

Storage

Lou *et al*.[65] investigated the impact of storage temperature and duration on the fate of Escherichia coli on packaged salads of iceberg and fresh-cut Romaine lettuce stored at 5^0C and 12^0C. The storage of fresh-cut Romaine lettuce at 5^0C allowed *E. coli* to survive but limited its growth while 12^0C storage facilitate proliferation of it more than 2.0 log cfu /g with significant decline in visual quality after 3 days. Spinardi and Ferrante[66] studied the effect of storage temperature (4^0C and 10^0C) on quality changes of fresh-cut baby lettuce. They observed increase in lipid peroxidation from 0.27 µmol/g to 0.36 and 0.49 µmol/g in leaves stored at 4 and 10^0C respectively. Total ascorbic acid slightly decreased initially and increased again after 7 days of storage. Grzegorzewska[67] compared the influence of three storage temperatures *viz.* 0^0C, 6-8^0C and 18-20^0C on storage ability of fresh-cut crisp lettuce. He observed good quality was maintained the longest time for 6 days at 0^0C and shortest time at 18-20^0C. After 2 days at 18-20^0C browning was visible and after 4 days there was symptoms of rotting. Serea *et al*.[68] determined that ascorbic acid, total phenolic content and antioxidant activity of the cultivars (Roderick, Markies and Locarno) of lettuce were depended not only temperature, but were also significantly affected by the duration of storage. Major loss were recorded after 6 days and for temperature above 2^0C for all cultivars when stored for different time periods (0,3,6,9,12 and 15) at storage temperatures (0,2,4, and 6^0C). Moreira *et al*.[69] studied the changes in lettuce leaf quality at two abusive temperature (8 and 15^0C) and compared with optimal storage temperature of 0^0C. They observed that degradation rate of ascorbic acid in lettuce leaves stored at abusive temperatures was from 2.7 to 2.9 times faster than at 0^0C. The microbial quality was retained 1-6 and 4 times longer at 0^0C with respect to 8 and 15^0C storage respectively. Manolopoulou *et al*.[70] investigated that fresh-cut cabbage could be stored upto 14 days and 7 days respectively at 0^0C and 5^0C when treated with ascorbic acid while citric acid (1%) combined with low temperature storage prolong the shelf life of minimally processed cabbage for 22 days and retained its color as well as overall acceptance than conventional chlorine treatment. Lu *et al.*[71] concluded that the growth rate and maximum permissible limit of bacteria in fresh cut lettuce samples stored at 0^0C are lower with longer lag time as compared to that of samples stored at 4 and 25^0C. They also opined that the efficacy of preservation of fresh-cut lettuce treated with chlorinated water was significantly better than that of non-treated lettuce under the same storage temperature.

Hurdle technology

Hurdle technology is the combination of different preservation techniques as a preservation strategy. The most important hurdles commonly used in food preservation are based on controlling temperature, water activity, acidity, redox potential and use of preservatives, modified atmosphere and competitive microorganisms (e.g. lactic acid bacteria)[72]. Lee *et al.*[73] reported that aqueous ClO_2 (50LL $^{-1}$) combined with UV-C radiations (10 KJm^{-2}) reduced the initial population of total aerobic bacteria in Romaine lettuce by 1.07 $logcfug^{-1}$ compared to 6.36 $logcfug^{-1}$ for control; it also reduced the population of yeast and mold in the sample of 1.85 log $cfug^{-1}$. Beltran *et al.*[23] studied the effect of UV-C activated ozonated water (10 and 20 mgL^{-1}) on fresh-cut lettuce and opined that respiratory activity of fresh-cut lettuce was delayed which maintained visual appearance and helped in controlling browning during storage in air. The active MAP (4 kPa O2+ 12 kPa CO2) helped in controlling total microbial growth as well as reduction in count by 2.0 log in relation to sample stored in air at the end of storage for 13 days at 4^0C. Diaz and Hotchkiss[74] studied the effect of gas mixtures of 0/10/90, 3/0/97, 5/30/65, 20/0/80 (O2/CO2/N2, v/v) on shredded lettuce inoculated with nalidixic acid-resistant *E. coli* (ATCC 35150) and placed at 13 or 22°C and observed that the aerobic plate count (APC) growth was inhibited in 5% O_2/30%CO_2/65%N_2 at 13^0C compared to all other atmospheres. Further, they opined that the extended shelf-life in Modified atmosphere allowed *E. coli* to grow to higher numbers compared to air-held lettuce.

CONCLUSION

In India, the ready-to-eat fruit and vegetable market is under developmental stage even though it is having number of health associated benefits. The awareness needs to be heightened for the newly developed novel products like fresh cut salads. There is need to develop the simple and cost effective processing line for fresh cut vegetables like lettuce. Due to stricter regulations on the use of chlorine urge to find the new alternatives which satisfies consumers and maintains a balance between sensory and quality. The intelligent packaging systems like MAP and SMART packaging with nano based food products and nano biosensors should be evaluated with combination of novel methods of packagings and storage, edible as well as antimicrobial films. There is need to investigate the antimicrobial effect of super atmospheric oxygen in fresh-cut product safety. Optimization of wide variety of polymers and gas mixtures available for packaging of individual commodity is necessary and should be given more focus in regard with MAP in future.

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