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The Effect of Film Wrapping and Nitrogen Flushing of Produce Atmosphere on the Storage Quality of Lettuce

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Abstract: Freshly harvested lettuces of the iceberg variety were used after vacuum cooling in these trials on storability at 5, 15 and 22^0. Five treatments were observed; four of them consisted of wrapping the lettuces with 4 different types of polymeric films. The 5th treatment consisted of placing some of the lettuces previously wrapped with one of the films into polyethylene bags, then flushing each bag with nitrogen gas for 2 min and sealed with a heat sealer. The samples were stored at 5, 15 and 22°C for 21 days and assessed for weight loss and quality. All the lettuces stored at 5°C retained their quality for 21 days. Nitrogen flushing gave best results with respect to weight loss and overall quality. Lettuces stored at 15 and 22°C spoiled rapidly and 50% or more were considered unsalable after 12 and 9 days, respectively.

Key words: Film wrapping, nitrogen flushing storage quality, lettuce

INTRODUCTION

Leafy vegetables are valued in many parts of the world for their flavour and nutritional value. Lettuce, *Lactuca sativa* (L) is a good example of a popular green salad which is an important item of diet around the globe. It is eaten fresh and therefore has to reach the market in very good condition, i.e. with minimal weight loss, wilting, discolouration, or fungal growth. Compared with other leafy vegetables, lettuce deteriorates rapidly. This is mainly due to water loss and fungal attack which causes decay. This short shelf life, allied to the fact that the market demand for vegetables such as lettuce is irregular means it has to be stored in order to meet peak market demands (Schofield *et al.*, 2005).

The storage of lettuce can be improved by vacuum cooling immediately after harvest, to remove field heat. It can also be stored at low temperatures immediately above freezing and in controlled atmosphere which involves modification of the oxygen/carbon dioxide ratio in the storage atmosphere. A modified atmosphere, which relies on respiratory activity for the adjustment of the internal atmosphere within film wrapped lettuce, may also be used.

There is an increasing level of use of polymeric films for fruit and vegetable packaging, the main objective being to obtain uniform produce appearance. Another objective is to prolong the shelf life of produce such as fruits and vegetables. Due to differential permeabilities of polymeric films to oxygen, carbon dioxide and water vapour, they modify the atmosphere within the film wrapping. Polymeric films also slow down respiratory and transpiratory activities of produce and thereby extend the shelf life. In general, polymeric films are used as open top containers to protect the heads from scratching and bruising. They do not, however, significantly extend the shelf life of produce since the tops are open.

Harvested fruits and vegetables are living tissues and therefore continue to respire and lose water in transpiration. They are also subject to chemical and physiological changes, all of which contribute to gradual deterioration of the produce after harvest.

The storage life of fresh fruits and vegetables is limited by the rate of biochemical, physiological and microbial deterioration. Storage losses arise from the continuation of normal metabolic processes such as respiration, water loss and the development of fungal or bacterial diseases; these losses are caused by the environmental conditions under which the produce is held such as temperature, humidity, presence of micro-organisms and composition of the atmosphere surrounding the produce (Ryall and Lipton, 1979). Browning is another major problem with fresh-cut lettuce and is controlled by packaging in <1% O_2 and 10% CO_2 atmospheres (Lopez-Galvez *et al.*, 1996; Smyth *et al.*, 1998).

Techniques used to preserve fresh fruits and vegetables during transportation, storage and handling through the market chain are generally based on refrigeration with or without the control of ambient gas atmospheres. The overall objective of these treatments is the slowing down of respiration and disease development in order to extend produce life. Control of humidity, generally by relatively crude techniques, is also exercised to retain product weight and quality. The storage life of untreated and non-cooled lettuce is low compared with other vegetables; various storage and transport methods have therefore been developed. These include keeping lettuce under refrigeration for 2 to 3 weeks (Aharoni and Ben-Yehoshua, 1973). Refrigerated storage entails the control of environmental factors such as temperature, relative humidity and movement in the storage environment. Commodities such as lettuce which require prompt cooling after harvest and storage at a controlled and constant temperature must be held in refrigerated storage. Refrigerated storage of lettuce, provided that the temperature is above freezing, extends the keeping quality of the leaves. Due to a combination of low temperatures and high humidity, however, only a relatively few vegetables can be stored for six months or more, even when held in environments optimum for the commodity. Many vegetables such as lettuce are not adapted for prolonged cold storage, for such commodities, refrigerated storage is usually at a wholesale level, with the purpose of avoiding over supply in the retail market during periods when supply of the commodity exceeds consumer demand.

Other techniques include hypobaric storage, controlled atmosphere storage and modified atmosphere storage.

Hypobaric storage is a storage system in which the produce is maintained at a given temperature in a sealed container at a constant sub-atmospheric pressure. It is also ventilated with air saturated with water vapour by continuously evacuating the container with a sealed vacuum pump. The storage of fresh produce at sub-atmospheric pressures results in reduced oxygen levels, thus reducing the respiratory rate. Hypobaric storage is new and it is rather complex and very expensive. Additional engineering developments and economic analyses would be needed before commercial application of the method becomes a reality (Salunke and Wu, 1973). Applications are currently limited to high value produce such as cut flowers and meat.

In controlled atmosphere storage, the product is confined in an air-tight chamber at the desired temperature. For lettuce and all other vegetables this is well below 5°C (Cantwell and Suslow, 2006). Product respiration consumes oxygen and gives off carbon dioxide; this lowers the oxygen level and causes carbon dioxide levels to rise. When the desired concentration of carbon dioxide and oxygen is reached after several days or even weeks, the oxygen at this level is maintained by ventilating with fresh air.

Experimental storage of lettuce in controlled atmospheres was carried out by Lipton (1971) to determine the effects of various combinations of oxygen and carbon dioxide on lettuce quality.

They found that lettuce stored in atmosphere containing 2.5, 5 or 10% carbon dioxide developed brown skin lesions, while those held in 0% carbon dioxide did not show any such symptoms.

They also observed that the percentage of affected heads increased progressively with increasing carbon dioxide concentrations. They also found that incidence of brown skin, severe enough to be objectionable to the consumer, increased from 3.8% of the heads after 7 days at about 3.3°C to 23.7% after an additional 4 days at about 10°C. These workers also found that low oxygen concentration increased the susceptibility of lettuce to brown skinning. They also noted that butt discolouration was less severe at higher carbon dioxide levels than at lower concentrations after 7 days in controlled atmospheres at 3.3°C. This result agreed with the finding of Watada *et al.* (1964), who also noted that carbon dioxide inhibited butt discolouration in lettuce. They concluded that the danger of injury from increased carbon dioxide levels in a controlled atmosphere outweighs any minor benefits the system might provide for lettuce.

In modified atmosphere storage, the commodity is sealed in a plastic film such as polyethylene, polyvinyl chloride or polypropylene. The respiration process consumes oxygen and causes carbon dioxide to accumulate within the film covering. The plastic bag serves as a modified atmosphere compartment and retains water vapour while allowing slow entry of oxygen. Unlike controlled atmosphere storage, the concentration of oxygen and carbon dioxide is not precisely controlled but is governed by a complex relationship of temperature, type of product and quantity and the gas permeability characteristics of the film used.

Stewart *et al.* (1967) investigated the effect of film wrapping of lettuce on quality, in a series of simulated experiments at transit temperatures of 0-5°C. They found that film wrapping significantly reduced the percentage of heads with commercially important crushing and bruising, giving an average reduction of 50% in bruising, compared with the control. They also found that film wrapping significantly retarded butt discolouration and that butt discolouration did not increase rapidly during 4 days of storage at a higher temperature of about 10°C.

The most important factor affecting the storage quality of lettuce is temperature, with the optimum around 0°C (Pratt *et al.*, 1954), since the leaves are easily damaged by freezing. The temperature of harvested lettuce should be lowered as quickly as possible by vacuum cooling (MAFF, 1978; Lipton and Barger, 1965). Another important factor affecting storage quality of lettuce is relative humidity. Ceponis and Haufman (1968) in their experiments have shown that unwrapped lettuce lost 15.2% of its weight at 5% relative humidity, 12.6 at 70% relative humidity and only 4.1 at 90% relative humidity. Although high relative humidity in other film wrapped lettuce tested resulted in a high incidence of decay, the optimum relative humidity recommended for the storage of lettuce is 95% (MAFF, 1978).

The objective of this study is to determine the effect of wrapping lettuce of the headed or 'Iceberg' variety using films of varying permeabilities, allied with nitrogen flushing of the plastic film package, on the shelf life of lettuce.

MATERIALS AND METHODS

Freshly harvested iceberg lettuces were used in this experiment; they were obtained from G.S. Shropshire Ltd., near Ely, Cambridgeshire, from a uniform population of first grown lettuces. Harvesting was carried out during the day and 175 heads were selected at random. They were then randomly allocated to the various treatments, i.e. wrapped in different types of film. The heads were vacuum cooled for approximately 30 min to remove field heat and were then transported rapidly to the storage chambers (Appendix 1).

The treatments used in this experiment were:

Treatment A = Lettuce wrapped in Vitafilm MT (PE
Treatment B = Lettuce wrapped in Resinite PF-M
Treatment C = Lettuce wrapped in "Letis Wrap" Polypropylene
Treatment D = Lettuce wrapped in "Grace Film" and used as control
Treatment E = Lettuce wrapped in "Grace Film" overwrapped with polyethylene film and flushed with nitrogen gas for 2 min.

Vitafilm MT and Resinite PF-M were supplied by TDRI, Slough. Letis Wrap and Grace Film by G. Shropshire Ltd.

The Treatments were to be held at 3, 12 and 25°C in storage cabinets representing, respectively average temperatures in a cold store, in a room in autumn or winter and in the summer. In reality, however, temperature attained were 5, 15 and 22°C over the three-week period of the test, due to temperature control problems in the storage cabinets. Average humilities in all three cabinets were 58%, although 90% relative humidity was required in each one. The oxygen concentration within the wraps for samples of lettuce chosen at random was measured with the Servomex oxygen analyzer; carbon dioxide concentration could not be measured due to lack of appropriate equipment.

The treatments consisted of wrapping the heads of lettuce in the various films mentioned previously. A completely randomized factorial design was used for this experiment, the two factors being temperature and type of film with each treatment replicated 12 times, except for treatment C. Treatment C. was replicated 12 times at 3 and 12°C, but only 6 times at 25°C, due to an error in the supply of the right number of lettuces wrapped in this film.

The method used for the experiment was as follows:

In the field, the first 36 heads of lettuce were randomly chosen and wrapped in Vitafilm MT, i.e. Treatment A; the next 36 in Resinite PF-M, i.e. Treatment B and 72 in "Grace Film".

An additional 30 heads were supplied wrapped in "Letis Wrap" and allocated Treatment C (Appendix 2).

At Silsoe College, the 72 heads in the Grace Film were further subdivided into two treatments, D and E.

Treatment D consisted of 36 heads wrapped in "Grace Film" and used as control, the objective being to compare the commercially used film against the others. It was felt that this kind of control would be more meaningful than naked lettuce, since this condition actually never exists in commercial storage. Treatment E consisted of 36 heads wrapped in "Grace Film" each of which was then placed in a polyethylene bag, flushed with nitrogen gas for 2 min and sealed.

Each treatment was then divided at random into three groups of 12 replicates and stored at 5, 15 and 22°C for 21 days. Weight measurements were made on day 1, day 3 and at 3 day intervals thereafter.

Market quality was evaluated subjectively at 3-day intervals from day 9 from a standpoint of consumer acceptability. Overall quality including firmness, butt discolouration, decay, wilting and visual appearance, was evaluated rather than each one individually. This is because these defects generally appear together and are so considered by the consumer in deciding whether to buy the lettuce or not. The oxygen levels within the wrapped lettuce heads were measured by withdrawing 50mL of sample from the internal environment using a syringe and passing this through the Servomex oxygen analyzer. This measurement was done on the 12, 15, 18 and 21st days on the randomly chosen samples from each treatment and expressed as percent oxygen. Measurement of the internal oxygen was started on the 12th day and at 3-day intervals thereafter, instead of starting on day 1, with a view to causing minimum disturbance within the packages (Appendix 3 and 4). The data on weight loss were subjected to analysis of variance according to Steel and Torie (1980).

APPENDIX I

Specifications of films used

1. Vitafilm MT (polyethylene)

Water vapour transmission : 465 g/m2/day @ 38°C/ 90% R.H. (estimate 90g/ m2/day @75% R.H. according to Dr. J. New of TDRI)

Oxygen transmission : 7750 cc/m2/day/atm @ 23°C

Carbon dioxide transmission : 52700 cc/m2/day/atm @ 23°C

Manufacturer : Goodyear (UK) Ltd

Address unknown.

2. Resinite PF-M (PVC)

Water vapour transmission : 120 g/m2/day @ 25°C/ 75% R.H

Oxygen transmission : 12300 cc/m2/day/atm @ 23°C

Carbon dioxide transmission : 2000 cc/m2/day/atm @ 23°C

Manufacturer : Borden (UK) Ltd, Thermoplastics Division

Colley Lane Estate, Bridgewater,

Somerset TA6 5LA

3. Letis wrap (polypropylene)

No water vapour, oxygen and carbon dioxide transmission rates available. Appendix II for manufacturer and technical specification.4. Grace film Cryovac-MPY (polyolefin)

No specifications available.

Manufacturer : W.R. Grace Ltd, Northdale House North Circular Road, London NW10 7HU

APPENDIX II

LETISWRAP

Technical specification

Gauge : 15 micron

Yield : 74 M2/Kilo

Tensile properties		ASTM D. 822
Tensile Strength Nmm^{-2}		
Machine Direction	:	24.86
Cross Direction	:	21.38
Elongation at break %		
Machine Direction	:	668.5
Cross Direction	:	635.9
Impact Resistance $g\ m^{-1}$		
ASTM D 1709	:	10.32
Burst strength N/Cm2	:	8.9
Optical Properties		
Haze % ASTM D 1003	:	1.6
Gloss % ASTM D 523	:	94

APPENDIX III

Table showing relative humility of air over various saturated solutions of salts

Saturated salt solution	Temperature 5°C	10°C	15°C	20°C	25°C	30°C	35°C	40°C	50°C
	Relative humidity (%)								
Potassium sulphate	96	98	97	97	97	96	96	96	96
Potassium nitrate	96	95	94	93	92	91	89	88	85
Potassium chloride	98	88	87	83	85	85	84	82	81
Ammonium sulphate	82	82	81	81	80	80	80	79	79
Sodium chloride	76	76	76	76	75	75	75	75	75
Sodium nitrite	-	-	-	66	65	63	62	62	59
Ammonium nitrate	-	73	69	65	62	59	95	53	47
Sodium diodromite	59	58	58	55	54	52	51	50	47
Magnesium nitrate	58	57	56	55	53	52	50	49	46
Potassium carbonate	-	47	44	44	43	43	43	42	-
Magnesium chloride	34	34	34	33	33	33	32	32	31
Potassium acebate	-	21	21	22	22	22	21	20	-
Lithium chloride	14	14	13	12	12	12	12	11	11
Potassium hydroxide	14	13	10	9	8	7	6	6	6

APPENDIX IV

Statistical analysis of variance

1. **Preliminary analysis**

Block effect - F11, α0.05= 1.79 > 1.5872
No significant block effect at the 5% level
Treatment effect - F14, α0.05= 1.67 < 3.3808
Significant treatment effect at the 5% level

2. **Further analysis**

Factor 1 Effect - F4, α0.5 = 2.37 <6.4607
Significant factor 1 effect at the 5% level
Factor 2 Effect - F2, α0.05 = 3.00 5.0261
Significant factor 2 effect at the 5% level
Interaction - F8, α0.05 = 1.94 >1.4295
No significant interaction at the 5% level
LSD = T X S.E of differences between means
LSD for Factor 1 = 1.960×13.71
= 26.87
LSD for Factor 2 = 1.960×10.62
= 20.82

3. Test of differences between means

Factor 1

A	B	C	D	E
372.26	421.27	289.43	375.28	423.44

Factor 2

5°C	15°C	22°C
377.02	404.12	407.87

Any pair of means linked by a line is not significantly different.
Any pair of means not linked is significantly different.

RESULTS

Weight loss and saleability: Table 1 shows the mean weights of samples in each treatment measured every three days for the duration of the tests. The blank spaces indicate that more than 50% of the samples in the treatment had become unsalable and the test stopped at that stage.

Tables 2-4 indicate percentage of the samples of lettuce per-treatment which were unsalable after 9 days, based on subjective evaluate of quality described earlier.

Analysis of variance: The treatments were subjected to a preliminary analysis of variance according to Steel and Torie (1980) and the results indicated that there were significant differences between treatments at the 5% level of probability, but not between blocks.

Two factors were used in these trials, factor 1 was the type of film used and factor 2 was the storage temperature and therefore a further analysis of variance was carried out on the treatments. The results indicated significant differences between factor 1 treatments and between factor 2 treatments, but no significant interaction between factors 1 and 2.

The treatment means were tested by the least significance, or LSD method. These tests indicated no significant differences between treatment B and treatment E means. There was a significant difference between means of treatments E, A, C and D, but no significant difference between the means of treatments C and D and between A and C.

Furthermore, no significant difference between means at 15 and 22°C were indicated, but there were significant differences between means at 5, 15, 5 and 22°C.

Visual quality: Figures 1-3 show lettuces wrapped with the different films and stored at the indicated temperatures. The samples used for photography were obtained by random sampling.

Internal atmosphere composition: The servomex oxygen analyzer was used to monitor the oxygen concentration in treatment E samples.

Table 1: Mean weights of samples in grams with time of storage

		Day							
Temperature °C	Treatment	1	3	6	9	12	15	18	21
5	A	250.8	350	348.71	346.49	345.1	343.46	341.93	340.4
	B	425.6	424.8	423.28	420.93	419.2	417.28	415.46	413.83
	C	352	350.4	347.1	342.86	340.5	337.98	335.6	333.2
	D	394	392	389.84	386.25	383.4	380.28	377.7	375.23
	E	409.4	409.34	409.7	409.35	409.2	409.14	408.91	408.68
15	A	411.5	409	406.96	404.6	400.9	396.88	-	-
	B	424.9	423.82	421.48	418.6	415.43	412.55	-	-
	C	412.9	411.2	408.64	405.63	401.02	396.12	-	-
	D	390	389.1	384.2	278.73	374.58	370.2	-	-
	E	426.8	426.53	426.2	426.04	425.82	425.23	-	-
22	A	369.8	368	361.84	352.89	332.53	-	-	-
	B	470.4	467.5	459.55	444.68	424.42	-	-	-
	C	414.4	412.2	402.53	391.5	375.1	-	-	-
	D	401.2	398.96	389.94	379.31	363.5	-	-	-
	E	451.3	451.24	450.13	449.03	447.5	444.66	-	-

Table 2: Percentage of total number of heads of lettuce per treatment which were unsalable with time of storage at 5°

Treatment	Days in storage 9	12	15	18	21
A	0	0	0	8	42
B	0	0	0	0	0
C	0	0	0	0	8
D	0	0	0	8	17
E	0	0	0	0	0

Table 3: Percentage of total number of heads of lettuce per treatment which were unsalable with time of storage at 15°

Treatment	Days in storage 9	12	15	18	21
A	75	100	100	100	100
B	25	75	92	100	100
C	25	75	83	83	100
D	0	33	75	83	100
E	0	25	50	83	100

Table 4: Percentage of total number of heads of lettuce per treatment which were unsalable with time of storage at 22°

Treatment	Days in storage 9	12	15	18	21
A	100	100	100	100	100
B	92	100	100	100	100
C	50	100	100	100	100
D	92	100	100	100	100
E	75	100	100	100	100

Fig. 1: Twenty one days old lettuce wrapped with Cryovac MPY and flushed with nitrogen, stored at 5°C

Fig. 2: Tweleve days old lettuce wrapped with Cryovac MPY and flushed with nitrogen, stored at 15°C

Fig. 3: Tweleve days old lettuce wrapped with Cryovac MPY and flushed with nitrogen, stored at 22°C

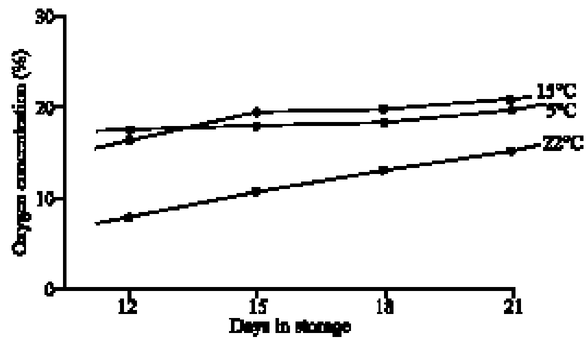

Fig.4: Effect of storage on oxygen concentration within polythene bags containing film wrapped heads of lettuce.

Due to the use of holes and slits in treatment C and D to ease evaporation during vacuum cooling and the presence of obvious voids in the wrappings of treatments A and B, no meaningful variations in the atmospheric oxygen level could be expected.

Figure 4 shows the effect of storage on the oxygen concentration within the nitrogen-flushed bags. It shows an increase in the oxygen concentration within the bag with the time of storage, this increase being greatest at 22°C.

DISCUSSION

The results show that there is a steady loss of weight during storage at all temperatures, this finding agrees with published information on the evapo-transpiration which relates the amount of water loss during storage to temperature, thus the higher the temperature, the greater the water and hence weight loss (Ryall and Lipton, 1979; Hughes *et al.*, 1981, Saltveit, 1997). It further reveals that the weight loss was least in those samples flushed with nitrogen, at all temperatures, compared with the other treatments. The negligible weight loss among treatment E samples, Table 1, at 5°C might be a significant contributory factor to attaining the 100% saleability at the end of 21 days. Although treatment B gave the same result at this temperature, the samples flushed with nitrogen had the best overall quality, with less butt discolouration, firmer heads, fresher and with no brown discolouration or wilting, compared with the other treatments. The above trend is similar at the higher temperatures, where the nitrogen treated samples had slightly better overall quality compared with the others, though still unsalable.

These findings compare well with the results of a preliminary test during which two types of film and nitrogen flushing were compared against unwrapped controls stored at 15°C. It was found that nitrogen flushing resulted in the least weight loss and best overall quality at the end of the trial, which lasted for 17 days.

The reduction in weight loss caused by overwrapping with films was expected, since this would minimize evapotranspirational loss. It agrees with Lipton and Barger (1965) who showed that packing of lettuce in plastic films reduced weight loss by reducing water loss. Furthermore, the extension of quality for 21 days at 5°C agrees with Steward *et al* (1967) who showed that film wrapping significantly improved lettuce quality at 0-5°C.

Statistical analysis of results indicated that both of treatment and temperature were important in these trials so that the type of treatment used and the temperature of storage would determine the overall quality of the lettuce after storage. However, the results of the analysis indicated no difference between the means of treatments B and E, but indicated that treatment E was significantly better than the other treatments. Table 3 indicates that there was no difference in salability for treatments E and B after 21 days, compared with the others. All the evidence indicates that nitrogen flushing gave the best result and was therefore the best treatment, for improving storage quality, this finding agrees with the work of Aharoni and Ben-Yehoshua (1973) which showed that flushing of polymeric bags containing head lettuce resulted in better quality. The improvement in quality obtained from nitrogen flushing may result from the very low oxygen atmosphere which exists in the bags after flushing, typically 0.5- 2% from previous tests after 2 min of flushing. This low oxygen level may then reduce metabolic activity to very low levels, resulting in reduced transpirational weight loss and extended storage life. The rise in oxygen concentration as shown by Fig. 4, may be partly caused by diffusion of atmospheric oxygen into the bag as a deficit exists between the oxygen level inside the film and that outside the film.

Problems experienced during the trials included lack of information on nitrogen transmission rates for all the films used and the variability in the amounts of nitrogen gas released from the cylinder as the pressure inside fell, for the chosen 2 min time limit.

CONCLUSIONS

Nitrogen flushing resulted in lower weight loss, extended storage life and the best overall quality at all temperatures.

For extended storage and best quality, lettuce should be stored at low temperatures such as 5°C for 21 days of storage.

Lettuce could not be stored more than 12 days at 15°C and 9 days at 22°C, although even at these temperatures, nitrogen flushing resulted in less weight loss than other treatments.

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