**Note** ～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～～

# Moisture-Proof Corrugated Fibreboard Box Effectively Reduces Water Loss in Lettuce during Storage

Daniel Z. K. WAMBRAUW*, Yuko SATO*, Naoki SUGINO*,**,
Saki MATSUMOTO* and Hiroaki KITAZAWA*,***

Reducing water loss is important for the maintenance of the commercial quality of lettuce. In this study, we evaluated the water loss of lettuces that were not enveloped in any plastic films during storage for 6 days at 10 °C using a moisture-proof corrugated fibreboard box (MPB) and a non-moisture-proof corrugated fibreboard box (control). We also evaluated the changes in SPAD value as an index of green colour, soluble solids, and ascorbic acid content during storage. The water loss in lettuce packaged into MPB was significantly lesser than that of control after 6 days of storage. In addition, MPB did not negatively influence other qualities of lettuce. Thus, we conclude that the use of MPB would contribute to keeping the quality of lettuces during distribution.

Keywords：Brix, corrugated fibreboard box, fresh produce, SPAD value, ascorbic acid

## 1. Introduction

Lettuce is recognised as one of the 14 designated vegetables in Japan[1]. The shipment and output of lettuce in 2017 were $543.3\times10^6$ kg[2] and JPY $101.8\times10^9$ [1], respectively. Therefore, maintaining the quality of lettuce during distribution, including storage and handling is important. However, it is known that lettuce deteriorates easily due to its high water content. Thus, water loss is a major factor that reduces the quality of lettuces after harvest[3]. Moreover, for distribution during business to business (B to B), lettuces are sometimes packaged directly into corrugated fibreboard box i.e. corrugated fibreboard box without being enveloped in plastic films, and this consequently leads to the increase of water loss.

Maintaining a high relative humidity inside the package contributes to the reduction of water loss in fresh produce[3]. Considering that fact, recently, the use of moisture-proof corrugated fibreboard box (MPB) has been proposed for quality maintenance of fresh produce especially with regards to the water content[4]. However, little is known about the ability of MPB to effectively maintain the quality of lettuce during storage. Thus, in this study, we investigated the applicability of MPB, with regards to its effectiveness in preventing water loss in lettuce.

## 2. Materials and Methods

### 2.1 Corrugated fibreboard boxes

Two different types of single-layered corrugated fibreboard boxes: a water-repellent liner board

*Food Research Institute, National Agriculture and Food Research Organization (NARO), 2-1-12, Kannondai, Tsukuba, Ibaraki 305-8642, Japan
**Headquarters, NARO, 3-1-1, Kannondai, Tsukuba, Ibaraki 305-8517, Japan
***Corresponding author, Tel: +81-29-838-7191, Fax.: +81-29-838-7996, E-mail: ktz@affrc.go.jp (H. Kitazawa)

(Rencoat, Rengo, Osaka, Japan) as the control and MPB with dried latex films and fillers on both surfaces of the liner board of the box (Damp-Proof 2, Rengo, Osaka, Japan). The corrugated box used as control did not have any water-proof property, although the surface of the liner boards was coated with a water-repellent agent. The flute type of both corrugated fibreboard boxes was “A” (K280 × HS200× K280), and the thickness was approximately 5 mm. The external dimensions of the box were approximately 400 × 600 × 360 mm (width × length × height) (Fig. 1).

## 2.2 Details of Lettuce

A head lettuce (*Lactuca sativa* L. cv. unknown) was harvested from a local farm in Ibaraki Prefecture, Japan. Each lettuce was enveloped in a polystyrene film and packaged into a conventional corrugated fibreboard box and delivered to Food Research Institute 1 day later. The average mass of lettuce was approximately 450 g. Prior to the storage test, all the lettuces were stored at 10°C and approximately 90 % relative humidity (RH) for 24 h in the dark.

## 2.3 Storage Conditions

The storage test started from 17 October, 2019. Eight boxes were prepared: four boxes for control and four boxes for MPB. The number of lettuces in each box was 16. Considering a B to B situation, the polystyrene films were removed from the lettuces, and the lettuces were packaged directly into the boxes. All the boxes were placed in two rows of four tiers for 6 days of storage. The boxes were stored at 10°C and approximately 90 % RH in the dark, and they were randomly shuffled every day throughout the experimental storage period.

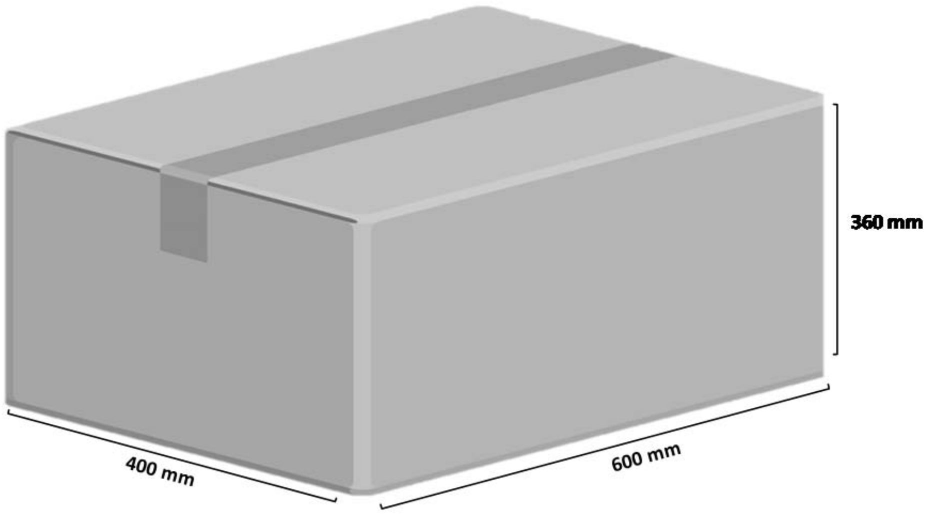


**Fig. 1** Dimensions of corrugated fibreboard box

## 2.4 Measurement Items

### 2.4.1 Mass Loss and SPAD Value as an Index of Colour Change

The mass values of 6 lettuces out of 16 were taken at 0 (initial mass prior to storage examination), 1, 4, and 6 days of storage period. The comparison of initial mass at 0 day and each storage period was used to determine mass loss in the lettuces.

Changes in the green colour of the leaves were also determined at the same period as the measurement of mass loss, using a non-destructive chlorophyll meter (SPAD-502plus, Konica Minolta Optics, Tokyo, Japan). The chlorophyll content was calculated as the SPAD value[5)]. The third to sixth leaves from the outer part of the lettuces were used to determine the SPAD value as an index of colour change.

### 2.4.2 Soluble Solids Content and Ascorbic Acid Content

A whole lettuce was cut diagonally into eight parts, and the core was removed. Next, each cut sample except the core was mixed. Exudates from approximately 5 g of sample were extracted by a hand squeezer and dropped onto the prism of a saccharimeter (PR-201α, Atago, Tokyo, Japan). We then measured the soluble solids content (Brix %).

Samples (10 g) were immediately dipped into 90 mL of 5 % metaphosphoric acid solution, homogenised using a homogeniser (HG30, Hitachi Koki, Tokyo, Japan), and filtered using a filter paper (No. 6, 110 mm, Advantech Toyo, Tokyo, Japan). Ascorbic acid (L-ascorbic acid) content in the filtered liquid was measured using a reflectometer (RQflex 20®, Merck, Darmstadt, Germany). The measured values were corrected considering the concentration of ascorbic acid due to water loss, using a formula described in a previous study by Kitazawa et al. (2011)[6)].

The measurement periods and the number of measured lettuces for each item were same as described in **2.4.1** and **2.4.2**.

## 2.5 Statistical Analysis

All results are presented as means ± standard deviations. For pair-wise comparison between control and MPB, we did not assume the variance of homogeneity. Therefore, we used Welch's *t*-test within the same measurement period using a spreadsheet program (Excel 2016, Microsoft Japan, Tokyo, Japan). The *p*-value was set to 0.05.

## 3. Results and Discussion

For the control, the mass loss (%) of lettuces on days 1, 4, and 6 after the start of the experiment was 1.1, 2.3, and 3.5, respectively (Fig. 2). For MPB, the values on the same days were 0.6, 2.0, and 1.9, respectively. Significant differences were found between both boxes on day 6 after the start of the experiment; the mass loss for MPB was 40 % lower than that for the control during storage. The most important factor associated with mass loss in fresh produce is the decrease in water content via evapotranspiration[7), 8)]. In addition, the degree of water loss of fresh produce depends on the surrounding relative humidity[9)]. Thus, in this study, our results suggest that MPB maintained a high humidity and reduced the water loss in stored lettuces.

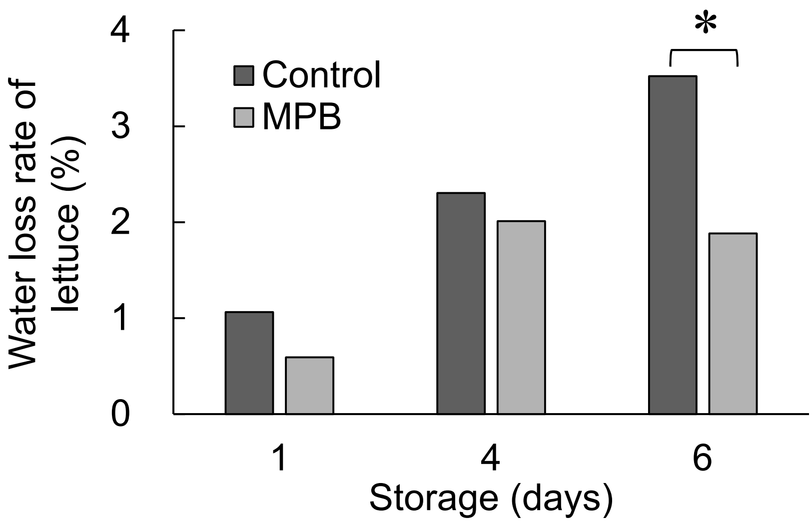


**Fig. 2** Effect of storage in different corrugated fibreboard boxes on the water loss rate (%) of lettuces. Control: non-moisture-proof corrugated fibreboard box, MPB: moisture-proof corrugated fibreboard box. Asterisk indicates a significant difference by Welch's *t*-test ($p < 0.05$; n=6). Different samples were traced for each day.

The change in SPAD value as an index of the green colour, soluble solids content, and ascorbic acid of lettuces during 6 days of storage is shown in Table 1. The loss of green colour of fresh produce is caused by chlorophyll degradation[3)] and accelerated by dark conditions[10)]. However, there were no significant differences between the SPAD values of both boxes on each day of the storage period. In addition, no relationship was observed between the SPAD values and storage period. The green colour of head lettuce is usually lighter than that of other vegetables, and hence, one of the reasons a change was not observed in its green colour was possibly because the initial content of chlorophyll might be too low for a noticeable change during storage.

**Table 1** Effects of different corrugated fibreboard boxes on the colour, soluble solids content, and ascorbic acid content of lettuce during storage

| Storage (days) | Green colour (SPAD Value) | | Soluble solids content (Brix %) | | Ascorbic acid content$^{z}$ (mg kg$^{-1}$ FW) | |
|---|---|---|---|---|---|---|
| | Control | MPB | Control | MPB | Control | MPB |
| 0 | 3.1 ± 1.4$^{y}$ | | 2.2 ± 0.4 | | 68 ± 8 | |
| 1 | 4.5 ± 1.1 | 4.8 ± 1.0 ns$^{x}$ | 2.2 ± 0.3 | 2.1 ± 0.3 ns | 69 ± 6 | 67 ± 10 |
| 4 | 2.9 ± 0.8 | 2.7 ± 1.3 ns | 2.2 ± 0.2 | 1.9 ± 0.2 ns | 65 ± 6 | 64 ± 5 |
| 6 | 2.2 ± 1.0 | 3.2 ± 1.1 ns | 2.0 ± 0.3 | 2.0 ± 0.5 ns | 52 ± 7 | 53 ± 7 |

Control and MPB: Refer to Fig. 2.

$^{z}$ L-ascorbic acid

$^{y}$ Mean ± standard deviation (n = 6).

$^{x}$No significant difference compared to control by Welch's *t*-test ($p < 0.05$).

In the present study, the soluble solids content remained stable throughout the storage period, which was consistent with the findings of similar studies involving asparagus[3),11)]. For lettuce, the soluble solids content only slightly changed during storage. The value of ascorbic acid for both boxes decreased during the storage period. This result supports that of a previous study by Lee and Kader[12)] which showed that post-harvest water loss in leafy vegetables causes rapid loss of ascorbic acid. Although there were no significant differences between both boxes on each day of the storage period, our study demonstrated that the use of MPB did not accelerate the reduction of ascorbic acid in the lettuces.

## 4. Conclusion

This study demonstrated the use of MPB to prevent water loss in lettuce during storage. In addition, our results suggest that MPB did not affect other qualities such as green colour, soluble solids content, and ascorbic acid content. Thus, we can conclude that the use of MPB would contribute to the keepability of lettuces during distribution. In further studies, we will focus on the changes in bacteria communities on lettuce during storage using MPB from the point of hygiene management.

## Acknowledgement

This work was supported by the Cabinet Office, Government of Japan, Cross-ministerial Strategic Innovation Promotion Program (SIP), and “Technologies for Smart Bio-industry and Agriculture” (funding agency: Bio-oriented Technology Research Advancement Institution, NARO).

## References

1) Ministry of Agriculture, Forestry and Fisheries, Japan, *Yasai-wo Meguru Jyousei* (The State of Vegetables) (2017), http://www.maff.go.jp/j/seisan/ryutu/yasai/yasai_jousei_0111.pdf (28th Nov. 2019)
2) Statistics Bureau of Japan, *Sakumotsu Toukei Chousa* (Statistical Survey on Crops) (2017), https://www.e-stat.go.jp/ (28th Nov. 2019)
3) B.M. Mampholo, D. Sivakumar, B. Beukes, W.J. van Rensburg, Effect of modified atmosphere packaging on the quality and bioactive compounds of Chinese cabbage (*Brasicca rapa* L. ssp. *chinensis*). J. Sci. Food Agric., **93**, p. 2008 (2013)
4) M. Shimizu, “Dynamic Transport Packaging Design Handbook for Practitioners” (Supervisor: K. Saito), Techno-System, Tokyo, Japan. p. 441 (2018)
5) P.L. Adrián, Z.V. Sonia, F. Diana, C. Alicia, C. Angel, Estimation of chlorophyll contents by correlations between SPAD-502 meter and chroma meter in butterhead lettuce, Comm. Soil Sci. Plant Anal., **38**, p. 2877 (2007)
6) H. Kitazawa, S. Motoki, T. Maeda, Y. Ishikawa, Y. Hamauzu, K. Matsushima, H. Sakai, T. Shiina, Y. Kyutoku, Effects of storage temperature on the postharvest quality of harvested in spring, J. Jpn. Soc. Hortic. Sci., **80**, p. 76 (2011)
7) S.J. Kays, R.E. Paull, “Postharvest Biology”. Exon Press, Georgia, U.S.A. p. 355 (2004)
8) P. Mahajan, G. Rux, O. Caleb, M. Linke, W. Herppich, M. Geyer, Mathematical model for transpiration rate at 100% humidity for designing modified humidity packaging. Acta Hortic., **1141**, p. 269 (2016)

9) G.G. Bovia, O.J. Caleb, M. Linke, C. Rauh, P.V. Mahajan, Transpiration and moisture evolution in packaged fresh horticultural produce and the role of integrated mathematical models: A review. Biosyst. Eng., **150**, p. 24 (2016)
10) A.M. Büchert, P.M. Civello, G.A. Martínez, Effect of hot air, UV-C, white light and modified atmosphere treatments on expression of chlorophyll degrading genes in postharvest broccoli (*Brassica oleracea* L.) florets. Scien. Hortic., **127**, p. 214
11) B. Brueckner, A. Schwarzbach, R., Correlation between sugar and saponin contents and sensory attributes of white asparagus. J. Consum. Prot. Food Safety, **5**, p. 305 (2010)
12) S.K. Lee, A.A. Kader, Preharvest and postharvest factors influencing vitamin C content of horticultural crops. Postharvest Biol. Technol., **20**, p. 207 (2000)


(Received 09 December 2019)
(Accepted 27 February 2020)

# 防湿段ボール箱はレタス貯蔵中の水分減少を効果的に抑制する

ワムブラウ ダニエル Z. K.[*]、佐藤裕子[*]、杉野直輝[*,**]、
松元咲樹[*]、北澤裕明[*,***]

レタスの流通中に品質を保持するためには、その水分減少を抑制することが重要である。本研究では、防湿加工された段ボール箱（防湿段ボール箱）と、されていない段ボール箱（対照）を用いた 6 日間の貯蔵試験によって、プラスチックフィルムで個包装されていないレタスの水分減少について調査した。またその間、その他の品質項目として、SPAD 値（緑色の指標）、可溶性固形物含量およびアスコルビン酸含量の変化についても調査した。その結果、防湿段ボール箱を用いた場合、6 日後の水分減少が有意に抑制されることが明らかとなった。また、防湿段ボール箱による貯蔵は、その他の品質項目に悪影響を及ぼすことはなかった。以上のことから、防湿段ボール箱の使用によって、レタス流通中の品質を良好に保つことができると考えられた。

**キーワード**: Brix、段ボール箱、青果物、SPAD 値、アスコルビン酸含量