



## Visible light as a new tool to maintain fresh-cut lettuce post-harvest quality



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### ARTICLE INFO

#### Keywords:

Minimally processed lettuce  
Light treatment  
Photosynthetic activity  
Browning  
Water loss

### ABSTRACT

Fresh-cut lettuces are susceptible to tissue browning and quality deterioration during post-harvest storage, even if they are kept in cold temperature. In this study we tried to counteract these undesirable physiological disorders by testing either storage under continuous light or after short treatments (2 d) with intermittent light (2 h on/2 h off) followed by storage in darkness. Two light intensities, 50 and 150  $\mu\text{mol m}^{-2} \text{s}^{-1}$ , were studied. Continuous light (50 or 150  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) significantly inhibited tissue browning but stimulated dehydration. However, intermittent light during 2 days minimized browning and water loss and showed a global positive residual physiological change during the following 5 d of storage in darkness. All light treatment maintained the photosynthetic capacity of fresh cut lettuces excepting for high continuous light (150C). The photosystem II efficiency was negatively affected by both the continuous and intermittent light at 150  $\mu\text{mol m}^{-2} \text{s}^{-1}$  but not by the moderate intermittent light (50  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ). Finally, among the overall conditions tested, the short treatment (2 d) of fresh-cut lettuce by intermittent moderate level light (50  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) followed by storage in darkness appeared to be the best compromise. Although not yet ideal, this treatment could maintained the product quality by reducing browning, minimizing weight loss and respiration and also keeping high level of photosynthetic capacity. Future studies in this context of visible light based post-harvest treatments are consequently promising.

### 1. Introduction

Fresh-cut fruit and vegetables belong to the market segment that had shown the highest economical progression in the food industry history (Martín-Belloso and Soliva-fortuny, 2011) and has continued to grow up to 5–10% per year during the past decade. Salads represent 50% of this segment and tissue browning is the most common and serious disorder easily detected and rejected by consumers.

Cut-edge browning results from an oxidation of the phenolic compounds present in the samples or neosynthesised after cutting mainly by polyphenoloxidase (PPO). The operations involved in the processing of fresh-cut lettuce such as cutting and drying cause tissue damage leading to a rapid quality deterioration and shelf-life reduction. Chemical treatments such as reducing agents or enzyme inhibitors have been studied to control the phenolic metabolism that leads to browning. However, the industry still needs a strategy to prevent browning without the use of chemical agents. Several physical methods have been proposed to extend the shelf-life such as modified atmosphere packaging (MAP) (Charles et al., 2005), heat treatments (Campos-Vargas et al., 2005; Djioua et al., 2009; Salman et al., 2008, 2007) or UV light application (Allende et al., 2006; Charles et al., 2013).

Visible light exposure represents a novel approach, environmental-friendly, that can be used to preserve the overall quality of fresh-produce. It is well known that in plants, darkness induces the expression of genes implicated in chlorophyll, protein and chloroplast degradation and an increase of the reactive species of oxygen (Wada and Ishida, 2009). However, in many cases, light is not controlled during post-harvest storage and products are commercially stored in darkness, which induces senescence and accelerates this process significantly. Light exposure can retard tissue browning of fresh-cut romaine lettuces (Zhan et al., 2012), fresh-cut celery (Zhan et al., 2013c) and delayed the yellowing of broccoli (Büchert et al., 2011).

Light has also a positive effect on nutritional quality. Continuous light (around 35  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) maintained soluble sugars and ascorbic acid in fresh-cut romaine lettuce (Zhan et al., 2013a). In spinach leaves, the endogenous pool of some vitamins (such as ascorbic acid, folate,) showed higher values when leaves were stored under visible light than in the dark (Lester et al., 2010). Light could enhance ascorbic acid synthesis due to the increase of the photosynthetic capacity that induce the availability of soluble carbohydrates, especially glucose, enabling them to contribute to the control of the ascorbic acid pool size (Toledo et al., 2003).

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Recent works studied the effect of light applied in a not continuous way. Light pulses of  $30 \mu\text{mol m}^{-2} \text{s}^{-1}$  can be used to extend post-harvest shelf-life of spinach leaves (Gergoff Grozeff et al., 2013). Daily exposure for 2 h to  $30\text{--}37 \mu\text{mol m}^{-2} \text{s}^{-1}$  of light was sufficient to delay postharvest senescence of basil leaves and suppress chlorophyll and protein loss (Costa et al., 2013). In Lamb's lettuces, intermittent low intensity light cycles increased only partially photosynthesis, while the metabolism of the green tissues is still able to provide carbon moieties for the synthesis of bioactive molecules involved in delaying senescence (Braidot et al., 2014).

However, light could also decreased the quality of fresh-cut leek (Ayala et al., 2009), asparagus (Sanz et al., 2009) and cauliflower (Olarie et al., 2009).

Thus, the influence of light on fresh-cut vegetables during storage is controversial, since both positive and negatives effects on shelf-life and quality have been observed. Furthermore, the effects of postharvest illumination on chloroplast have not been extensively investigated. Most of the time, light is applied continuously and with medium intensity (around  $30 \mu\text{mol m}^{-2} \text{s}^{-1}$ ). This study wants first to investigate the effect of two light intensities: a medium and a high. Then, the impact of light cycle treatments will be analyzed by applying continuous and intermittent light and by studying the effect on the storage of fresh-cut lettuces. The objectives of this study were to characterize the effects of light on browning and on electron transport chain in Calvin cycle in order to determine whether the photosynthetic parameters from chlorophyll *a* fluorescence and gas exchanges are permanently influenced by light in post-harvest and whether they are also good senescence markers.

## 2. Materials and methods

### 2.1. Plant materials and fresh-cut process

Butterhead lettuces (*Lactuca sativa* L.) were purchased at a local market in Avignon, France, and were immediately transported to the laboratory. The outer leaves, the core and the upper part of lettuces were removed and leaves of uniform size and color (inner leaves) were selected. The leaves were washed in chlorinated water (80 mg L, pH = 7.4) for 10 min, rinsed with tap water for 5 min and then dried on blotting paper. Four leaves were packaged in OPP film (Oriented polypropylene film, CFS, France) of  $40 \times 30$  cm and  $40 \mu\text{m}$  thickness ( $p_{\text{eO}_2} = 2.43 \cdot 10^{-16} \text{ mol m}^{-1} \text{ s}^{-1} \text{ Pa}^{-1}$ ). Three bags were used per treatment and per time.

### 2.2. Visible light treatments

Packaged lettuce leaves were stored at  $6^\circ\text{C}$  in standard climatic chambers (Sanyo MLR-351-H, Panasonic, USA) equipped with cool white fluorescent lamps (Mitsubishi OSRAM 44ss w/37), according to the following conditions (Fig. 1): 7 d of darkness (control); 7 d of continuous visible light at  $150 \mu\text{mol m}^{-2} \text{s}^{-1}$  (150C) or  $50 \mu\text{mol m}^{-2} \text{s}^{-1}$  (50C); 2 d of intermittent light (2 h on/2 h off) at either  $150 \mu\text{mol m}^{-2} \text{s}^{-1}$  (50LD) or  $50 \mu\text{mol m}^{-2} \text{s}^{-1}$  (50LD) followed by 5 d of storage in darkness. The samples were arranged on the shelves in order to receive similar light irradiance. The photosynthetic active radiation (PAR) was performed by a radiometer (PMA2100, Solar

Light).

### 2.3. Cut-edge browning, weight loss and gas content

Visual quality was scored by a four member trained panel. The quality was evaluated considering the color of the cut-edges and according to a five points hedonic scale running from 5 = good quality (absence of cut-edge browning) to 1 = severe browning. In addition, color was assessed by a colorimeter CR-400 (Konica Minolta, Japan) under standard illuminant D65. The percentage of weight loss was calculated by deducting sample weighting of each day from J0.  $\text{O}_2$  and  $\text{CO}_2$  content inside each lettuce packages were measured by a gas analyzer (Checkmate 9900, PBI Dansensor, France).

### 2.4. Net photosynthesis and chlorophyll fluorescence

Net photosynthesis ( $P_n$ ) was measured using a portable photosynthetic analyzer (LI-Cor, Lincoln, NE, USA) equipped with a leaf dedicated chamber. Measurements were done under various light intensity, 400 ppm fixed  $\text{CO}_2$ , ambient temperature and humidity. Net photosynthesis ( $P_n$ ) was deduced under light as a result of gross photosynthesis ( $P_g$ ) and respiration ( $P_n = P_g - R$ ). The LI-Cor was also used to determine the lettuce light compensation point.

To assess the behavior of photosystem II (PSII), chlorophyll *a* fluorescence (expressed in relative units) was measured with a portable Handy-PEA (Hansatech, Kings Lynn, UK). Prior to measurement, lettuce leaves were dark-adapted for 30 min. JIP-test analysis was performed by the apparatus according to the energy flux theory of the thylakoid membrane to determine several indices (Strasser and Srivastava, 1995). In this study we focalized on Fv/Fm (maximal photochemical efficiency of PSII) and Pi (Performance Index) for all conditions.

### 2.5. Statistical analysis

A total of three packaging were analyzed for each day and each condition tested and all the analyses were performed on each leaves (four leaves per bag). Thus, data represent mean values of three replicates ( $n = 12$ ). Error bars represents standard error. Statistical significance of the data was determined according to the Kruskal-Wallis test followed by Dunn's comparative post hoc method ( $p = 0.05$ ). Significant groups of data are indicated on the figures by small letters.

## 3. Results

### 3.1. Cut-edge browning

Lettuces kept in darkness showed the strongest cut-edge browning (Fig. 2A). A score of 4 (browning initiation) was obtained after 4 d and it quickly reached a score of 3 (moderate browning) under the limit of marketability, at the end of storage. Samples stored under light conditions showed a better visual quality in terms of browning. Under continuous light and whatever the intensity (150C and 50C), no browning was observed during all the storage period. In addition, there was no browning with intermittent  $150 \mu\text{mol m}^{-2} \text{s}^{-1}$  light (150 LD). Finally, a slight cut-edge browning was observed at day 7 with the 50LD treatment.



Fig. 1. Light treatments of fresh-cut lettuces.

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