International Journal of Biological, Life and Agricultural Sciences
ISSN: 2415-6612
Vol:7, No:7, 2013

The Influence of Thermic Plastic Films on Vegetative and Reproductive Growth of Iceberg Lettuce 'Dublin'

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***Abstract*—**Photoselective plastic films with thermic properties are now available so that greenhouses clad with such plastics exhibit a higher degree of "Greenhouse Effect" with a consequent increase in night time temperature. In this study, we investigate the potential benefits of a range of thermic plastic films used as greenhouse cover materials on the vegetative and reproductive growth and development of Iceberg lettuce (*Lactuca sativa* L). Transplants were grown under thermic films and destructively harvested 4, 5, and 6 weeks after transplanting. Thermic films can increase night temperatures up to 2 °C reducing the wide fluctuation in greenhouse temperature during winter compared to the standard commercial film and consequently increased the yield (leaf number, fresh weight, and dry weight) of lettuce plants. Lettuce plants grown under Clear film respond to cold stress by the accumulation of secondary products (phenolics, and flavonoids).

***Keywords*—**Photoselective plastic films, thermic films, secondary metabolites.

I. INTRODUCTION

THE demand for an all year offer of horticultural products has considerably changed the scene in modern greenhouse production. Following these developments greenhouse grower turned to more sustainable methods of producing crops of high quality and in large quantities in an all year round basis. One such alternative method is the introduction of plastic films with specific spectral properties that manipulate the environmental parameters within the greenhouse growth environment. This technology's main aim is the modification of the microclimate of the greenhouses [1]. The performance and the properties of plastic films can be improved by the incorporation of a range of additives. Anti-dust, anti-static, pigments, UV absorbance, shade, light diffusers, and thermic additives have led to the production of a number of photoselective plastic films [2]-[7]. These films can transmit, block or reflect different wavelengths of the electromagnetic spectrum offering to the growers a greater control of crop yield and quality.

One of the most exciting applications of photoselective plastic films appears to be in the area of heat control. A number of photoselective plastic films with thermic properties now exist. Thermic films allow short wave infra red from the sun to heat up the soil and the contents of the tunnel, and when the air temperature drops the hotter objects radiate the heat back again as long infra-red wave and consequently lower heat losses by radiation. Vegetable grown under thermic films have a greater vegetative development and earlier harvests of greater quality [5], [8]. To our knowledge, little research has been carried out on the impact of thermic films on vegetative growth and crop quality.

The aim of this work was to study the effect of thermic greenhouse covering films on vegetative and reproductive growth of Iceberg lettuce (*Lactuca sativa* L., cv. Dublin).

II. MATERIALS AND METHODS

A. Greenhouse Facilities and Plant Husbandry

Experiment was conducted during the winter of 2009 at the School of Plant Science Field Unit, Shinfield, Reading. The experiment was carried out in suits of six tunnels, each measuring 3m X 2.7m X 6.8m (W X H X L), specifically designed for studying experimental covering materials. Three plastic films with different thermic properties were supplied by British Polyethylene Industries Agri, Stockton-on-Tees, UK. The spectral transmission of each film was measured using scanning spectroradiometer with a double monochromator (Macam Photometrics, Livingston, UK) and these data are presented in Fig. 1.

Seeds of Iceberg 'Dublin' (Elsoms Seeds Ltd., Lincolnshire, UK) were sown in plug trays containing peat-based compost. After 4 weeks they were transplanted into 0.5 m long peat-filled growbags (Bulrush Horticulture Ltd, UK) at a density of 3 plants per growbag. Plants were irrigated with a drip irrigation system (Field Ltd, Kent, UK). Plants were supplied with tap water for the 1st two weeks. Thereafter, a standard commercial nutrient solution (NPK, 3:1:1) (Sinclair Horticultural, Lincoln, UK) was applied through a drip irrigation system (Field Ltd., Kent, UK) (four drippers per grow bag) via a Dosatron (Dosatron International, Bordeaux, France) set to provide an electric conductivity of 1.4 ms.

Temperature in each tunnel was measured using Tinytag data loggers (Gemini Data Loggers Ltd., Sussex, UK). The temperature was measured every 10min and then the daily average temperature was computed. Total radiation in each

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tunnel was measured using tube solarimeters (in-house construction, [9]), suspended 10 cm below the film. Radiation in each tunnel was measured every 30s and half-hourly average was recorded in a data-logger (DT-500, Data Electronics, Cambridge, UK).

B. Experimental Design

The experiment was set out as two blocks in which treatments (plastic films) were randomly allocated. Three films were used and replicated twice (Luminal, Lumitherm and UVI/EVA as a control). Thirty growbags in tenth were randomly allocated to three plots within each tunnel. Data were analyzed using the ANOVA procedure in Genstat statistical package (version 11) (VSN International Ltd, Oxford, UK). Differences between means were compared using least significant difference test (LSD) at 5% level.

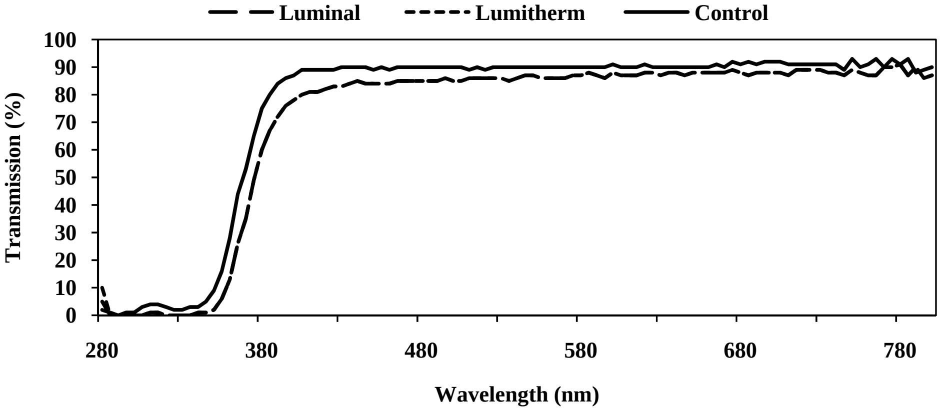


Fig. 1 Transmission spectra of each photoselective film compared with control polythene film

C. Growth and Physiological Measurements

Nine destructive plant samples after 28, 35 and 42 days of transplanting were randomly chosen from each tunnel (three plants per plot). Leaf number, fresh and dry leaf weights (g) were recorded at each destructive harvest. Dry weight measurements were carried out after drying to constant weight in a ventilated oven at 70°C. Chlorophyll fluorescence was measured on three different sunny days using a portable fluorometer (Handy PEA, Hansatech Instruments Ltd, Kings Lynn, UK). One leaf (the same age) was chosen per plant from six growbags in each tunnel. A total of 12 measurements per treatment were made. Fluorescence measurements included: Maximum quantum yield of PS II F_v/F_m was calculated as; $F_v/F_m = (F_m - F_o)/F_m$ [10]. Performance index of photosynthesis based on the equal absorption (PI_{ABS}) was calculated as reported by [11].

D. Measurement of Secondary Product

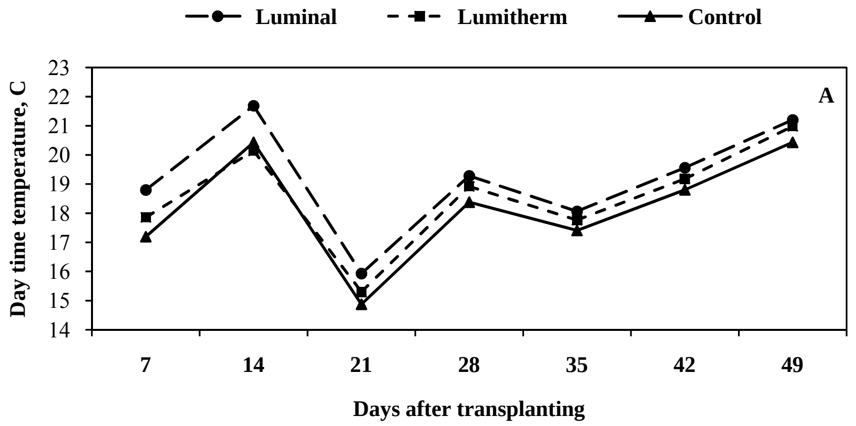
Six lettuce plants from each tunnel (2 plants / plot) were randomly chosen for the determination of phenolics and flavonoids. Five to six leaves from each plant were selected and the distal ends (4cm) were removed [12]. These leaves were blended in a food processor and a 1 g sub-sample was extracted with 10 ml of methanol: HCl (99:1, V/V) and placed in a vial. The mixture was stirred for 1-2 min with a hand held stirrer. The supernatant was then placed in a screw cap vial and placed in a cold room (2°C) overnight to facilitate extraction. An aliquot of the filtered extract was used the next day to determine total phenolics and flavonoids levels. Total soluble phenolics in the extract were determined spectrophotometrically (CECIL 1000 series, Cecil Instruments Ltd, Cambridge, UK) using the Folin-Ciocalteu colorimetric method [13], [14] which modified by [15]. Aliquots (0.125 ml) of the diluted extract (in distilled H_2O) were mixed with 0.5 ml H_2O. Then, 0.125ml of Folin-Ciocalteu reagent was added and after 6min 1.25ml of 7% aqueous sodium carbonate solution was added. Finally, water was added to adjust the final volume to 3ml and samples were allowed to stand for 90 minutes at room temperature. Results were expressed as mg of gallic acid per 100g of fresh weight. Total flavonoids content of the extracts was determined spectrophotometrically using a modified colorimetric method described by Meyers et al. [15]. Aliquots (0.25ml) of the diluted extract (in distilled H_2O) were mixed with 1.25ml H_2O followed by the addition of 0.075ml of $NaNO_2$ 5%. After 6 minutes, 0.15ml of 10% $AlCl_36H_2O$ were added and allowed to stand for 5 minutes. Then, 0.5ml of NaOH was added followed by the addition of water to adjust the final volume to 2.5ml and samples were allowed to stand for 90 minutes at room temperature. Results were expressed as mg of catechin per 100g fresh weight.

III. RESULTS AND DISCUSSION

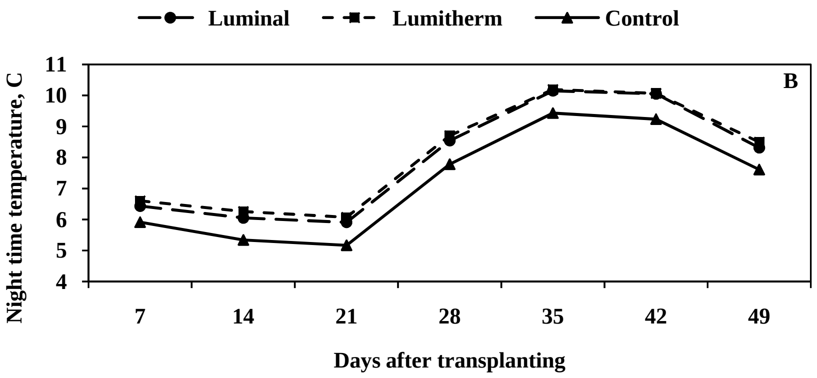
A. Greenhouse Microclimate

Light and temperature are the most two important environmental parameters that represent the climate of the greenhouse and strongly affected by the type of covering material and its properties [16]. The results of this study indicated that the environmental conditions such as the daily transmission of solar radiation and temperature have been strongly affected by the type of covering material. Of the tested photoselective plastic films, the non-thermic film

'Clear' exhibited approximately 10% higher light transmission compared to the other films with thermic properties. This result can be due to the absence of any additives in the Clear film leading to the highest light integral over the duration of the experimental period whereas, Luminal film had the lowest light integrals. (Data not shown). Temperature has been strongly affected by the type of covering material. The Luminal film appears to give constantly highest average day time temperature, while the Clear film gave the lowest temperature, and the Lumitherm film showed intermediate temperature characteristics. The average night time temperatures under the thermic films Luminal and Lumitherm, were approximately increased by 1.5C°, compared to the non-thermic film Clear (Fig. 2 (a) and (b)). This result can be attributed to the 'greenhouse effect' created under the thermic films. By the term 'greenhouse effect' is meant that solar radiation enters the greenhouse through the transparent cover, is absorbed inside the greenhouse and then it cannot escape in the form of thermal radiation because the cover is not transparent to thermal radiation [17].



(a)



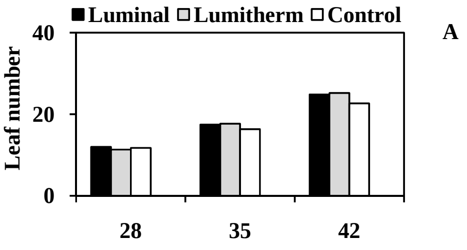
(b)

Fig. 2 Mean day time temperature (a) and night time temperature (b) for the duration of the experiment under luminal, lumitherm, and control plastic, growth and physiological parameters

The Iceberg lettuce 'Dublin' showed a significant growth response under thermic films. At 42 DAT (3rd harvesting time), total leaf number of plants grown under Luminal and Lumitherm were significantly ($P<0.001$) higher by 9% and 10% than under the Clear film, respectively (Fig. 3 (a)). At final harvesting plants grown under Luminal and Lumitherm films produced significantly ($P<0.001$) higher dry weight by 16% and 18% than plants grown under Clear film, orderly. A similar trend was observed for dry weight in both the 1st and the 2nd harvesting times (Fig. 3 (b)). Plants showed similar trends in fresh weight to those of dry weight. At the last harvesting time fresh weight yield of plants grown under Luminal and Lumitherm films was significantly ($P<0.001$) higher than those grown under the Clear one by 22% and 27%, respectively. Similar trends were observed for fresh weight in both the 1st and the 2nd harvesting times (Fig. 3 (c)). The significant effect of thermic films on leaf number, dry weight, and fresh weight of lettuce plants compared to the Clear film was in agreement with previous reported studies. For instance, [5] reported that growing vegetables under tunnels or greenhouses covered with thermic films have a greater vegetative development. Yield components including leaf number, dry weight, and fresh weight were positively correlated with the increase in the averages of night time temperatures over the experimental period. This was mainly due to the cooler night conditions under these films. This increase in yield components under the thermic films 'Luminal' and 'Lumitherm' can be linkage with the 'greenhouse effect' created under these films. This conclusion was drawn from the observed influence of the night time temperatures on all yield parameter measured at the time of last harvesting. These include the very strong correlation between night time temperature and the leaf number (R^2= 0.82), leaf dry weight (R^2=0.86), and also leaf fresh weight (R^2=0.94). Low night temperatures have been reported to reduce growth and delay the crop. Reference [18] showed an association between lettuce head weight and mean temperature in the period following crop emergence. As temperatures increased, yield of Romaine lettuce increased linearly with a similar slope. Reference [19] clarified that increasing the mean minimum temperature by 1°C increased marketable yield / plot by 0.9 head.

Temperature pattern has been reported to have an influence on the quality of the product for most vegetable species [20]. Reference [21] has shown that wide fluctuations in temperatures were detrimental to lettuce head formation. Also, [22] investigated the fluctuations of temperature effect in promoting fruit production compared to growth at constant temperature and they concluded that the stronger variations in temperature between day and night need to be avoided, because they can lead to a reinforced pushing off of the cucumber fruits. The present study indicated that thermic films effectively reduced the wide fluctuations in greenhouse temperature during the winter and consequently increased the yield of lettuce plants. However, low night temperatures do not necessarily reduce crop growth. References [23], [24] were able to grow chrysanthemum and petunia crops with equal dry weight gains at night temperatures of 5.6°C or 15.6 °C, provided day temperatures were equally in each instance.

No significant differences were observed in Fv/Fm ratio and/or PI of plants grown under Luminal and Lumitherm films compared to those grown under the Clear film (Figs. 4 (a) and (b)).



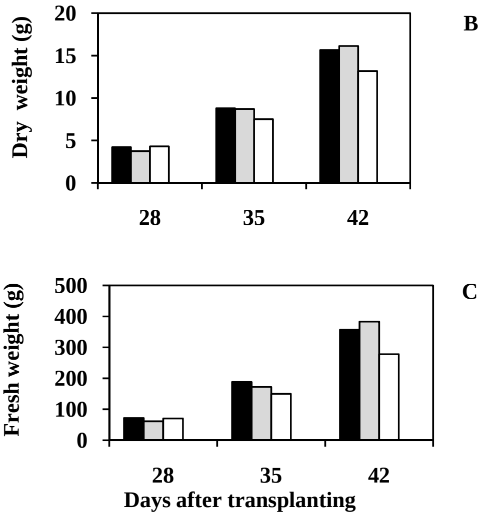


Fig. 3 Leaf number (a), dry weight (b), and fresh weight (c) of Iceberg lettuce plants cv. 'Dublin' grown under Luminal, Lumitherm, and Control plastic films. The vertical bars show LSD at 5% level

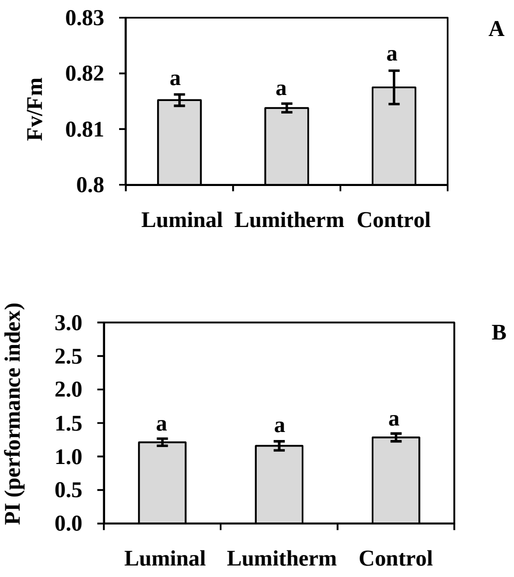


Fig. 4 Leaf fluorescence Fv/Fm (a) and performance index (PI) (b) of Iceberg lettuce plants cv. 'Dublin' grown under Luminal, Lumitherm, and Control plastic films. The vertical bars show ±S.E.M. Columns marked by different letters are significantly different at 5% level, n=12

B. Secondary Products

The Iceberg lettuce 'Dublin' used in this study showed a clear response to lower night temperatures under the non thermic film 'Clear' by increasing secondary metabolites. The total phenolic and flavonoid contents of plants grown under Clear film were the greatest compared to those grown under the thermic films; Luminal and Lumitherm. Plants under the Clear film had significantly ($P<0.05$) higher phenolic content than plants under the Luminal and Lumitherm films by 21%

and 22%, respectively (Fig. 5 (a)). Total flavonoid content followed a similar pattern to that of phenolics. Plants under Clear film had higher flavonoid content than plants under Luminal and Lumitherm films but the different was not statistically significant (Fig. 5 (b)).

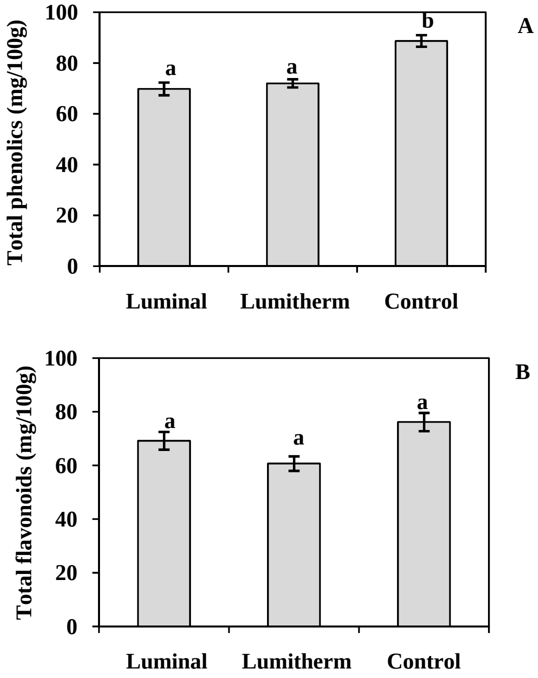


Fig. 5 Total phenolic (a) and flavonoid (b) content of Iceberg lettuce plants cv. 'Dublin' grown under Luminal, Lumitherm, and Control plastic films. The vertical bars show ±S.E.M. Columns marked by different letters are significantly different at 5% level. n = 6

The greater accumulation of phenolic and flavonoid in the presence of wide fluctuations in temperature under Clear film suggested that these products may have a protective role against low temperatures stress. Secondary metabolites have been reported to play a major role in the adaptation of plants to the changing of environment and in overcoming stress constraints [25]. The present study is in agreement with [26], who reported high levels of flavonoid compounds in the presence of low temperature stress. On the same side [7] reported an increased accumulation of phenolic compounds in lettuce occurred under high levels of UV radiation. However, it was particularly interesting to explore the growth inhibition under 'Clear' film. Reference [4] suggested that the growth inhibition in lettuce under ambient levels of UV radiation could be due to damage of photosynthetic apparatus. However, in the present study the Fv/Fm and PI were unaffected by the type of plastic used suggesting that phenolics and flavonoids had efficiently protected photosystem II. However, these compounds may have a high cost of plant protection such that the plants divert energy produced by photosynthesis to synthesis phenolic and flavonoid compounds.

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