Effect of Plastic Mulch Color on Leaf Anatomy of Lettuce (*Lactuca sativa* L.)

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**ABSTRACT**

Two lettuce (*Lactuca sativa* L.) varieties (Looseleaf and Romaine) were grown with five colored (silver, red, orange, yellow, green) plastic mulches to determine their effects on the leaf anatomical structure of lettuce. The experiment was laid in a Split Plot Randomized Completely Block Design. The result of the study showed that Romaine lettuce (8.80) had a higher number of stomata compared with Looseleaf (6.73) however, in terms of stomata length the Looseleaf had longer stomata (18.63 µm) compared with Romaine (16.42 µm). More stomata were observed in lettuce grown with red plastic mulch. However, the number of stomata did not differ among the various colored plastic mulches. Longer stomata were observed in lettuce grown with red plastic mulch, compared with lettuce grown with the other colored plastic mulches. The study showed that the color of the plastic mulch had a significant effect on the leaf anatomy of lettuce and red plastic mulch could be used to enhance the length of the stomata which may affect the growth of the lettuce.

**Key words:** Colored Plastic Mulch, Lettuce, Leaf Anatomy, Stomata.

**INTRODUCTION**

Lettuce is produced commercially in many countries worldwide and is also widely grown as a vegetable in home gardens[1]. It is especially important as a commercial crop in Asia, North and Central America, and Europe. China, U.S., Spain, Italy, India and Japan are among the world’s largest producers[2][3]. Colored mulch can affect the spectral balance and quantity of light that affects many plant developmental patterns[4] [5] [6] [7]. The spectral balance of light within the crop canopy can be influenced by the number of leaves, size of leaves and their proximity [8]. A significant greater density of stomata was observed in leaves of plants grown with full sunlight and a significant reduction of stomatal density was observed when plants were maintained in a filtered conditions[9]. Thus, a change in the microclimatic condition of the plant may also have an effect on the plants leaf anatomy. Hence, this study was conducted to determine the effects of the different colored plastic mulch on the leaf anatomy of lettuce.

**MATERIALS AND METHODS**

An experiment was conducted on January to February 2013 at the Vegetable Experimental Station in University of the Philippines Los Baños. Seeds of Looseleaf and Romaine lettuce were sown in seedling trays. One week after germination, the seedlings were pricked to ensure individual seedlings in each hole of the seedling tray. The seedlings were kept in a plastic house for three weeks. The study was laid out using the Split Plot Randomized Complete Block Design. The two lettuce varieties were the main treatments while the different colors of plastic mulch were the sub treatments. Plastic mulches were sprayed with their corresponding color (red, silver, orange, yellow, green) using a pressurized sprayer to obtain a uniform coating. One month old seedlings were transplanted late in the afternoon at a planting
distance of 45x20cm. Fifteen days after transplanting the lettuce received 5 grams of 46-0-0 per plant. 28 days after transplanting leaf samples were collected from each treatment and were subjected to leaf anatomical analysis. The leaf anatomical data (stomata density and length) were obtained using a Scanning Electron Microscope. For the scanning electron microscopy (SEM), lettuce leaves were cut and fixed in solution of 2.5% glutaraldehyde in 0.1 M cacodylate buffer pH 7.2 for 24 hours at 4°C. After fixation, the samples were rinsed in cacodylate buffer three times for 15 min, dehydrated in a series of graded ethanol from 50% to 100% alcohol (vol/vol of distilled H₂O). The samples were left for 15 min at 4°C for each concentration of alcohol. The samples were incubated for 30 minutes in a series of alcohol-isoamyl acetate mixtures with volume proportion of 50%-50% and 0-100%. Samples were then dried with CO₂ to a critical point, mounted on aluminum stubs and sputter coated with gold-palladium (JEOL JFC-1100, Japan). The cross sections of these specimens were observed on scanning electron microscope (JEOL JSM 5310, Japan) operated at an accelerating voltage of 20kV.

RESULTS AND DISCUSSIONS

Stomata Number

The two varieties varied considerably in stomata number (Table 1). Romaine lettuce had a higher number of stomata (8.8) than Looseleaf lettuce (6.73).

The variations of stomata number of the two varieties may be attributed to the genetic differences of the two varieties since every species had its own genetic performance. Furthermore, the pattern of stomata distribution is highly variable between species [10].

More stomata were observed in lettuce grown with red plastic mulch. However, it did not differ with the other colored plastic mulches. This indicates that the number of stomata was not affected by the color of the mulch. Although the level of light has been known to cause changes in stomata number [11][12]. The present study shows that the stomata number was not affected by the color of the mulch. The effect of variety and the color of the mulch interaction was also non significant.

Stomata Length

The length of stomata (Table 2) varied with the two varieties. Looseleaf lettuce had longer stomata (18.63 µm) compared with Romaine lettuce (16.42 µm). The differences could be due to genetic variation.

Lettuce grown within the red plastic mulch produced longer stomata compared with lettuce grown with the other colored plastic mulch such as silver, green, orange and yellow. This means that the color of the mulch affected the stomata length. The stomata length differences of lettuce grown with the different colored plastic mulches could be attributed to the differences in the quality of light reflected from

Table 1: Stomata number of leaf lettuce grown with different colored plastic mulch in Vegetable Crop Division experimental station

<table>
<thead>
<tr>
<th>Mulch Color</th>
<th>Loose leaf</th>
<th>Romaine</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Silver</td>
<td>8.33⁹</td>
<td>8.00⁹</td>
<td>8.16⁹</td>
</tr>
<tr>
<td>Red</td>
<td>8.00⁹</td>
<td>9.66⁹</td>
<td>8.83⁹</td>
</tr>
<tr>
<td>Orange</td>
<td>5.66⁹</td>
<td>9.66⁹</td>
<td>7.66⁹</td>
</tr>
<tr>
<td>Yellow</td>
<td>6.00⁹</td>
<td>8.66⁹</td>
<td>7.33⁹</td>
</tr>
<tr>
<td>Green</td>
<td>5.66⁹</td>
<td>8.00⁹</td>
<td>6.83⁹</td>
</tr>
<tr>
<td>Mean</td>
<td>6.73⁹</td>
<td>8.80⁹</td>
<td></td>
</tr>
</tbody>
</table>

Means in the same column or row followed by a common letter(s) are not significantly different at 5% level by LSD
the colored plastic mulch since every color would reflect a specific wavelength of light back onto the leaves of the plants. Application of colored plastic mulch could alter the spectral quality of light that is reflected from the surface of the mulch back onto the growing leaves. Different colors of the mulch create a specific environment which could have a considerable effect on plant growth and development \[13\] thus, light quality affects the stomatal apperture \[14\].

<table>
<thead>
<tr>
<th>Varieties</th>
<th>Stomatal apperture (µm)</th>
<th>Means in the same column or row followed by a common letter(s) are not significantly different at 5% level by LSD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mulch Color</td>
<td>Loose leaf</td>
<td>Romaine</td>
</tr>
<tr>
<td>Silver</td>
<td>19.55&lt;sup&gt;a&lt;/sup&gt;</td>
<td>17.15&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Red</td>
<td>22.69&lt;sup&gt;a&lt;/sup&gt;</td>
<td>21.73&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Orange</td>
<td>19.14&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.57&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Yellow</td>
<td>13.65&lt;sup&gt;a&lt;/sup&gt;</td>
<td>13.29&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Green</td>
<td>18.11&lt;sup&gt;a&lt;/sup&gt;</td>
<td>17.38&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Mean</td>
<td>18.63&lt;sup&gt;a&lt;/sup&gt;</td>
<td>16.42&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

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