ENHANCEMENT OF PHOTOSYNTHETIC RATE THROUGH PHOTOPERIOD USING LED IN CUCUMBER (Cucumis sativus)

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Abstract
Previous studies have demonstrated that photosynthetic rate in plants could be enhanced by manipulating their photoperiods. Therefore the aim of this study was to investigate the effect of day length using combination of red (R), blue (B) and yellow (Y) light provided by LED (light-emitting diode) on photosynthetic rate of cucumbers. Two cucumber cultivars (Mercy and KE-27187) were incubated under 8 h, 12 h, and 16 h photoperiod using a composition of RBY-LED light (80:10:10) and HPS + TLD lamps (as a control) inside growth chambers for 28 days. We obtained that 16 h photoperiod resulted in the best quality of plants, in terms of growth rate, sugar content, chlorophyll content, and mass balance among the treatments. However, LED-incubated plants consumed energy less efficiently compared to control plants. This indicates that precise LED specifications should be re-adjusted to maximize energy efficiency for plant production.

Keywords
Cucumber, Growth, LED, Photoperiod, Photosynthetic growth
1. Introduction

Cucumber (Cucumis sativus L.) is one of major horticultural commodities which popular as traditional medicine and food in Indonesia. Nutritional content per 100 g of cucumber consists of 12 calories, 0.6 g protein, 3.6 g carbohydrates, 0.1 g fat with omega-3 content of 5 mg and omega-6 28 mg, 45 IU Vitamin A, 0.03 g of vitamin B1, 12 mg of vitamin C, 14 mg of calcium, 15 mg of magnesium, phosphorus 24 mg, and 124 mg of potassium. However, current production of cucumber in Indonesia is not sufficient to meet market demand due to pests or disease problems and uncontrolled environmental factors of outdoor planting (Anonym, 2014). The damage caused by the contaminant and diseases can reach 30-100% each year (Prabowo, 2009). Therefore, one of the solutions to this problem is applying indoor agriculture technology or cultivation system in closed chamber that can prevent pests and disease. Moreover, this system offers more optimizing environmental factors during planting.

Light is one of the essential environmental factors that play a major role in photosynthesis and morphogenesis (Zukauskas et al., 2002) for example induction of chlorophyll synthesis (Hogewoning et al., 2010). Photosynthesis is a natural process that convert inorganic substances into organic matter in the form of carbohydrates. This is necessary due to carbohydrate is one of the component that form the building blocks of biomass and energy sources of metabolism in plants which used in synthesis and preserve biomass of plants (McKendry, 2002). Recently, new developed technology to provide great quality of light environment is by using LED (light-emitting diode). This technology is characterized by high radiant efficiency, long lifetime, cool emitting temperature, narrow emission spectra, and contain no mercury as most conventional light source do (Abidi et al., 2012).

LED usage in right amount of PAR and intensity may escalate photosynthesis and its result, as well as induce morphological changes, for example accelerating the flower induction. There are numerous studies on the effects of lighting quality using LED systems for various crops. According to Samuoliene et al, red wavelengths induced starch accumulation through photosynthesis (Samuoliene et al., 2010), while Kinoshita et al explains that blue wavelengths affected chloroplast development, chlorophyll production, and the opening of stomata (Kinoshita et al., 2001).
Cucumbers respond differently to different light. Red light can improve the capacity of photosynthesis and also increase chlorophyll content (Menard et al., 2006). On the other hand, blue light decreases the elongation growth, but increase the surface area of leaves (Novickovas et al., 2012). Therefore, LEDs lights may improve the quality of cucumber (Hao et al., 2012).

Photoperiodism may increase the production of plant biomass due to increase activity and the amount of chlorophyll per unit leaf area (Langton et al., 2003). Research conducted by Danielson (Danielson, 1944) showed the provision of light for 16 h lead to optimal amount of plant dry weight and sugar content. The more light that is absorbed in the light reaction, the more ATP and NADPH are formed, so that the sugar production in the reaction of carbon will increase. The increasing photosynthate leads to more part of the plant able to get nutrients and become food sink, for example the production of more number of fruits and greater fruit weight.

2. Materials and Methods

2.1 Materials

Materials of this study were cucumber seed, LED lamps, media, and fertilizer. Media for 66 polybag was charcoal husks (30.7 kg), cocopeat (21.5 kg), and soil (148 kg) mixed together. LED lamp composition for red, blue, and yellow light was 80:10:10. Power input for each LED is 1 watt and each treatment had 12 circuit LED, each circuit has an area of 50 x 25 cm.

2.2 Cucumber Plants Preparation and Treatment

Two cultivars of cultivars, Mercy and KE-27187 were cultivated in growth chamber with temperature 24-26 °C and air moisture content 5-6%. The initial step of preparation were pre-sowing and sowing. After 5 days, it was transferred to small pot. The next stage were transplantation and acclimatization for 2 weeks under the sun lighting. After that, plants were acclimatized under the HPS+TLD lamp with the length of exposure 16 h for 1 week. Then the plants were treated with the length of exposure under LED lamp in 8, 12, and 16 h.

Plants were watered 500 mL/day until 3 weeks after planted, then watered for 1000 mL/day. Basal fertilizer was given 10 g/plant with N: P: K (16:16:16). Second week fertilizer was Growmore N 1 g/plant, then each week, plants was given Grow more PK 1 g/plant.
2.3 Analysis of Growth Rate, Dry and Weight, and Chlorophyll Content

Plant height measurements was carried out 2 times a week. Cucumber biomass dry weight was measured on the initial conditions before the treatment (day 0) and the end of the treatment (day 28). Plant growth rate was calculated using the formula (Shuler & Kargi, 2002):

$$\mu = \frac{\ln x_t - \ln x_0}{t}$$  \hspace{1cm} (1)

The measurement of chlorophyll content were done by using SPAD meter.

2.4 Analysis of Sugar Content

Sugar content was calculated using ferricyanide reducing sugar assay (Taiz & Zeiger, 2002). Fresh, yet fully developed leaves were harvested on the last day of treatment. Leaf samples were weighed as much as 1 gram, grinded, then diluted with deion aqua, filtered with filter paper, thereby getting the supernatant subsequently dissolved by deion aqua up to 5 mL. Each sample was taken as much as 2 mL, then 1 ml alkaline cyanide solution and 1 mL potassium ferric cyanide was added. Once it was heated at a temperature of boiling water for 15 min, then add more 1 mL ferriamonium. Absorbance of the sample was measured with spectrophotometer at a wavelength of 700 nm. The amount of total sugar content in the sample was calculated by using the standard curve equation.

2.5 Statistical Analysis

Dry weight and chlorophyll content (SPAD) from both cultivars of plants were analyzed statistically using two-way ANOVA using Statistical Product Services Solution (SPSS 22) with P value = 0.05, followed by Duncan test.

3. Results and Discussion

3.1 Plant Growth Kinetics

The growth rate of cucumber Mercy and KE-27187 in 8 h, 12 h, and 16 h photoperiods were presented in (Figure 1) and (Table 1).
Figure 1: The growth curve of cucumber Mercy (a) and KE-27187 (b)

Table 1: Growth Rate Cucumber Mercy and Ke-27187

<table>
<thead>
<tr>
<th>Growth Rate (µ)</th>
<th>RBY 8</th>
<th>RBY 12</th>
<th>RBY 16</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mercy (cm/day)</td>
<td>0.0018</td>
<td>0.0040</td>
<td>0.0059</td>
</tr>
<tr>
<td>KE-27187 (cm/day)</td>
<td>0.0015</td>
<td>0.0034</td>
<td>0.0044</td>
</tr>
</tbody>
</table>

Plant exposed to 16 h produced the highest growth rate. The result was similar to Danielson study, reported that 16 h photoperiod resulted in the highest growth rate of cucumber compared to treatment with 8 and 12 h photoperiods (Danielson, 1944). Moreover, plants that get
longer exposure might receive the amount of light energy more than other plants, so that more ATP and NADPH were formed in the light reaction. Consequently, formation of sugar would increase as an energy source and a source of organic carbon for further metabolic processes in plants (Liu, Jing-fu, & Gui-bin, 2001). The more photosynthate obtained would likely to be used for growth and development.

3.2 Analysis of Total Sugar Content

Total sugar content was influenced by the process of photosynthesis in plants, and the process of photosynthesis itself was influenced by the light energy used to convert CO₂ and H₂O into glucose. Comparison of total sugar content in cucumbers with different photoperiods was presented in (Figure 2).

![Figure 2: Total sugar content in cucumber Mercy and KE-27187](image_url)

In both types of cultivars, the highest total sugar content was obtained from a plant with 16 h photoperiod, while the lowest value was obtained from a plant with 8 h photoperiod. The highest value of total sugar content in cucumber KE-27187 was 9178.6 ppm and the cucumber Mercy was 7748 ppm. This results was in accordance with the research conducted by Xu et al, the longer light exposure, the higher carbohydrates accumulated in plants (Xu et al., 2004).

Taken together, photoperiodism was very influential in the process of photosynthesis of plants that produce organic matter in the form of carbohydrates. According to Langton et al, the length of exposure would increase the production of plant biomass due to increased activity and
the amount of chlorophyll per leaf width unit (Langton et al., 2003). Increased activity and the amount of chlorophyll in plants led to increasingly high number of pigments that could absorb light energy in the form of photons. The more photons absorbed by chlorophyll, the more ATP and NADPH formed in reaction to light, so that the sugar produced in the reaction of carbon would increase. Increased activity in the reaction of carbon during a longer exposure would increase the rate of photosynthesis of plants (Sekizuka et al., 1995). Therefore, with sufficient light energy for photosynthesis, and the provision of water and good nutrition in plants, the total sugar content in plants would be higher.

3.3 Comparison of Chlorophyll Content

The amount of chlorophyll was measured in SPAD, so the amount of chlorophyll quantifiable was the total amount of chlorophyll. 12 and 16 h photoperiods resulted in the highest amount of chlorophyll in cucumber Mercy, while the highest chlorophyll content in the cucumber KE-27187 was obtained after 16 h photoperiod (Table 2).

### Table 2: Chlorophyll Content in Cucumber Mercy Dan Ke-27187

<table>
<thead>
<tr>
<th>Treatment</th>
<th>RBY 8</th>
<th>RBY 12</th>
<th>RBY 16</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mercy</td>
<td>40.727 ± 2.08&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>42.588 ± 1.70&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>42.258 ± 1.63&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td>KE-27187</td>
<td>39.391 ± 1.68&lt;sup&gt;c&lt;/sup&gt;</td>
<td>41.316 ± 1.77&lt;sup&gt;b&lt;/sup&gt;</td>
<td>42.403 ± 1.74&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Chlorophyll content value calculated from average chlorophyll content + Standard Error of Mean (SEM)

Different letter showed difference in significance based on Duncan Test (p < 0.005)

Chlorophyll synthesis is preceded by nitrate metabolic reactions in plants. Glutamate formed will produce proline, arginine and δ-aminolevulinic. Δ-aminolevulinic is an intermediate compound in the formation of chlorophyll. Longer exposure (12-16 h) would increase the activity of NADH and ATP formation in the light reaction as the more the light energy that could be captured by the plant. NADH produced played role in helping the process of reduction of
nitrate that occurred in plants, so the more NADH formed would eventually produced the higher chlorophyll in plants (Salisburry & Ross, 1995).

3.4 Mass Balance

Mass balance equation was constructed based on cucumber dry weight. Hypothetical mass balance was constructed based on the reaction of biomass formation *Atropa belladonna* root, CH$_{1.27}$O$_{0.43}$N$_{0.45}$ (Mr = 26.45 g/mol), and the reaction of photosynthesis was represented by sucrose as photosynathate. This model was used due to organ biomass composition which was closer to the composition of plant biomass than the composition of cell biomass. Input from the mass balance were sucrose (photosynthate), oxygen, and ammonium nitrate (fertilizer). It was assumed that the source of ammonium nitrate only derived from fertilizer, which was also the limiting reaction in this equation. Hypothetical mass balance equation of cucumber plant was presented in Table 3.

$$0.39 \text{C}_{12}\text{H}_{22}\text{O}_{11} + 0.23 \text{NH}_4\text{NO}_3 + 3.43 \text{O}_2 \rightarrow \text{CH}_{1.27}\text{O}_{0.43}\text{N}_{0.45} + 4.07 \text{H}_2\text{O} + 3.64 \text{CO}_2$$

<table>
<thead>
<tr>
<th>Initial (mol)</th>
<th>C$<em>{12}$H$</em>{22}$O$_{11}$</th>
<th>NH$_4$NO$_3$</th>
<th>O$_2$</th>
<th>CH$<em>{1.27}$O$</em>{0.43}$N$_{0.45}$</th>
<th>H$_2$O</th>
<th>CO$_2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reaction (mol)</td>
<td>0.93</td>
<td>0.55</td>
<td>8.20</td>
<td>2.39</td>
<td>9.73</td>
<td>8.70</td>
</tr>
<tr>
<td>Final (mol)</td>
<td>-</td>
<td>0</td>
<td>-</td>
<td>2.39</td>
<td>9.73</td>
<td>8.70</td>
</tr>
</tbody>
</table>

Fertilizer addition of 44 gr (equivalent to 0.55 mol ammonium nitrate) obtained the final biomass of 2.391 mol, or equivalent to 63.25 g. However, actual dry weight of plants was lower than dry weight of hypothetical. This could be affected by other factors that were not included in the calculation of the mass balance, i.e. water evapotranspiration, carbon dioxide, and nutrients to the soil. The light exposure that comes closest to the weight of hypothetical was 16 h (Table 4). According to Sekizuka et al, in the optimal temperature conditions for growth, extension of light exposure would increase the plant dry weight, because longer exposure time can increase the light energy absorbed by plants, so that more photosynathate produced for cell division and differentiation, thus increase dry weight (Sekizuka et al., 1995).
Table 4: Actual Dry Weight Cucumber Mercy and Ke-27187

<table>
<thead>
<tr>
<th>Treatment</th>
<th>RBY 8</th>
<th>RBY 12</th>
<th>RBY 16</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mercy (g)</td>
<td>3.00 ± 0.27&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.68 ± 1.08&lt;sup&gt;b&lt;/sup&gt;</td>
<td>15.64 ± 0.11&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>KE-27187 (g)</td>
<td>2.22 ± 0.04&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.27 ± 1.34&lt;sup&gt;b&lt;/sup&gt;</td>
<td>12.42 ± 1.51&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Dry weight value calculated from average dry weight + Standard Error of Mean (SEM)
Different letter showed difference in significance based on Duncan Test (p < 0.005)

3.5 Energy Efficiency

In the LED light treatment, the energy input derived from the 12 LED circuit, respectively 42 watt (1 watt x 42 lamp) during exposure of 8, 12 and 16 h in a day, for 28 days of treatment. Exposure of light for 8 h resulted in energy of ± 406 MJ, for 12 h at ± 610 MJ, and for 16 h at ± 813 MJ. It was assumed that plants could absorbed the energy efficiency of the lamp by 5%, so that the energy input accepted by the plant for the exposure of 8 h was ± 20 MJ, for 12 h at ± 30 MJ, and for 16 h at ± 41 MJ. Measurements using a quantometer showed the value of PAR received by plants from the use of LED lights was ± 44 µmol/m2s. On the measurement of the intensity of the LED lights indicated the value of ± 807 lux.

In the control plants (treated with 2 HPS lamps and 12 TL lamps), the energy input of HPS lights was 400 watts and TLD was 36 watts. Energy received by the plant for 28 days of treatment for 16 h exposure was equal to ± 99 MJ. PAR value received by the plant with the use of HPS lamps + TLD was ± 120 µmol/m2s, and the intensity of the lamp had a value of ± 3787 lux.

Result showed that growth kinetics parameter total sugar content, the amount of chlorophyll and dry weight of cucumber in control was higher than those treated with LED lights. This suggested that lamp energy, PAR value, and the intensity of the LED light given for treatment in this study was not sufficient for the cucumber to perform well and produced maximum growth and field. According to Torres and Lopez, cucumber need PAR approximately 350 µmol/m²s, while PAR accepted by each cucumber with LED treatment in this trial is only ± 44 µmol/m²s. Therefore, LED lights used in this experiment was only able to meet the needs of PAR of cucumber about 12%, so that the result of growth was not optimal. Thus, to produce
maximum plant growth and photosynthesis, plant should be given appropriate quantity and quality of light so that the energy requirements, PAR and the intensity received by the plants were adequate to their needs.

4. Conclusion and Suggestion

Photoperiodism influenced the rate of photosynthesis of cucumber. Giving the length of exposure for 16 h was better than 8 and 12 h, because it would increase the rate of photosynthesis with higher growth rate, dry weight, amount of chlorophyll and total maximum sugar content in cucumber Mercy and KE-27187. Plants that get longer exposure might receive the amount of light energy more than other plants, so that more ATP and NADPH were formed in the light reaction, led to higher sugar accumulation as an energy source and a source of organic carbon for further metabolic processes in plants. The maximum value of the total sugar content in cucumber Mercy was 7748.1 ppm and cucumber KE-27187 was 9178.6 ppm.

LED was designed for better plant growth because it has a high light efficiency and low energy usage. However, this study used the LED lights with lower energy efficiency. Therefore, further research need better LED lights that can meet the energy needs of the plant, for example PAR and intensity that can be received by the plant to grow and maintain metabolic process optimally.

5. Acknowledgment

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