Real-time Monitoring of Photosynthesis and Transpiration of a Fully-grown Tomato Plant in Greenhouse

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A real-time photosynthesis and transpiration monitoring system was developed and applied to monitor the time course of photosynthesis and transpiration of fully-grown tomato plant in a semi-commercial greenhouse. The system is composed of an open-bottom chamber and a sensing unit. The chamber encloses a two-meter high tomato plant with transparent film. The two fans equipped at the top of the chamber exhaust interior air and the open bottom allows exterior air to move into the chamber. The inflow- and outflow-air were continuously sampled, and their CO2 and H2O concentrations were measured. The developed system successfully traced the time courses of net photosynthetic and transpiration rates on typical sunny and rainy days. A decrease in net photosynthetic rate, which might be due to the stomatal closure caused by drought stress, was observed on the sunny day. On the other hand, negative net photosynthetic rate and transient increase in net photosynthetic rate and transpiration rate caused by the supplemental lighting were observed on the rainy day. In addition, the dark respiration rate was recognized during nighttime. These results suggest that the developed system can be a useful tool to evaluate the dynamic changes in photosynthesis and transpiration of crops in greenhouse.

Keywords : drought stress, photosynthesis, supplemental lighting, transpiration

INTRODUCTION

Maximization of crop net photosynthesis is one of the most important objectives of environmental control in greenhouses (Takayama, 2013). To increase the crop photosynthesis, instrumentations such CO2 enrichment, supplemental lighting and so on have been installed in commercial greenhouses (Vanthoor et al., 2011). To establish the appropriate environmental control in greenhouse, plant diagnosis techniques are important and the Speaking Plant Approach (SPA) is regarded as a sophisticated concept (Udink ten Cate et al., 1978; Hashimoto, 1980). In the plant diagnosis techniques, measurement of photosynthetic rate is important to monitor the plant physiological status and performance of assimilation. Nevertheless, many open gas-exchange systems have been designed to measure leaf photosynthetic rate (Dutton et al., 1988) and there is no appropriate instrumentation to monitor fully-grown/full-size crop photosynthetic rate in commercial greenhouse.

Many studies estimated the crop photosynthesis by using the previously measured photosynthesis light response curve at single leaf level, incoming radiation, canopy light profile and the leaf area index (Spitters et al., 1989; Jones, 1992; Cannell and Thornley, 1998; Li et al., 2014). However, the environmental response of photosynthesis at single leaf level does not represent the crop photosynthesis (Dutton et al., 1988). Especially, Paradiso et al. (2011) reported that the spectral dependence of light absorption and photosynthesis at the canopy level is different from that at leaf level. Furthermore, canopy architecture of tomato plant has a large impact on crop light distribution and photosynthesis (Sarlikioti et al., 2011). On the other hand, Zekki et al. (1999) and Teitel et al. (2011) proposed a monitoring of CO2 balance at a greenhouse level to measure the net photosynthetic rate of all the plants grown in the greenhouse. However, it is difficult to evaluate the fluctuated ventilation rate of the greenhouse at high time resolution, so these techniques provide low time resolution data. Therefore, a real-time monitoring of photosynthesis of a full-size plant grown in greenhouse has been required for SPA. There are several whole plant level open chambers for trees or herbaceous plants (Munakata, 1970; Miller et al., 1996; Ferrai et al., 2016), however they are...
not able to be applied for the plants in greenhouse because of their massive and complex assembly.

The main objective of this study was real-time monitoring of photosynthesis and transpiration of a full-grown tomato plant in greenhouse. To achieve this objective, we developed an open chamber method-based photosynthesis and transpiration monitoring system for whole tomato plant. Then, we applied the developed system to monitor the daily change in photosynthesis and transpiration of tomato plant grown in greenhouse under several conditions including supplemental lighting.

MATERIALS AND METHODS

Plant materials and instrumentation of the experimental greenhouse

Tomato (*Solanum lycopersicum* Mill. ‘Taiankitijitsu’) plants were transplanted to rockwool cubes (Grodan Delta, GRODAN Group, Roermond, The Netherlands, 100 mm [W]×100 mm [H]×65 mm [D]) after germination, and then the rockwool cubes were set onto rockwool slabs (Grotop Expert, GRODAN Group, Roermond, The Netherlands, 1.0 m [W]×0.2 m [H]×75 mm [D]) on 3rd October 2011 for Experiment 1-2 and on 20th August 2013 for Experiment 3 in a semi-commercial greenhouse (23 m [N-S]×44.8 m [E-W]; the details of Experiment 1-3 are described below) of Ehime University (33°50’ N, 132°47’ E). Four rockwool cubes were set on a rockwool slab at an interval of 0.25 m and each cube was supplied with nutrient water (A-type recipe of Otsuka House Solution, Otsuka Chemical Co., Ltd., Japan) through a dripper. Each plant was attached to a nylon tie for support, which in turn was connected to a steel wire located above the canopy. Professional growers managed the tomato plants and routine crop maintenance operations were done once a week. A full-grown 8-10-month-old tomato plant, which had a 7.9 m length stem and 13-19 leaves within 1.6 m from the shoot apex, was used for the experiments. In the greenhouse, nine 360 W high-pressure sodium lamps (NH360FLS, IWASAKI ELECTRIC Co., Ltd., Tokyo, Japan) are installed for supplemental lighting. These lamps were fixed at 1.4 m above the tomato canopy at an interval of 6 m along the north-south axis and 3 m along the east-west axis. The light intensity of the supplemental lighting at the top of the tomato canopy was at photosynthetic photon flux density (PPFD) of 75 μmol m⁻² s⁻¹. The experimental plant was located in the center of the supplemental lighting area. The CO₂ enrichment was accidentally done by turning on the heater (CG-205SL, NEPON Inc., Tokyo, Japan) equipped in the greenhouse.

The open-bottom chamber

Figure 1 shows a schematic diagram of the open-bottom chamber developed in this study. The size of the chamber was 0.35 m [W]×0.7 m [D]×2.1 m [H]. The bottom was opened to the greenhouse-air allowing the greenhouse-air flow into the chamber. The chamber was consisted of a transparent film (Bejitaron super kirinashi AAA, SEKISUI FILM Co., Ltd., Osaka, Japan) supported by thin and lightweight steel frames. So, weight of the chamber is lower than 5 kg and installation is very easy. The transmittance of the film was over 90% in PPFD range. The chamber encloses a full-grown tomato plant. Two ventilation fans (MB630-B, ORIENTAL MOTOR Co., Ltd., Tokyo, Japan) were equipped at the top of the chamber to exhaust the air inside the chamber, which made a stable upward airflow in the chamber. The airflow rate (F) of the ventilation fan was 0.36 m³ min⁻¹ for each, 0.72 m³ min⁻¹ in total and the estimated ventilation rate of the chamber was 84.0 times h⁻¹.

The CO₂ and H₂O concentrations were measured with a portable photosynthesis system (LI-6400; LI-COR, Lincoln, USA). Two sets of CO₂ and H₂O sensors of the LI-6400 were used at the same time. By using the air pump of the LI-6400, the inflow air coming into the chamber (greenhouse air) was sampled and exposed to one set of CO₂ and H₂O sensors. An air pump, EAP-01 (AS ONE, Osaka, Japan) with a flow rate of 1000 ml min⁻¹, was used for air sampling of the outflow air from the chamber and exposed to the another set of CO₂ and H₂O sensors.
CALCULATION OF NET PHOTOSYNTHETIC RATE, TRANSPIRATION RATE AND TOTAL CONDUCTANCE

Net photosynthetic rate \( (P_n) \) [\( \text{mmol} \text{ plant}^{-1} \text{ s}^{-1} \)] and transpiration rate \( (T) \) [\( \text{mmol} \text{ plant}^{-1} \text{ s}^{-1} \)] were calculated by the following equations:

\[
P_n = F \left( \frac{[\text{CO}_2]_{in} - [\text{CO}_2]_{out}}{N_{pist}} \right)
\]

\[
T = F \left( \frac{[\text{H}_2\text{O}]_{in} - [\text{H}_2\text{O}]_{out}}{N_{pist}} \right)
\]

where \( F \) is the airflow rate [\( \text{mol s}^{-1} \)] of the ventilation fan, \([\text{CO}_2]_{in} \) is the \( \text{CO}_2 \) concentration of inflow air [\( \text{mmol mol}^{-1} \)], \([\text{CO}_2]_{out} \) is the \( \text{CO}_2 \) concentration of outflow air [\( \text{mmol mol}^{-1} \)], \([\text{H}_2\text{O}]_{in} \) is the \( \text{H}_2\text{O} \) concentration of inflow air [\( \text{mmol mol}^{-1} \)], \([\text{H}_2\text{O}]_{out} \) is the \( \text{H}_2\text{O} \) concentration of outflow air [\( \text{mmol mol}^{-1} \)], and \( N_{pist} \) is the plant number inside the chamber. The effect of the transpiration on the air volume can be regarded as negligible because of it is less than 1%.

The calculation of total conductance \( (g_t) \) [\( \text{mmol plant}^{-1} \text{ s}^{-1} \)] is based on the assumption that the leaf temperature equals the air temperature. So, the \( g_t \) is calculated by the following equations:

\[
g_t = T \left( \frac{\text{VPD}}{P} \right)
\]

where \( \text{VPD} \) is vapor pressure [\( \text{mmol mol}^{-1} \)] deficit [\( \text{kPa} \)], \( P \) is atmospheric pressure [\( \text{kPa} \)]. The \( g_t \) consists of stomatal conductance and boundary layer conductance. In the chamber, the boundary layer conductance can be assumed constant because of the stable airflow in the chamber. Hence, the dynamic changes in \( g_t \) can be regarded as the changes in stomatal conductance.

**Experiments**

Three experiments, Experiment 1−3, were conducted to monitor the change in \( P_n \), \( T \), and \( g_t \) of tomato plant grown in greenhouse under the several conditions. Experiment 1 was carried out on typical sunny day and Experiment 2 was done on a typical rainy day and evaluated the effects of supplemental lighting on crop photosynthesis and transpiration. Experiment 3 was conducted during nighttime with supplemental lighting and measured dark respiration rate of full-size tomato plant in greenhouse. Experiment 1 and 2 used same tomato plant and Experiment 3 used other tomato plant. Data acquisition rate was 2 minutes and calculated average value for 6 minutes. Light intensity was measured by a line PPFD sensor (SEN-301/S/S, Apogee Instruments Inc.) attached above the chamber.

**Experiment 1 - Measurement on a typical sunny day**

On a typical sunny day, we monitored the change in \( P_n \), \( T \), and \( g_t \) of tomato plant grown in greenhouse with the developed system. The measurement was done from 7:00 to 15:00 on 12th June 2012. The supplemental lighting was turned on for 30 minutes at an interval of 30 minutes during the measurement.

**Experiment 2 - Measurement on a typical rainy day with supplemental lighting**

On a typical rainy day, we monitored the change in \( P_n \), \( T \), and \( g_t \) of tomato plant grown in greenhouse with the developed system. The measurement was done from 20:00 on 1st June 2014 to 4:00 on 2nd June 2014. The supplemental lighting was turned on for about 4 hours from 22:00 on 1st June 2014 to 2:00 on 2nd June 2014.

**RESULTS AND DISCUSSION**

**Experiment 1 - Measurement on a typical sunny day**

Figure 2 shows the time course of PPFD, air temperature, relative humidity and \( \text{VPD} \) measured on typical sunny day (13th June 2012). The PPFD was up to 1200 \( \text{mmol} \text{ m}^{-2} \text{ s}^{-1} \) before 12:00 and maintained 1200 ± 300 \( \text{mmol} \text{ m}^{-2} \text{ s}^{-1} \) until 14:00. Accompanied by the increase in solar radiation, the air temperature increased and relative humidity decreased. The \( \text{VPD} \) showed continuous increase and reached the maximum value of 7 [kPa] at 14:00. Figure 3 shows the changes in \([\text{CO}_2]_t \) (A) and \([\text{H}_2\text{O}]_t \) (B). The \([\text{CO}_2]_t \) was in the range of 350–420 [\( \text{mmol mol}^{-1} \)] and \([\text{CO}_2]_t \) kept lower value compared with \([\text{CO}_2]_m \) during the measurement because of the photosynthesis. The \([\text{H}_2\text{O}]_t \) was in the range of 20–32 [\( \text{mmol mol}^{-1} \)] and \([\text{H}_2\text{O}]_t \) kept higher value compared with \([\text{H}_2\text{O}]_m \) during the measurement because of the transpiration.

Figure 4 shows the time courses of \( P_n \) (A), \( T \) (B) and \( g_t \) (C) of the tomato plant inside the chamber. The \( P_n \) reached the maximum value of 13 [\( \text{mmol plant}^{-1} \text{ s}^{-1} \)] at 9:00 and then decreased slightly and kept at 6 ± 2 [\( \text{mmol plant}^{-1} \text{ s}^{-1} \)] from 10:30 to 14:30 even though the PPFD from 10:30 to 14:30 was higher than that around 9:00 (Fig. 2A). The \( T \) showed prompt increase at 8:00 and then maintained a stable value of 1.6 ± 0.6 mmol plant$^{-1}$s$^{-1}$ until 14:30 even though the \( \text{VPD} \) showed continuous increase until 14:00 (Fig. 2B). On the other hand, the \( g_t \) increased quickly from 9:00 to 15:00 on 12th June 2012. The supplemental lighting was turned on for 30 minutes at an interval of 30 minutes during the measurement.
8:00 to 9:00 and reached the maximum value at just after 9:00 and then showed dramatical decrease from 9:00 to 11:00 and kept lower value than 50 mmol plant\(^{-1}\) s\(^{-1}\) until the end of the measurement. The decline in \(g_t\) during 9:00 to 11:00 implied stomatal closure, which might be caused by drought stress induced by the rapid increase in VPD during 9:00 to 10:00 (Fig. 2B), and moreover, this can be the main reason of the slight decrease in net photosynthesis rate at that time (Fig. 4A).

**Experiment 2 - Measurement on a typical rainy day with supplemental lighting**

Figure 5 shows the time course of PPFD, air temperature, relative humidity, VPD measured on typical rainy day (12th June 2012). The PPFD was less than 300 \(\mu\text{mol m}^{-2}\text{s}^{-1}\) and the supplemental lighting at an interval of 30 minutes (Time zone in gray in Fig. 5) provided additional light at PPFD of 75 \(\mu\text{mol m}^{-2}\text{s}^{-1}\). Air temperature, relative humidity and VPD were almost stable during the measurement. Figure 6 shows the time course of \([\text{CO}_2] (A)\) and \([\text{H}_2\text{O}] (B)\). The \(\text{CO}_2\) concentrations \([\text{CO}_2\text{in}]\) and \([\text{CO}_2\text{out}]\) showed a prompt increase at around 14:00 because of the \(\text{CO}_2\) enrichment. Although \([\text{CO}_2\text{in}]\) sometimes showed higher values compared with \([\text{CO}_2\text{out}]\), \([\text{H}_2\text{O}\text{out}]\) kept higher value compared with \([\text{H}_2\text{O}\text{in}]\) during the measurement.

Figure 7 shows the time course of \(P_n\) (A), \(T\) (B) and \(g_t\) (C) of the tomato plant inside the chamber. The \(P_n\) showed negative values from 9:00 to 11:30 because of the quite low light intensity (Fig. 5A). However, supplemental light-
Figure 7 Time course of photosynthetic rate (A), transpiration rate (B), total conductance (C) on a typical rainy day (12th June 2012).

Figure 8 Time course of PPFD and air temperature (A), relative humidity and vapor pressure deficit (B) at night (1st - 2nd June 2014). Gray period shows a time that supplemental lighting was turned on.

Figure 9 Time course of CO₂ concentration (A), H₂O concentration (B) at night (1st - 2nd June 2014).

REAL-TIME MONITORING

The supplemental lighting was turned on.

Experiment 3 - Measurement at night with supplemental lighting

Figure 8 shows the time course of PPFD, air temperature, relative humidity, VPD measured during nighttime (from 1st June 2014 to 2nd June 2014). The PPFD was 75 µmol m⁻² s⁻¹ during the time period that supplemental lighting was turned on (Time zone in gray in Fig. 8). Air temperature gradually decreased from 29 °C to 26 °C until 4:00. According to the air temperature, the relative humidity was slightly increased and the VPD was gradually decreased. Figure 9 shows the time course of [CO₂] (A) and [H₂O] (B). [CO₂]ᵢₑ was maintained high value at 1.5 and slightly increased from 11:30 to 13:30. Similar to the time course of [CO₂]ᵢₑ, supplemental lighting increased T and gₛ during 9:00-11:00. Furthermore, the gₛ showed almost similar time course that of T. These results prove that supplemental lighting at PPFD of 75 µmol m⁻² s⁻¹ certainly drives crop photosynthesis and induces stomatal opening, which results in the increase in transpiration rate, at least during the 9:00-11:00 time period.

Figure 10 shows the time course of Pₛ (A), T (B) and gₛ (C) of the tomato plant inside the chamber. The Pₛ kept negative values at −1.5 ± 0.5 µmol plant⁻¹ s⁻¹ under dark condition (20:00-22:00 and 2:30-4:00, time zone in white in Fig. 10), which must be caused by the dark respiration of the plant inside the chamber. During the time period of supplemental lighting (time zone in gray in Fig. 10), the T and gₛ were kept at almost constant values throughout the night regardless of the presence or absence of supplemental lighting (Fig. 10B and C). These responses of T and gₛ to supplemental lighting during nighttime were significantly different from those observed during nighttime (9:00-11:00 in Fig. 7B and C). These results suggested that the supplemental lighting during nighttime drives crop photosynthesis but does not effect on stomatal opening and transpiration. This phenomenon might be attributed to the lack of blue light, which is a key for stomatal opening and is not included in the supplemental lighting, during nighttime (Assmann and Shimazaki, 1999).
In this study, we developed a real-time photosynthesis and transpiration monitoring system and applied it to monitor the time course of photosynthesis and transpiration of fully-grown tomato plant in a semi-commercial greenhouse under three different environmental conditions such as daytime of sunny day, daytime of rainy day, and nighttime of fine day. The developed system successfully traced the time courses of net photosynthetic rate, transpiration rate and total conductance of the tomato plant inside the monitoring system. On the sunny day, a stomatal closure, which might be caused by drought stress, and relating net photosynthesis decrease were recognized. On the rainy day, supplemental lighting induced increases in net photosynthesis and transpiration and stomatal opening was observed. During night-time, dark respiration rate was quantified, and supplemental lighting induced increase in net photosynthesis was observed without any changes in transpiration and stomatal opening. These results imply that the developed system can be a useful tool to evaluate the crop photosynthesis/respiration, transpiration and stomatal behaviour under greenhouse conditions. For the next step, we have to compare the developed system with other measurement methods to validate the reliability of this system.

**CONCLUSION**

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