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Observation system for the control of the hive environment by the honeybee (*Apis mellifera*)

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The honeybee can control its hive environment to survive drastic changes in the field environment. To study the control of multiple environmental factors by honeybees, in this experiment, we developed a continual and simultaneous monitoring system for the temperature, moisture, and carbon dioxide (CO_2) concentration in a honeybee hive. Changes in hive weight, CO_2 production rate, and honeybee behavior were also monitored to estimate energy costs and behavioral activity for the environmental regulation. Measurements were conducted in August 2008. We found that the honeybee hive has a microclimate different from the ambient climate, and that the difference was partly accompanied by changes in honeybee activity. Our results also suggest that hive temperature, humidity, and CO_2 concentrations are controlled by different mechanisms. Additional monitoring of the hive environment and honeybee behavior for longer periods would enable us to understand the mechanisms of environmental control by honeybees, which is one of the behaviors that define honeybees as social insects.

Honeybees are known to control their hive environment to survive drastic changes in the field environment (Jones & Oldroyd, 2007). Thermoregulation of the hive is well known as the key to the ecological success of social insects (Wilson, 1990). Characteristics and mechanisms of thermoregulation have been intensively studied, and honeybees have been shown to maintain their brood nest temperature within a fairly narrow range—between about 33ºC and 36ºC—over a wide range of outside temperatures (Jones, Myerscough, Graham, & Oldroyd, 2004; Jones & Oldroyd, 2007). Kronenberg and Heller (1982) found that workers regulate temperature by fanning hot air out of the nest when the temperature rises above 38ºC. On the other hand, metabolic heat production increases when the temperature is below 30ºC, and honeybees begin clustering tightly at 15ºC. Control of the hive humidity and carbon dioxide (CO_2) concentration has also been suggested (Simpson, 1961), but there is less information about this than for thermoregulation. Seeley (1974) reported that fanning behavior was observed at high CO_2 concentrations inside the hive and the concentrations were more precisely controlled in large colonies than in small colonies. Fanning behavior at high humidity inside the hive was also observed (Human, Nicolson, & Dietemann, 2006), but the mechanisms are poorly understood.

In spite of many studies on the environmental control of honeybee hives, little is known about how honeybees integrate the controls of multiple factors such as temperature, CO_2 concentration, and humidity to stabilize the hive environment. There is little information on how environmental factors change differently in a hive compared with ambient environmental factors at the same time. It is also unclear how the hive environment is established actively by honeybees and passively by the hive location, orientation, material, and architecture. Human et al. (2006) suggested that there might be trade-offs in controlling multiple environmental factors. Since the metabolic heating of a honeybee normally accompanies changes in the respiratory emission of CO_2 and the evaporation of hive water and nectar, the stability of the nest's CO_2 environment and humidity could be disturbed. Fanning behavior has been shown to be important not only for cooling the hive (Kronenberg & Heller, 1982) but also for ventilation (Seeley, 1974). However, it has not been determined whether fanning for these purposes is performed separately. There might be optimal and suboptimal limits for each environmental factor, and a honeybee might control an environmental factor within the suboptimal limits of the other factors.

Continuously measuring multiple factors of the hive environment simultaneously with honeybee behavior and metabolic heating might reveal the mechanism of the integrated regulation of multiple environmental factors by honeybees. In this study, we developed a continual and si-

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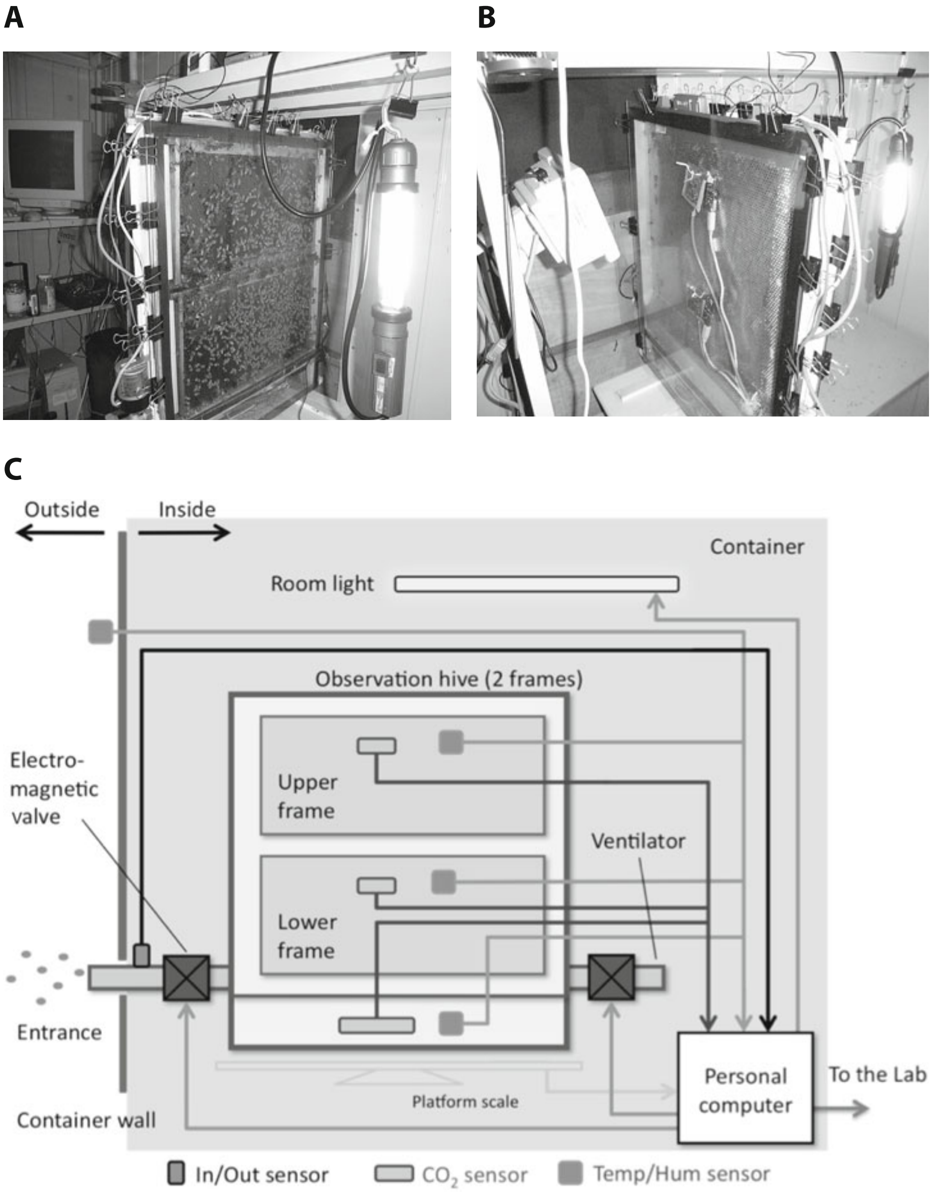

Figure 1. Measurement system for environmental regulation in the honeybee hive. (A) The bee room of the box. Two frames were located in the observation hive. Behaviors in the hive were observed through the glass plate. (B) The sensor room of the observation hive. Temperature/humidity sensors and CO_2 sensors were located behind the hive frames. (C) Temperature, humidity, and CO_2 concentration in the hive were measured and recorded automatically. Hive weight, CO_2 production, and coming/going conditions were also recorded on a personal computer.

multaneous monitoring system for temperature, moisture, and CO_2 concentration in a honeybee hive. Changes in the hive weight, CO_2 production rate, and honeybee behavior were also monitored to estimate energy costs and behavioral activity for the environmental regulation.

METHOD

A colony of European honeybees (*Apis mellifera*) of all ages, and a queen, were reared in a custom-made two-frame observation hive consisting of a glass box, with a wooden frame, measuring 660 mm long, 560 mm wide, and 72 mm deep. The hive temperature, humidity, and CO_2 concentration were monitored automatically (see Figure 1). The inside of the box was divided vertically into two rooms by a porous metal plate. Honeybees were in one room and the temperature, humidity, and CO_2 sensors were in the other room. Honeybees were prevented from entering the sensor room by the metal plate. One entrance hole and one ventilator hole, each 1 cm in diameter, were made in the lower part of the box. The opening and closing of the two holes were controlled by electronic magnetic valves to measure the CO_2 production of the hive.

Hive temperature, humidity, and CO_2 concentration were monitored every 5 min at a point in each of the upper, lower, and bottom

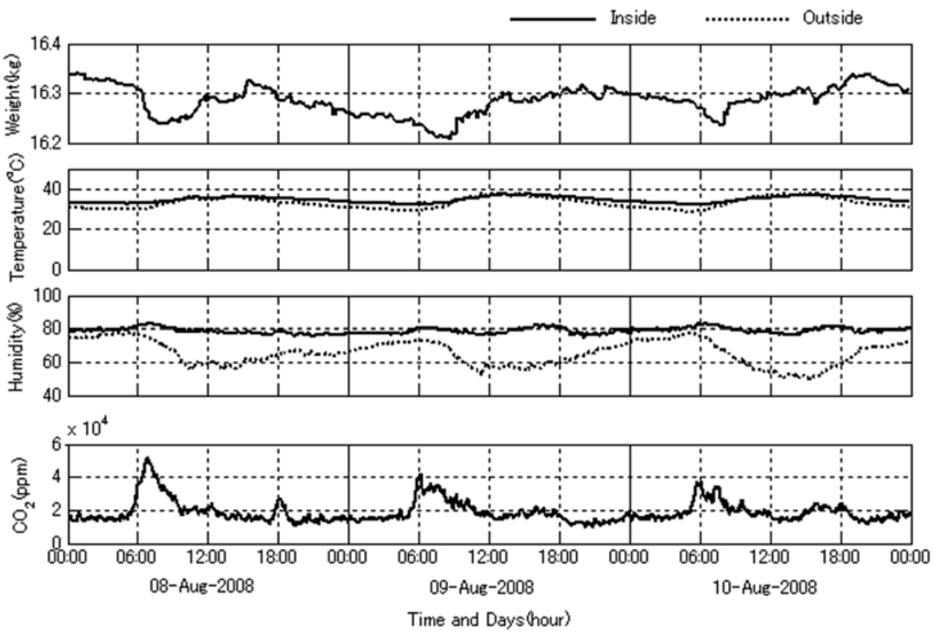

Figure 2. Temporal changes in weight, temperature, humidity, and CO_2 concentration measured in the middle part of the honeybee hive and in the ambient, August 8–10, 2008. The daily cycle of the hive weight was an increase due to foraging in the day and a decrease due to consumption at night. The temperature in the hive changed synchronously with the ambient temperature, but the humidity in the hive was kept constant. A significant increase in the CO_2 concentration was observed around 6 a.m. each day. Another CO_2 peak was observed in early evening.

parts of the glass box, using bandgap and polymer sensors (SHT-11, Sensirion, Staefa, Switzerland). The CO_2 concentration was measured every 10 sec at the same locations in the hive using nondispersion infrared radiation analyzers (GMT220, GMD20, Vaisala, Vantaa, Finland, and K30-10, SenseAir, Delsbo, Sweden). The outside air temperature and humidity around 1 m above the ground were also monitored 1 m from the hive, using the same sensors. The box was placed on a platform scale (FG-60KAL and FG-30KAM, A&D, Tokyo, Japan) to measure the hive weight continuously. The increasing rate (ppm/sec) of the CO_2 concentration in the hive was measured every 1–2 h as an index of the CO_2 production rate of the hive. The electronic magnetic valves were closed for 5 min, and the CO_2 increase during the period was monitored.

The honeybee behavior was monitored continuously using a charge-coupled device camera (MTV-D-54K0, Akizuki Denshi, Tokyo, Japan). Since temporal changes in a pixel's grayscale in a movie image reflect the movements of bees (Okada, Ikeno, Aonuma, & Ito, 2008), we analyzed the changes as time-series signals for each pixel. The grayscale signal was separated into low (approximate) and high (detailed) frequency components, using the discrete wavelet transform. Spatial properties of honeybee movements on the frame were calculated by averaging the detailed components, which correspond to rapid honeybee movements, over 3 sec. In our analysis, the first Daubechies (Haar) function was used for the mother wavelet. We used the image processing toolbox and wavelet toolbox of the MATLAB (version 7.6.0) software (The MathWorks, Inc., Natick, MA) for our image analysis.

Measurements were conducted in August 2008, at the campus of the University of Hyogo in Himeji, central Japan. The CO_2 production rate and honeybee movement index were obtained every 1–2 h for 24 h on August 6th and 7th. In our measurements, the sampling frequencies of signals were determined by their temporal properties; that is, the hive weight was measured every 10 min, temperature and humidity were measured every 5 min, and the CO_2 concentration was measured every 10 sec. These data were recorded and logged in a personal computer.

The weather was fine and calm during the study periods. The glass hive box was placed in a large container measuring 2.6 (width) × 1.4 (depth) × 2.0 (height) m, where the light environment was controlled as a fixed 14:10-h light:dark cycle.

RESULTS AND DISCUSSION

The hive mass decreased in the morning and increased in the afternoon (Figure 2). The difference between highest and lowest masses in a day was around 100 g on each day, but there was only a net 50-g decrease over the 3 experimental days. The small change in the mass of the hive might have been caused by honey collection by the honeybees.

The temperature within the hive increased during the day and decreased at night. The hive temperature at night was slightly higher, by around 3ºC, than that outside. The peaks of the temperature inside the hive appeared later than those of the ambient. Humidity within the hive was stable, but outside it increased by night and decreased by day. The hive humidity was always higher than that of the ambient, which fluctuated by up to 30%. There were no differences in temperature and humidity among the sensor locations in the hive. The magnitude and fluctuation patterns of the active hive were different from those of the ambient, suggesting that honeybees controlled these environmental factors actively. However, since passive regulation by hive materials and hive structure has also been suggested to control the environment (Jones & Oldroyd, 2007), additional studies, such as a comparison of active and empty hives, are necessary to reveal the mechanisms

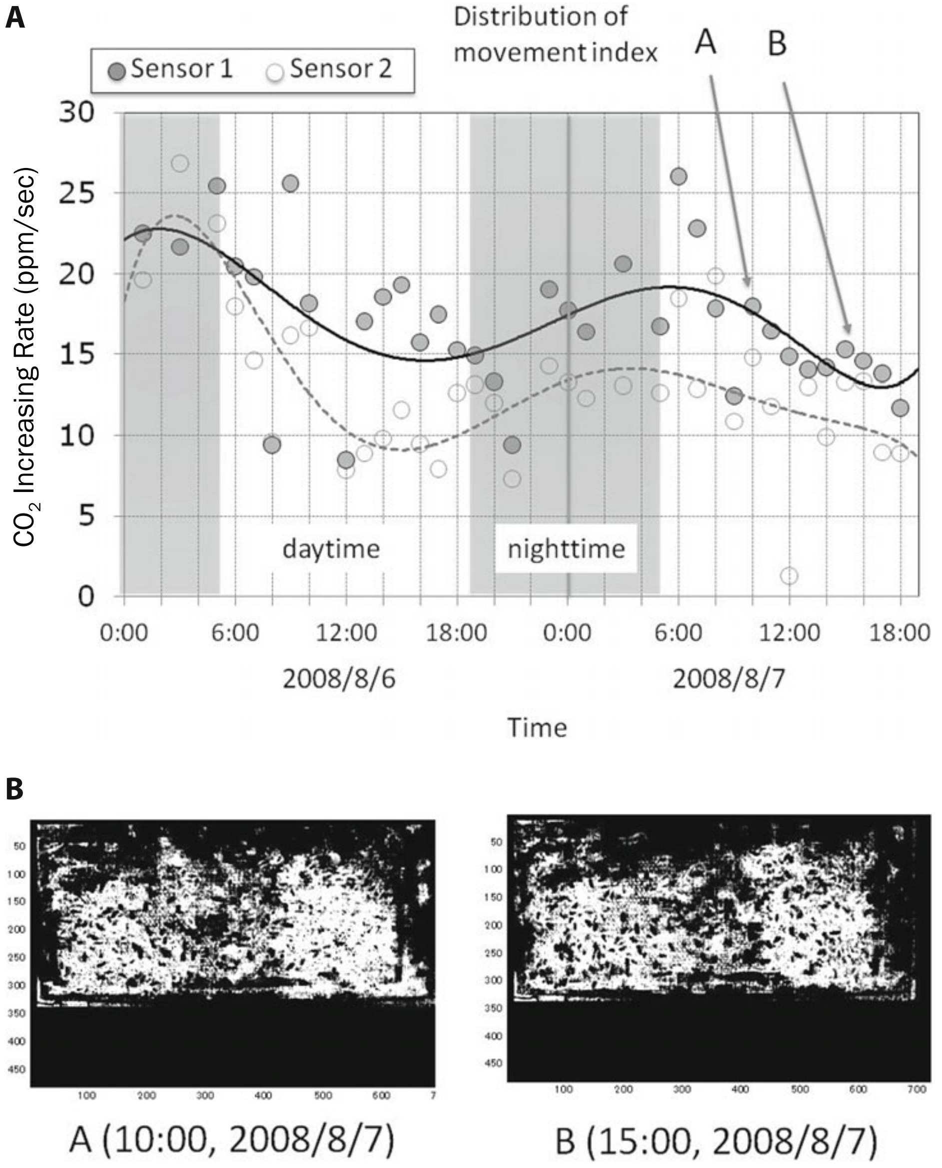

Figure 3. (A) CO_2 production and (B) movement index on summer days. The CO_2 production rate was estimated from the rate of the CO_2 increase with both the hive entrance and ventilator closed for 5 min. Honeybee movements in a morning (10 a.m.) and afternoon (3 p.m.) are shown in a grayscale image of the movement index.

of microclimate controls in the hive. In the present study, the different patterns and magnitudes of humidity inside and outside the hive were not similar to the patterns of temperature. This suggests that honeybees might control the two factors with different mechanisms.

The hive CO_2 concentration had two peaks per day: a higher peak in the early morning and a smaller peak in the evening. This pattern was observed by all CO_2 sensors. Since the outside CO_2 concentration normally decreases during the day, because of the photosynthesis of plants and the air turbulence resulting from high wind speeds (Luo & Zhou, 2006), this unpredicted increase might be caused by a circadian activity of honeybees (Kronenberg & Heller, 1982).

CO_2 production in the hive was high in the early morning and low in the afternoon, which corresponded with changes in the movement index of the colony (Figure 3). The high CO_2 production in the early morning might be caused by the high activities of honeybees, because CO_2 concentration had also increased rapidly at that time. The rapid decrease in hive weight following the increases in hive CO_2 production and CO_2 concentration suggests that foragers had flown from the hive to forage.

In the present study, we developed a system for monitoring multiple factors of a hive environment and honeybee behavior continuously for several days. Our system can also measure the CO_2 production of a honeybee colony continuously and automatically by regulating the closure of the entrance and ventilation holes. This system will be very useful in determining the mechanisms of nest climate control by honeybees, which is one of the behaviors that define honeybees as social insects. Monitoring the hive weight and CO_2 production together with honeybee behavior over a year using this system will reveal the energetic efficiency of a honeybee colony that enables it to survive severe winters for years with a large population. Simultaneous measurements of hive temperature and humidity, honeybee behavior, and energy consumption of the hive will clarify the sociophysiological mechanisms for honeybee control of environmental factors. The various data streams obtained in this system will contribute to understanding of the colony homeostasis of the honeybee from many perspectives.

However, there are still improvements to be made to the system. One is to make it possible to monitor honeybee behavior from both sides of the hive. This will be achieved by separating the amplifier from each sensor and placing these small sensors in a hive without disturbing colony activity.

Long-term measuring systems for animal behavior and climatic conditions are crucial for understanding biological mechanisms and functions. Since the dynamics of these objectives are unsteady and unpredictable, a traditional approach, such as verification of a hypothesis, is insufficient for studying the nature of animals and ecosystems. We established open-ended systems for monitoring honeybee behavior and a colony environment. Long-term experiments using the system will increase our understanding of the mechanisms of the social behaviors of this insect and its role in natural processes.

CONCLUSION

Previous studies suggested the controlling of multiple environmental factors, such as temperature, humidity, and CO_2 concentration, in the hive of the honeybee, but it is still unclear how these environmental factors fluctuate differently from those outside the hive, and how honeybees control these factors purposely. To answer these questions, we developed a system to monitor the honeybee hive environment and honeybee behavior and energy consumption. Our system revealed the peculiar environment of the honeybee hive, which is partly related to honeybee behavior. Additional monitoring of the hive environment and honeybee behavior for longer periods would enable us to understand the mechanism of environmental regulation by honeybees, which characterizes them as social insects.

AUTHOR NOTE

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