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LETTER TO THE EDITOR

# RFID monitoring indicates honeybees work harder before a rainy day

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Dear Editor,

Storms are usually accompanied by a drop in temperature, and an increase in wind and barometric pressure and rainfall, which have negative impacts on most activities, survival and reproduction in insects (Gillot, 2005). The majority of studies have mainly focused on how the flight activity of various flying insects such as honeybees, bumble bees, horse flies and leafminers were directly influenced by intraday weather changes (Burnett & Hays, 1974; Lundberg, 1980; Casas, 1989; Vicens & Bosch, 2000). However, accumulating evidences showed that animals can make behavioral changes before storms, which is enormously important for their survival in severe weather conditions. Before upcoming storms birds unusually chirp and bathe with sand; native frogs croak and hide their egg masses; spiders spin shorter and produce thicker webs and wasps hide their comb before rains (Galacgac & Balisacan, 2009; Acharya, 2011). In early 1893, honeybees were reported as more active before storms (Inwards, 1893). In this study, we compared the working habits of foragers on days that were followed by a sunny day and those that were followed by a rainy day using the Radio Frequency Identification (RFID), which was developed and manufactured by the Honeybee Research Institute of Jiangxi Agricultural University in collaboration with the Guangzhou Invengo Information Technology Co., Ltd., and we showed that honeybees worked harder before a rainy day.

Three honeybee (*Apis mellifera*) colonies were maintained at the Honeybee Research Institute, Jiangxi Agricultural University, Nanchang, China. Each colony had four full frames, with approximately 6000 workers and a laying queen. Newly emerged workers ($n = 300$) of each experimental colony were each glued with an RFID tag on the dorsal surface of the thorax with shellac. Tagged workers were introduced to their natal colonies, and monitored 24 h per day during a period of 34 days by an RFID reader (Fig. 1). When tagged workers entered and exited a colony, the RFID system recorded their unique ID, time stamp and direction. Duration for each trip was calculated based on the exit and entrance time stamps after sorting the data based on each RFID. Flights with duration longer than 5 min were considered as foraging trips (Calderone & Page, 1988). To be conservative, we used only data from bees that were older than 21 days. We used foraging data only from good foraging days: those that had temperatures from 15 to 34°C, sunny, no precipitation and wind $< 12$ km/h. Rainy days were defined as those with daily precipitation $> 5$ mm. We summed the duration of all foraging trips per day of each forager as "foraging duration", and defined "quitting time" as the last returning trip. For further statistical analysis, we collected a delta time of each bee by using daily sunset to subtract their quitting time. Sunset time for each day was obtained from the website (http://sunrise.supfree.net/). We classified the foraging data in two categories, days that were followed by a sunny day, and days that were followed by a rainy day. Foraging duration and the delta time between sunset and quitting time were transformed by taking their square root, which made the data to be normally distributed (Bartlett, 1947). The transformed data were then analyzed with analysis of variance (ANOVA) in Statview 5.0 (SAS Institute Inc., Gary, NC, USA). In addition, data of meteorological factors including humidity, temperature, barometric pressure and precipitation during the experimental period were recorded by Jiangxi Agricultural University weather station which is approximately 500 m away from experimental hives. Foraging data and its relative weather data for the next day were analyzed using generalized linear model analysis in R statistical

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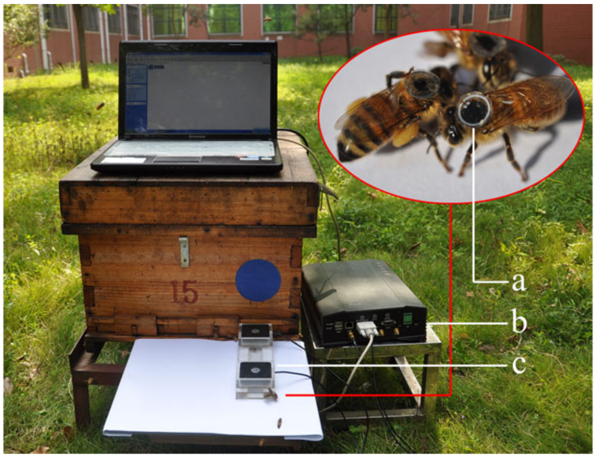


**Fig. 1** One of the experiment colonies, honeybee Radio Frequency Identification (RFID) system and tag-marked workers. a: tag; b: recorder; c: reader. The tag has a diameter of 3 mm and is 0.08 mm thick and weighs 1 mg.

package (https://www.r-project.org/) for testing the correlation between them.

Our results showed that there was a significant difference in foraging duration depending on the next day's weather ($F_{1,832} = 47.38$, $P < 0.001$; Fig. 2A), with bees spending longer total times outside the hive on days that were followed by a rainy day compared to those followed by sunny days. Bees also quit later on days that were followed by a rainy day (delta time followed by a rainy day was significantly shorter than that followed by a sunny day, $F_{1,819} = 26.10$, $P < 0.001$; Fig. 2B). These suggest that honeybees may attempt to collect more food before rainy days as a food shortage prevention. These behavioral changes possibly can lead to increasing colony productivity prior to inclement weather. Moreover, there was a strongly correlation linked the honeybees' foraging duration and the delta barometric pressure, delta humidity, delta temperature and precipitation of the next day ($P <$ 0.01), while the delta time between sunset and quitting time was in strong relation only with the delta barometric pressure ($P < 0.01$) and precipitation ($P < 0.05$) (Table 1).

Pervious studies revealed that cats may use their hair to detect slight changes of static electricity in the air during weather change for weather forecasting (Acharya, 2011). Sharks are extremely sensitive to the barometric pressure even it drops of a few millibars before rain (Toothman, 2008). Honeybees have been shown to be able to detect changes in carbon dioxide levels, relative humidity, temperature and air pressure (Southwick & Moritz, 1987), and even electric changes before thunderstorms (Warnke, 1976). These findings, and our results in Table 1, are consistent with the interpretation that honeybees may have an ability of measuring changes of meteorological factors such as barometric pressure, humidity and temperature to forecast the coming weather.

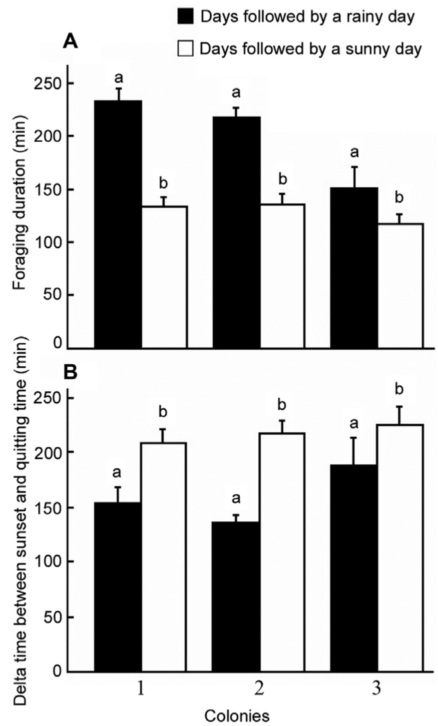


**Fig. 2** (A) Mean (±SE) foraging duration (summed total time spent on foraging) by foragers on days that were followed by a rainy day (solid) or those by a sunny day (open). There were significant differences between the two types of days and among colonies ($F = 47.38$, $P < 0.001$; $F = 9.61$, $P < 0.001$) but interactions between the foraging duration and colonies were not significant ($F = 2.63$, $P > 0.05$). Data were analyzed after square root transformation but presented here without transformation. (B) Mean (±SE) delta time between sunset and quitting time on days that were followed by a rainy day (solid) or a sunny day (open). Quitting time was defined as the return time of the last foraging trip. Delta time was collected by using daily sunset to subtract the quitting time of each forager. There were significant differences between bees working on the types of days ($F = 26.10$, $P < 0.001$), but differences among colonies and interactions between the colony and type of days were not significant ($F = 0.41$, $P > 0.05$; $F = 1.97$, $P > 0.05$). Data were analyzed after square root transformation but presented here without transformation. Different letters "a" and "b" on the top of each bar indicate significant differences ($P < 0.05$, analysis of variance test).

Honeybees are widely acknowledged as a model species for the studies of social behavior, social organization and even neurology (Robinson *et al.*, 2005; Qin *et al.*, 2014). Our results showed that honeybees change their habits by increasing foraging duration and working

**Table 1** Correlation analysis of weather factors for the next day with honeybee foraging durations and delta time between sunset and quitting time.

| Behavioral types | Weather factors | df | Mean square | *F* | *P*-value |
|---|---|---|---|---|---|
| Foraging duration | ΔHumidity | 1 | 283166.06 | 17.85 | $<0.01$ |
| | ΔTemperature | 1 | 461890.63 | 29.11 | $<0.01$ |
| | ΔBarometric pressure | 1 | 201768.67 | 12.72 | $<0.01$ |
| | Precipitation | 1 | 759915.05 | 47.89 | $<0.01$ |
| Delta time | ΔHumidity | 1 | 279.31 | 0.01 | 0.92 |
| | ΔTemperature | 1 | 401.37 | 0.02 | 0.90 |
| | ΔBarometric pressure | 1 | 488494.09 | 18.87 | $<0.01$ |
| | Precipitation | 1 | 142385.63 | 5.50 | $<0.05$ |

Note: Delta humidity, barometric pressure and temperature were calculated by the maximum subtracted to the minimum of each day. Data of honeybee flight activities and weather factors were analyzed with generalized linear model analysis in R statistical package, where foraging duration and delta time were dependent variables, weather types of next day were factors and weather factors were covariates.

later in the afternoons. Because these changes are at the social level, and not just at individual level, the question remains how honeybee workers can communicate the need to work "harder" after perceiving the predictors for the impending weather changes. Future studies should focus on what factors in the weather system are perceived and how bees communicate the need for behavioral changes to the whole colony.

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