Defecation initiates a stereotyped behavior in the cricket *Gryllus bimaculatus*

Keisuke NANIWA[a], Yasuhiro SUGIMOTO[b]
Koichi OSUKA[b], Hitoshi AONUMA[a]
[a]Research Institute for Electronic Science, Hokkaido University, Japan
naniwa@hokudai.ac.jp
[b]Dept. of Mechanical Engineering, Osaka University, Japan

1 Introduction

In insects, feces contain pheromones and attracts homogeneous insects. On the other hand, the odor of feces also would risk insects by recruiting parasites. It might be necessary for insects whether staying in the same place or leaving after defecation. Previous studies mainly focused on the metabolic system of defecation [1]. In insects, it is known that the peripheral nervous system controls food digestion and excretion [2], and information on intestinal pressure is detected in sensory nerves derived from the abdominal ganglion. On the other hand, there are few findings on behavioral expression associated with defecation.

We here focus on the sequence of behavior associated with defecation in the cricket. In our previous study [3], we focused on walking initiated by defecation. In this paper, we found that defecation initiates not only walking but a stereotyped behavior. We demonstrate the cricket shows stereotyped behavior associated with defecation where cricket starts walking backward several steps just before defecation, bent the abdomen towards the ground and then walks forward immediately after defecation. We also demonstrate that ascending neuronal information from the terminal abdominal ganglion introduce the stereotyped behavior associated with defecation.

2 Material and method

2.1 Animals

Crickets *Gryllus bimaculatus* (DeGeer) used were raised in a laboratory colony. They were reared on a 14h: 10h light and dark cycle at 28 ± 2°C (lights on at 6:00 h), and fed a diet of insect food pellet (Oriental Yeast Co., Tokyo, Japan) and water ad libitum. Adult crickets that had molted within 2 weeks before the experiment were used in this study.

2.2 Behavioral experiments

Test cricket was placed in an acrylic rectangular parallelepiped arena of the plastic sheet whose floor surface was mat-processed, 300 mm in width, 200 mm in depth, 100 mm in height. Behavior experiments were performed from 7:00 pm to 9:00 am on the next day in order to avoid the influence of the diurnal activity, and the LED light always irradiates the arena during the recording. A Web camera recorded the behavior with USB connection Manufactured by Logicool Co., Ltd. for later analysis. Behavior patterns before and after defecation were classified into three types: walking forward, walking backward, and resting when crickets were stationary for more than 10 minutes. Behavior pattern and the direction of the waling were measured.

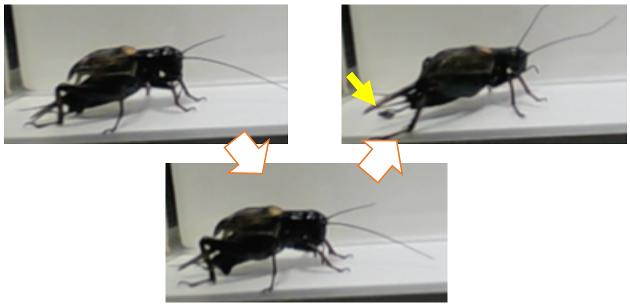

Figure 1: Stereotyped behavior when cricket defecates. Cricket first stopped walking. Cricket then bent the abdomen while slightly moving backwards and defecation. After defecation, cricket begins to walk forward. The yellow arrow shows defecated fece.

3 Behavior analysis results before and after defecation

3.1 Behavior analysis between intact cricket and headless cricket

The defecation behavior of crickets was expressed about every 3 hours in both intact crickets and headless crickets. The behavior associated with defecation was a stereotyped shown in Fig. 1. A sequence of behavior was that cricket stops moving in the arena, lifting the body while walking backward several steps just before defecation, bending the abdomen onto the ground, and then defecating feces. Fig. 2 shows behaviors immediately before and after defecation of intact crickets and headless crickets.

Backward walking immediately before defecation of the intact cricket was about 30%, and walking distance was about 14 mm. On the other hand, in the headless cricket,

Table 1: Expression of backward walking followed by excreting of feces in crickets. Backward walking was observed in both intact and headless crickets but not when the abdominal nerve cord was cut. N: number of animals, n: times of defecation. Sham: sham operation (only cuticle of the abdomen was cut)

Speciments	Intact	Headless	Nerve cord cut	Headless and nurve cord cut	Headless and sham
Number of speciments	N(n) 7(18)	7(33)	16(66)	15(33)	10(40)
Frequency of stereotyped defecation walking	N(n) 2(5)	6(17)	0(0)	0(0)	6(14)

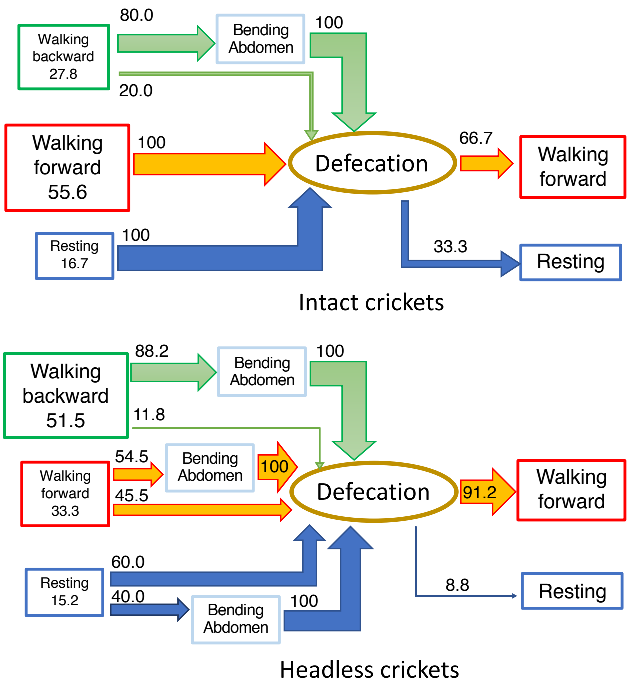

Figure 2: Ethograms for defecation behavior of crickets. These ethograms indicate the behavior of crickets before and after defecation. Top ethograms for defecation behavior in intact crickets (n=18). Bottom ethograms for defecation behavior in headless crickets (n=33). Each number indicates the exact value (%) of the occurrence or transition probability.

backward walking just before defecation at a frequency of about 50%, and the distance was longer than intact crickets (about 27 mm). After defecation, both intact crickets and headless crickets moved forward, and the frequency of forwarding movement immediately after defecation especially at the headless cricket was much higher (exceeding 90%). The behavior of bending the abdomen just before defecation was seen only in stereotyped behavior in intact crickets. On the other hand, in headless crickets, the behavior increased in all pre-behaviors.

Behavior analysis of intact and headless cricket before and after defecation showed a stereotyped behavior of "walking backward a few steps just before excretion, bent the abdomen towards the ground and walking forward immediately after excretion" regardless of the presence or absence of the head. In particular, the series of behaviors became more prominent by resecting the head. Since the signal from the head suppresses the series of reflex behaviors associated with defecation, it is suggested that a series of stereotyped behaviors were caused by reflection inside the cricket other than the head.

3.2 Behavioral analysis in cricket that cut abdominal nerve

To examine whether the neuronal signals mediated defecation-related walking from the abdominal nervous system, we cut the abdominal nerve cord at different positions and observed the resultant behavior. The result of the behavioral experiment is the table shown in Table 1.

Based on these results, it is suggested that signals from the abdominal terminal ganglion cause stereotyped behavior. The sensory nerve that detects the state of the rectum of insect originates from the abdominal terminal ganglion. In the cricket, as the abdominal terminal ganglion detects the expansion of the volume of the rectum just before defecation, it induces a receding movement just before defecation and the forward movement immediately after defecation is induced by the volume reduction of the rectum after defecation we concluded.

4 Conclusion

We here found that the cricket expresses stereotyped behavior accompanying backward movement, bent the abdomen towards the ground and forward movement before and after defecation. We suggest that the stereotyped behavior associated with defecation is induced by the signal from the abdominal terminal ganglion. As a future study, it is necessary to identify the neuronal circuit to introduce stereotyped behavior associated with defecation for further understanding.

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