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## Directionality of Cricket Giant Interneurons to Escape Eliciting Unidirectional Air-Current

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**ABSTRACT**—Response properties of six giant interneurons (GIs) 8-1, 9-1, 9-2, 9-3, 10-2 and 10-3 of the cricket, *Gryllus bimaculatus*, were investigated with a short duration unidirectional air-current stimulus which elicits escape behavior of the insects effectively. Air-currents were presented to the insects from 12 different directions in the horizontal plane in order to explore the directional characteristics of the GIs. The frequency dependency of velocity thresholds (threshold curves) of the GIs changed with the direction of stimulus air-current. Preferential directions of the GIs to the air-currents were ipsilateral-rear in GI 8-1, ipsilateral-front in GI 9-1, ipsilateral-rear in GI 9-2, ipsilateral-front in GI 9-3, ipsilateral-rear in GI 10-2 and ipsilateral-front in GI 10-3, with respect to the ventral nerve cord containing the axons. Intensity-response curves suggest that GIs 8-1 and 9-1 show the maximum response to air-current velocities lower than 300 mm/sec regardless of the stimulus direction. On the other hand, response magnitudes of GIs 9-2 and 9-3 increased proportional to the logarithm of the stimulus velocity up to 300 mm/sec and did not show a plateau even at 300 mm/sec regardless of the stimulus direction. Based on the directional characteristics revealed in the present study, a possible neural mechanism for encoding the direction of air-current by the combinational activity of GIs 9-2 and 9-3 is discussed.

### INTRODUCTION

Several giant interneurons (GIs) have been identified both physiologically and morphologically in orthopteran and blattellan insects (Boyan and Ball, 1989; Boyan *et al.*, 1989; Kämper, 1984; Kanou and Shimozawa, 1984; Shen, 1983; Westin *et al.*, 1977). In the terminal abdominal ganglion (TAG) of the central nervous system of the insects, GIs receive inputs from a large number of wind-sensitive filiform hairs on the cerci, a pair of appendages at the abdominal tip. In some cases, the information from filiform hairs may be relayed by local non-spiking interneurons in the TAG (Baba *et al.*, 1995; Bodnar, 1993; Bodnar *et al.*, 1991; Kobashi and Yamaguchi, 1984). The cricket GIs, which have their somata in TAG, extend their axons to the dorsal region of the deutocerebrum of brain (Hirota *et al.*, 1993). Information about air-disturbances detected by cercal filiform hairs is transmitted to the other parts of the body mainly via the GIs. Cercal inputs are effective to elicit some stereotyped behaviors such as escape running, defensive posture or kicking in crickets and cockroaches (Bentley, 1975; Camhi *et al.*, 1978; Dumpert and Gnatzy, 1977; Gnatzy and Heißlein, 1986; Kanou *et al.*, 1995). The cercus-to-GI system is, therefore, an ideal object of a case study in neuroethology (Boyan and Ball, 1989; Gnatzy and Heißlein, 1986; Liebenthal *et al.*, 1994).

A short duration unidirectional air-current (air-puff) is biologically significant. It elicits escape behavior in crickets and cockroaches. Their escape behaviors are highly directional, i.e. the insects escape away from the stimulus source in most cases (Camhi and Tom, 1978; Gras and Hörner,

1992; Kanou *et al.*, 1995). Encoding of the air-current direction is indispensable for successful escape from predator attacks. Therefore, investigation of directional properties of the sensory system must extend our knowledge about the relation between neural activities and resulted escape behaviors. Thus many studies have dealt with the directional characteristics of the cercus-to-GI system of the insects (Bacon and Murphey, 1984; Edwards and Palka, 1974; Gnatzy and Tautz, 1980; Kämper, 1984; Matsumoto and Murphey, 1977; Westin *et al.*, 1977).

Earlier studies of cricket GIs have contributed to reveal many significant aspects of the directional characteristics of GIs of *Acheta domestica* and *Gryllus campestris*, which are closely related species with *G. bimaculatus* used in the present study (Edwards and Palka, 1974; Kämper, 1984; Levine and Murphey, 1980; Murphey *et al.*, 1977). However, those studies were not enough to reveal the real directionalities of the neurons because a bidirectional air-motion (e.g. tones) was employed as a stimulus. In order to understand the role of each cricket GI for encoding the wind direction and for triggering the directional escape, the directionalities of all the GIs must be investigated with quantitatively controlled unidirectional air-currents. However, unidirectional air-motion was used only in a small number of studies of cricket GIs (GI 8-1: Tobias and Murphey, 1979, GIs 9-3 and 10-2: Miller *et al.*, 1991, GI 10-3: Jacobs and Miller, 1985; Jacobs *et al.*, 1986; Miller *et al.*, 1991). Although the directional sensitivity of all the GIs of american cockroach (*Periplaneta americana*) have been well investigated by using unidirectional wind-puff stimulus (Comer and Camhi, 1984; Daley and Camhi, 1988; Vardi and Camhi, 1982; Westin *et al.*, 1977), such study in cricket has not been done yet.

The aim of the present study was to investigate how the cricket GIs respond to the stimulus which elicit escape

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behaviors on the insects effectively, and to compare the response characteristics of the GIs with each other. This kind of investigation must be essential in order to understand the roles of GIs for the elicitation of the directional escape behaviors. In the present study, frequency dependency of velocity thresholds of 6 pairs of cricket GIs were investigated in combination with their directional characteristics by using a unidirectional air-current stimulus. This is the first case that the thresholds and directional characteristics of the 6 pairs of GIs have been investigated by the same stimulating method and compared with each other.

A preliminary account of this work has partly been reported (Kanou, 1991).

## MATERIALS AND METHODS

### Animals

Adult female crickets, *Gryllus bimaculatus*, 1-2 weeks after imaginal molt, were used for the experiments. The insect colony was reared in the laboratory. They were fed with "insect chow" (Oriental Koubo) and carrots. A shell vial filled with water and plugged with a cotton ball was served as water supply and egg laying place. The breeding room was kept at 28°-30°C, and the photoperiod was DL 10:14.

### Air-current stimulus

The stimulus was a short duration unidirectional air-current that was produced by a wind-tunnel consisting of a pair of push-pull driven loud speakers (Kanou and Shimozawa, 1984). The electrical signal fed to the speakers was a half cycle cosine wave, i.e. it started from the bottom of the signal and ended at the next peak of the signal. The electrical potential at the peak was held unchanged for 500 msec and then returned to the initial level. The initial part of the signal drove the speakers in one direction (Fig. 1B) and resulted a quantifiable unidirectional air-current in the tunnel because speaker displacement is proportional to the driving signal. The air-current made in the tunnel was, therefore, a kind of air-puff. As the signal of the returning phase was not specifically controlled, only the initial part of the air-motion was used as the stimulus. The time course of the velocity change and an example of the neural response to the stimulus are shown in Figure 1. The peak velocity and the frequency of the air-current were controlled separately. Although a whole cycle or a continuous oscillation of sinusoidal air-motion was not used in the present experiments, the term "frequency" is used in this paper in order to express the rate of air-displacement, because the stimulus wave form was a part of a sinusoid, and it was more intuitively understandable to express the air-displacement in terms of frequency than of the time of 1/2 cycle. The inter-stimulus interval was 2 sec for threshold measurement and was 5 sec for the measurement of intensity-response relations. Details about the stimulus equipment including the calibration procedures were the same as was reported in previous papers (Kanou, 1991; Kanou and Shimozawa, 1984).

### Animal preparation, neural recording and threshold measurement

The head, thorax and ovipositor were removed from the abdominal part of the cricket. The abdominal tergites and viscera were removed in order to expose the ventral nerve cord. The specimen was pinned on a cork board with the dorsal side up and was set at a center of the wind-tunnel. The responses of GIs were intracellularly recorded from the axons in the right ventral nerve cord between the 4th abdominal ganglion and the TAG. The direction of the stimulus air-current was changed in the horizontal plane by 30° step around the insect by rotating the wind-tunnel. Threshold curves of previously identified GIs 8-1 (MGI), 9-1 (LGI), 9-2, 9-3, 10-2 and 10-3 (Kanou

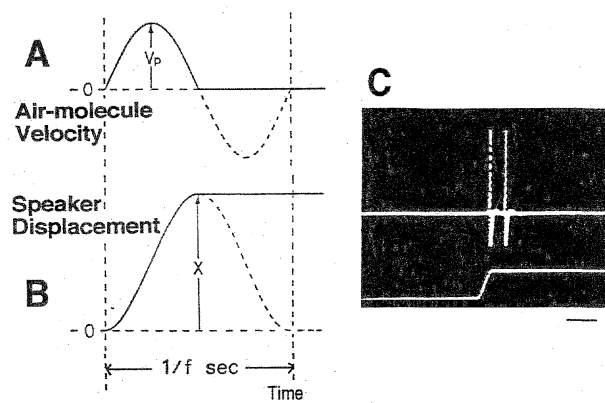


Fig. 1. A: velocity of air-molecules when the speakers were moved as in B.  $V_p$ : peak velocity of air-current. B: speaker displacement.  $X$ : stroke amplitude. C: an example of a GI response to a unidirectional air-current stimulus. Time scale: 20 msec.

and Shimozawa, 1984; Mendenhall and Murphey, 1974) were measured by the air-current stimulus from 12 different directions by changing the stimulus velocity and frequency independently. In order to express the directional characteristics of the GIs more clearly, threshold velocities at 5 and 50 Hz were plotted on a polar coordinate to make directionality curves (Fig. 12). The threshold was defined as the minimum peak velocity of the stimulus required to elicit an action potential at a certain frequency. Although GIs 10-2 and 10-3 are spontaneously active (Kanou and Shimozawa, 1984), their stimulus driven impulses could easily be discriminated from spontaneous ones by using both audio monitor and storage oscilloscope (Sony Tektronix, R5111A).

Five percent Lucifer Yellow CH dissolved in 0.3 mol/l LiCl, which was placed in the glass microelectrode beforehand, was iontophoretically injected after the physiological investigation for the morphological identification of GIs based on previous papers (Kanou and Shimozawa, 1984; Mendenhall and Murphey, 1974).

### Intensity-response curves

Intensity-response relations (peak velocity of air-current vs. number of action potentials) were measured at each stimulus direction. At each air-current velocity, ten successive 50 Hz stimuli were applied with 5 sec intervals, and the averaged number of action potentials per stimulus was calculated. The averaged number of action potentials was again averaged by the number of animals used. Therefore, each point on the intensity-response curves was an average of  $10 \times N$  ( $N$ = number of animals) trials.

## RESULTS

Velocity thresholds of six GIs in the cricket *G. bimaculatus* were measured with a unidirectional air-current stimulus in order to explore the directional characteristics of them. The thresholds of the GIs changed with frequency and direction of the stimulus. Thresholds at 5 and 50 Hz were plotted on polar coordinates in order to express the directionalities more clearly. In four out of the six GIs, intensity-response relations were investigated in order to explore how the GIs encode the velocities of stimulus air-current. Followings are the response characteristics of each GI revealed in the present study.

*GI 8-1 (MGI)*

Threshold curves of GI 8-1 for different stimulus directions moved almost in parallel fashion along the ordinate (Fig. 2). The GI showed high-pass (or low-cut) characteristic regardless of the stimulus direction, i.e. the threshold curves showed negative slopes between 2 and 20 Hz, and showed almost flat slopes between 20 and 100 Hz. The slope between 2 and 20 Hz was ca. -16 dB/decade (threshold velocity decreases 16 dB for a decade increase of frequency: ref. Kanou and Shimozawa, 1984) on the average.

Velocity thresholds plotted on a polar coordinate revealed that GI 8-1 showed the lowest threshold to the air-current from ipsilateral-rear direction (R120 and R150) with respect to the ventral nerve cord containing the axon, and was slightly insensitive to air-currents from the opposite direction at both 5 (Fig. 12A; open circles) and 50 Hz (Fig. 12A; filled circles). However, when compared to other GIs investigated in the present study, the sensitivity of GI 8-1 was not as strongly biased to one particular direction.

On the other hand, intensity-response curves measured for air-currents from 12 different directions revealed that the response magnitude of GI 8-1 showed the largest increasing rate to the air-currents from R60 and R90 directions (Fig. 3). Therefore, in the supra-threshold domain, GI 8-1 shows greater

response to air-currents from R60 and R90 than from other directions. In most cases, the number of impulses show maximum between 100 and 300 mm/sec.

*GI 9-1 (LGI)*

Threshold curves of GI 9-1 showed negative slopes between 2 and 20 Hz, and showed flat or slightly positive slopes between 20 and 100 Hz (Fig. 4). Therefore, the GI has high-pass (or low-cut) characteristic as does GI 8-1. The change in stimulus direction resulted threshold differences mainly at lower frequencies (2~5 Hz).

Directionality curves made by the thresholds at both 5 (Fig. 12B; open circles) and 50 Hz (Fig. 12B; filled circles) have oval shapes. Thus GI 9-1 is relatively sensitive to air-currents from ipsilateral-front (R30 and R60) and contralateral-rear (L120 and L150) directions, and is insensitive to air-currents from the orthogonal directions. Intensity-response curves of GI 9-1 measured by air-currents from 12 different directions also reveal that the GI 9-1 shows the largest response to air-currents from R60 direction (Fig. 5).

Regardless of the stimulus direction, GI 9-1 showed maximum responses to stimuli between 10 and 100 mm/sec. For example, when the stimulus was from R60 direction, the GI showed a maximum response to 30 mm/sec air-current

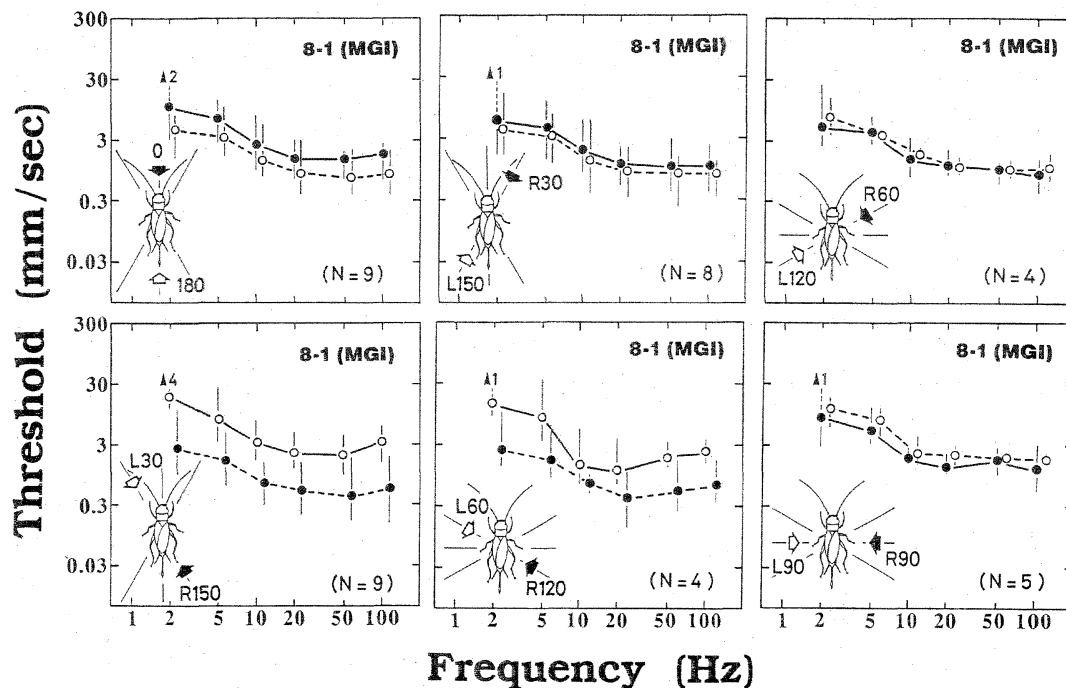


Fig. 2. Threshold curves of GI 8-1 (MGI) measured by a unidirectional air-current from 12 different directions. Stimulus directions of the threshold curves with filled circles and solid lines are shown by filled (black) arrows in the insets. Threshold curves with open circles and dashed lines are for stimulus directions shown by open (white) arrows. Air-currents from 180° opposite directions were made by inverting the speakers driving signal. Regardless of the stimulus direction, GI 8-1 shows a high pass characteristic. See text for details. Each point shows the averaged threshold velocity of "N" animals. Vertical bars show variations of measured samples. In some cases, thresholds were higher than the highest velocity that the wind-tunnel could deliver. In such cases, the thresholds were tentatively determined as the averaged value of those could be measured, and dashed lines with upward arrowheads were used for the variation bars. The number beside the upward arrowheads indicates the number of animals with higher thresholds above the wind-tunnel could deliver.

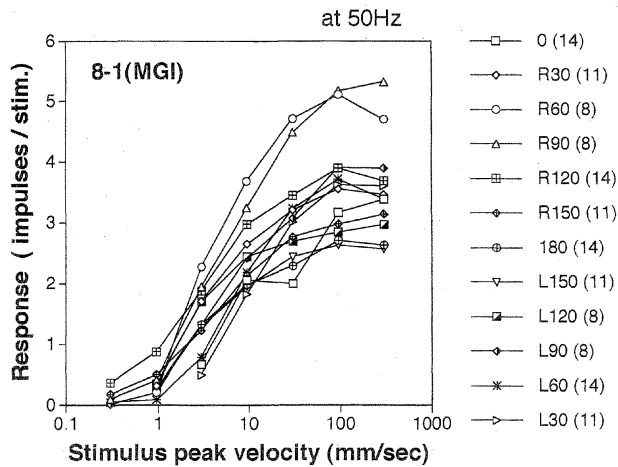


Fig. 3. Intensity-response curves of GI 8-1 measured by a unidirectional air-current from 12 different directions. The GI shows bigger responses to air-currents from R60 and R90 than from other directions. Numbers of impulses reached a plateau between 100 and 300 mm/sec in most cases. Numbers of animals used for the measurement are in parenthesis after the symbols showing stimulus directions.

and the response magnitude decreased when the velocity was higher than 30 mm/sec.

#### GI 9-2

According to the change of the stimulus direction,

threshold curves of GI 9-2 changed more drastically than those of GIs 8-1 and 9-1 (Fig. 6). Between 2 and 10 Hz, some of the threshold curves of GI 9-2 had negative slopes (ca. -20 dB/decade). Above 10 Hz, however, most threshold curves were almost parallel with the abscissa or showed slightly positive slopes.

The thresholds of GI 9-2 showed wide variations from animal to animal, unlike those of GIs 8-1 and 9-1. In particular, the thresholds to air-currents from L30, L150 and R30 had wider variations than other GIs.

Directionality curves of GI 9-2 at both 5 and 50 Hz are pear shaped. It suggests that the GI is the most sensitive to air-currents from R120 and R150 directions, and is less sensitive to air-currents from 0, L90 and L120 directions. The main receptive field of GI 9-2 is, therefore, in the ipsilateral-rear direction.

Intensity-response curves of GI 9-2 confirm that the GI is sensitive to air-currents from ipsilateral-rear directions (R90, R120 and R150) (Fig. 7). Unlike GIs 8-1 and 9-1, the numbers of impulses increase in proportion to the logarithm of the stimulus velocity up to 300 mm/sec regardless of the stimulus direction. As the response magnitude does not show a plateau even at 300 mm/sec, GI 9-2 may possibly encode air-current velocities higher than 300 mm/sec.

#### GI 9-3

Threshold curves of GI 9-3 moved almost in parallel along ordinate when the stimulus direction was changed (Fig. 8).

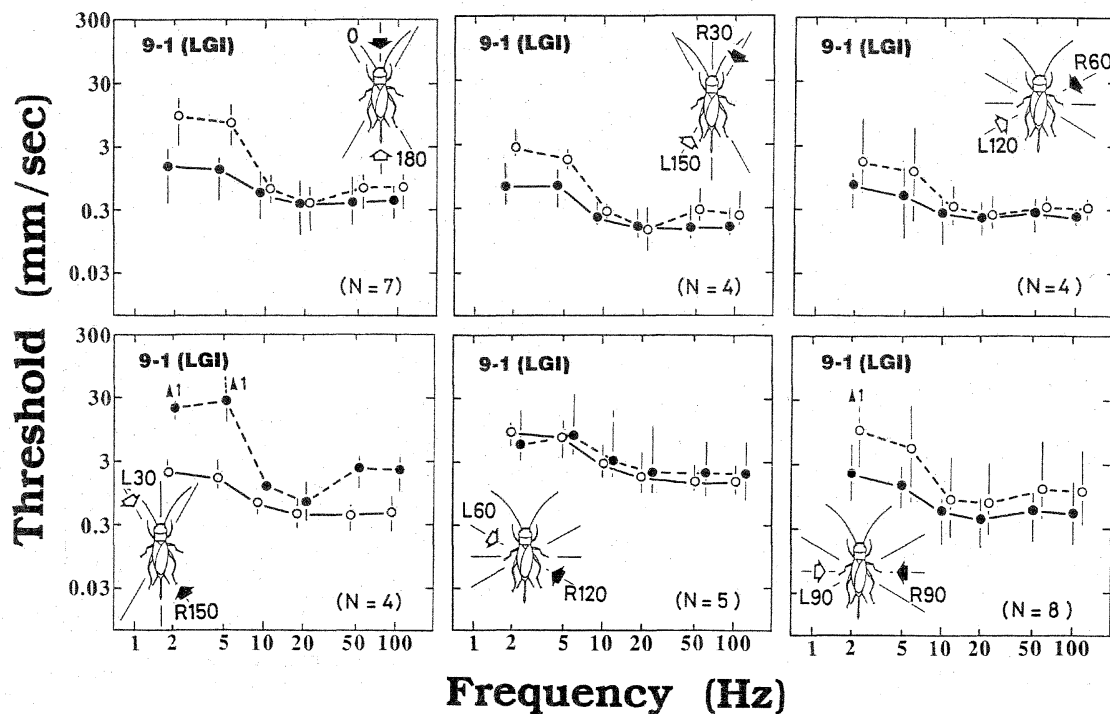


Fig. 4. Threshold curves of GI 9-1 (LGI) measured by a unidirectional air-current from 12 different directions. Regardless of the stimulus direction, GI 9-1 shows a high pass characteristic. Thresholds at lower frequencies change more widely with the change of stimulus direction than those at higher frequencies. See legends of Fig. 2 for other explanations.

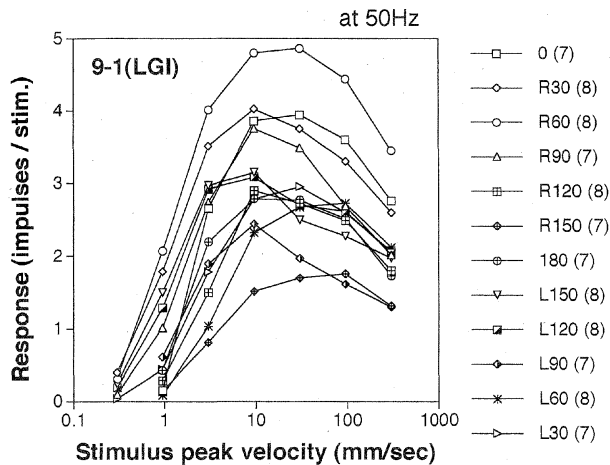


Fig. 5. Intensity-response curves of GI 9-1 measured by a unidirectional air-current from 12 different directions. All the intensity-response curves show their peaks between the air-current velocities of 10 and 100 mm/sec. See text for details. See legends of Fig. 3 for other explanations.

Most threshold curves of GI 9-3 measured for air-currents from 12 different directions showed slightly negative slopes and insensitivity to stimuli lower than 5 Hz. The threshold curves reveal that GI 9-3 is more sensitive to air-currents from R30, R60 and R90 than the air-currents from other directions. When

compared to the other GIs investigated in the present study, GI 9-3 is less sensitive; the lowest threshold was ca. 3 mm/sec.

Directionality curves show that the GI 9-3 is sensitive to air-currents from ipsilateral-front directions (R30, R60 and R90) and is less sensitive to air-currents from contralateral-front directions at both 5 and 50 Hz (Fig. 12D). Although the GIs 9-2 and 9-3 have different receptive fields, the shape of directionality curves of GIs 9-2 and 9-3 are very similar to each other, i.e. their receptive fields are symmetrical with respect to the transverse axis of the insect body (front-rear symmetry).

Intensity-response curves of GI 9-3 tell that the GI is sensitive to air-currents from ipsilateral-front directions (Fig. 9). The intensity-response curves are again very similar to those of GI 9-2, i.e. impulse numbers increased proportionally to the logarithm of stimulus velocity up to 300 mm/sec regardless of the stimulus direction. As all the intensity response curves did not show a plateau even at 300 mm/sec, the GI 9-3 could encode air-current velocities higher than 300 mm/sec as could GI 9-2.

#### GI 10-2

Threshold curves of GI 10-2 changed according to the change of stimulus direction (Fig. 10). Threshold differences caused by the stimulus direction were larger for higher frequencies. Most of the threshold curves of GI 10-2 measured

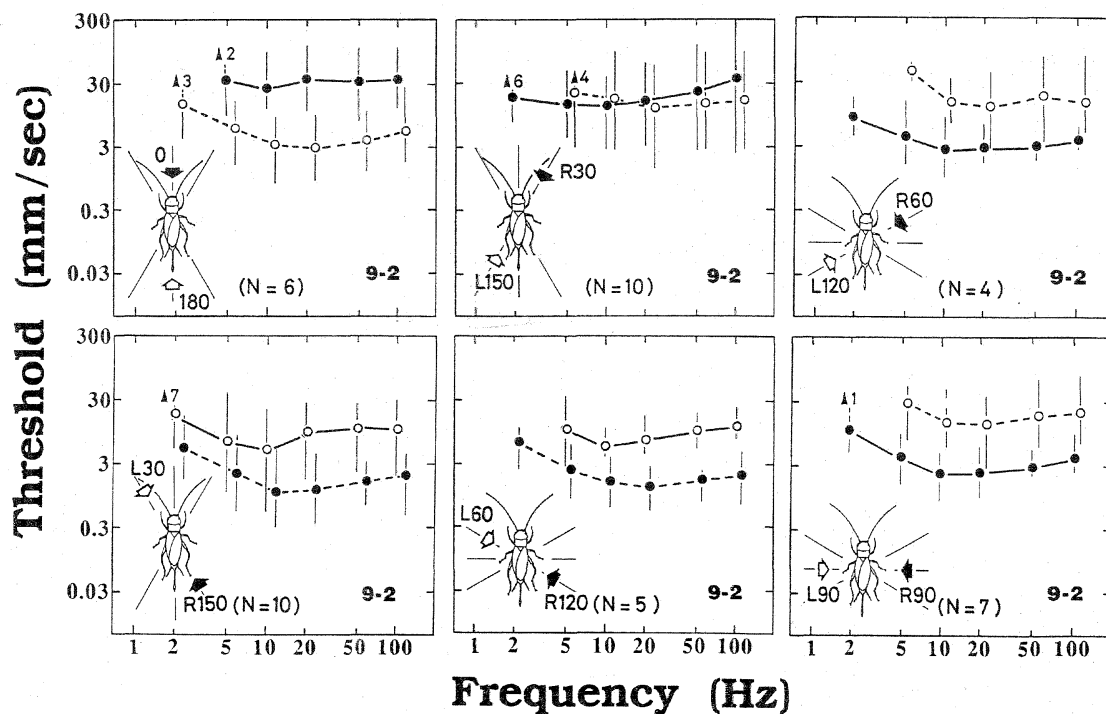


Fig. 6. Threshold curves of GI 9-2 measured by a unidirectional air-current from 12 different directions. Some of the threshold curves show negative slopes in lower frequencies, but slightly positive slopes in higher frequencies. See text for details. See legends of Fig. 2 for other explanations.

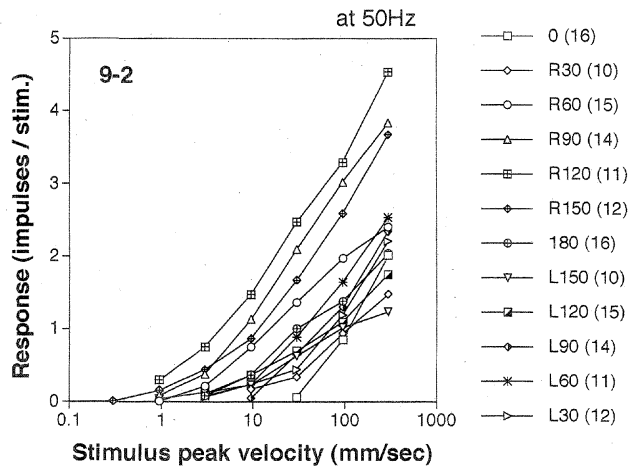


Fig. 7. Intensity-response curves of GI 9-2 measured by a unidirectional air-current from 12 different directions. The GI is sensitive to the air-current from ipsilateral-rear directions. Regardless of the stimulus direction, numbers of impulses increase almost in proportion to the logarithm of the velocity of the stimulus air-current and are not saturated even at 300 mm/sec. See legends of Fig. 3 for other explanations.

by air-currents from 12 different directions showed slightly negative slopes between 2 and 5 (or 10) Hz and slightly positive

slopes above these frequencies (Fig. 10). Thus GI 10-2 showed the highest sensitivity between 5 and 10 Hz. The slope of the threshold curves measured by air-currents from 0, L60 and L90 was about +20 dB/decade at higher frequencies. This means that the thresholds of GI 10-2 depend on the amount of the displacement of air-molecules in such stimulus conditions. Although most of the thresholds showed relatively small variations at each measured point, only the thresholds measured by air-currents from R30 showed large variations and higher thresholds than other cases. GI 10-2 was more sensitive than other GIs and the lowest threshold was ca. 0.03 mm/sec.

Directionality curves of GI 10-2 showed oval shapes (Fig. 12E). At 5 Hz stimulus, the GI was equally sensitive to air-currents from ipsilateral-rear and contralateral-front directions, and was insensitive to air-currents from the orthogonal directions. When the stimulus frequency was 50 Hz, the sensitivity to air-currents from ipsilateral-rear was still high, but the sensitivity to the air-currents from contralateral-front decreased by about 15-20 dB.

#### GI 10-3

The threshold curve of GI 10-3 changed according to the change of the stimulus direction (Fig. 11). Most of the threshold curves of GI 10-3 for air-currents from 12 different directions have slightly negative slopes in the lower frequency range (2

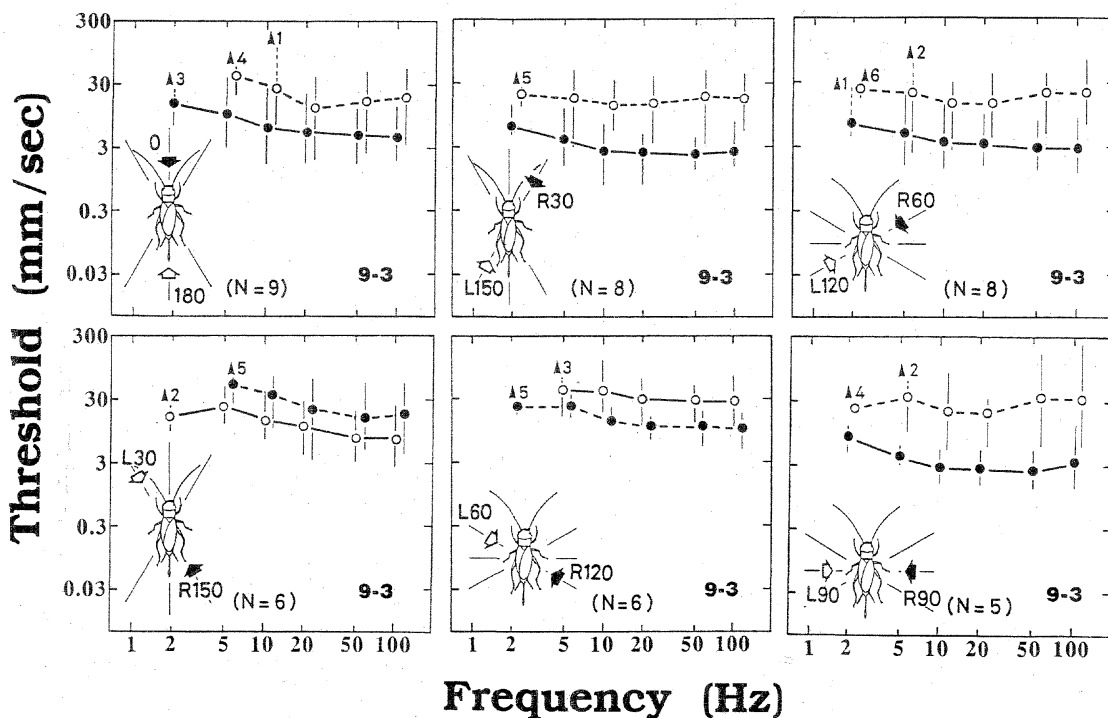


Fig. 8. Threshold curves of GI 9-3 measured by a unidirectional air-current from 12 different directions. Most of the threshold curves show slightly negative slopes. Threshold curves move almost in parallel along the ordinates when the stimulus direction was changed. See legends of Fig. 2 for other explanations.

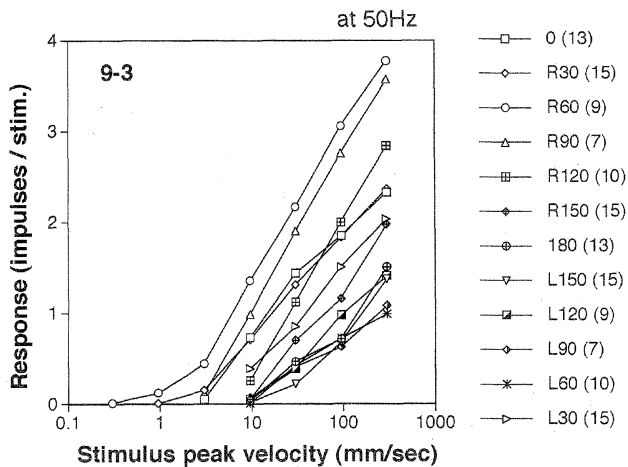


Fig. 9. Intensity-response curves of GI 9-3 measured by a unidirectional air-current from 12 different directions. The GI is sensitive to air-currents from ipsilateral-front directions. Regardless of the stimulus direction, numbers of impulses increase in proportion to the logarithm of the velocity of the stimulus air-current and are not saturated even at 300 mm/sec. See legends of Fig. 3 for other explanations.

~ 5 or 10 Hz) and have positive slopes in the higher frequency range. Therefore, GI 10-3 showed the highest sensitivity between 5 and 10 Hz. Such frequency characteristic is very similar to that of GI 10-2. The slope of the threshold curves measured by air-currents from R150, 180, L150, L120 and L90 was about +20 dB/decade in the higher frequency range. This suggests that the thresholds of GI 10-3 depend on the amount of the displacement of air-molecules in such stimulus conditions. Like GI 10-2, GI 10-3 is highly sensitive, i.e. the lowest threshold was 0.03 mm/sec or less.

The GI 10-3 showed an oval or dumbbell shaped directionality curve (Fig. 12F). At 5 Hz stimulation, the GI 10-3 was equally sensitive to the air-currents from ipsilateral-front and contralateral-rear directions. At 50 Hz, sensitivities to the air-currents from ipsilateral-front directions are almost same with those at 5 Hz stimulation. However, sensitivities to the air-currents from contralateral-rear directions at 50 Hz were about 10 dB higher than those at 5 Hz stimulation. This directional characteristic is again very similar to that of GI 10-2. The directionality curves of GIs 10-2 and 10-3 are, therefore, symmetrical with respect to the transverse axis of the insect body (front-rear symmetry) as are the directionality curves of GIs 9-2 and 9-3.

## DISCUSSION

### Frequency dependency of velocity thresholds

In our previous study, GIs 8-1, 9-1, 9-2 and 9-3 were

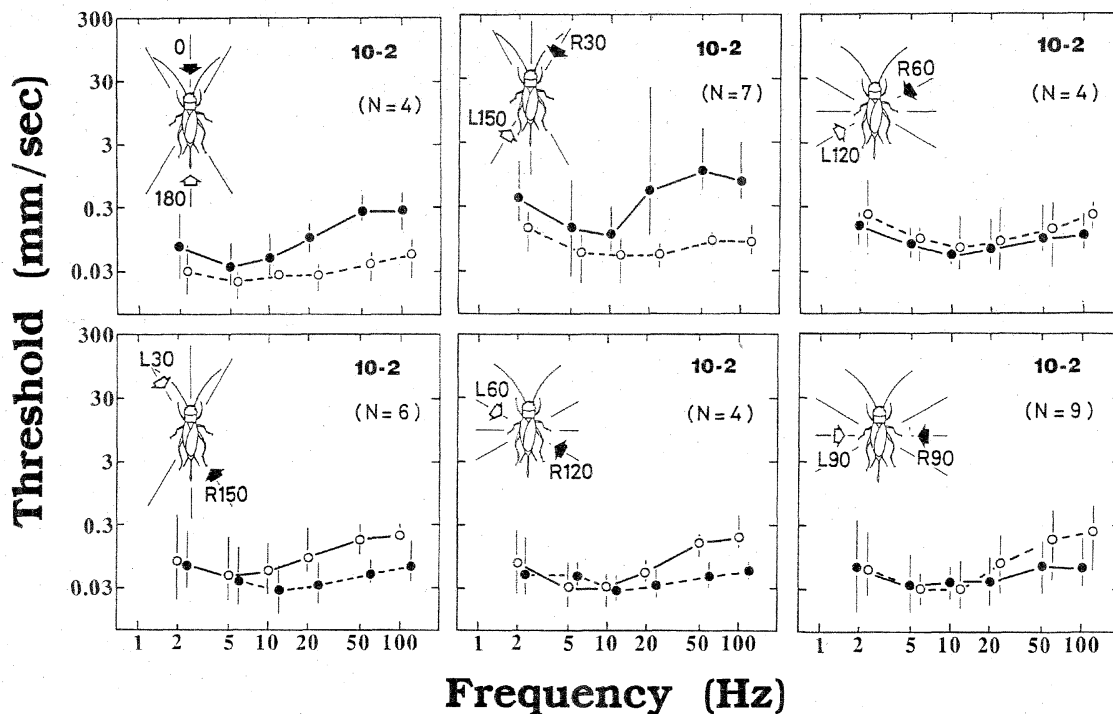


Fig. 10. Threshold curves of GI 10-2 measured by a unidirectional air-current from 12 different directions. Regardless of the stimulus direction, the GI shows the highest sensitivity between 5 and 10 Hz. Thresholds at higher frequencies change more widely than those at lower frequencies. Some of the threshold curves show positive slopes of ca. +20 dB/decade in higher frequencies. See text for details. See legends of Fig. 2 for other explanations.



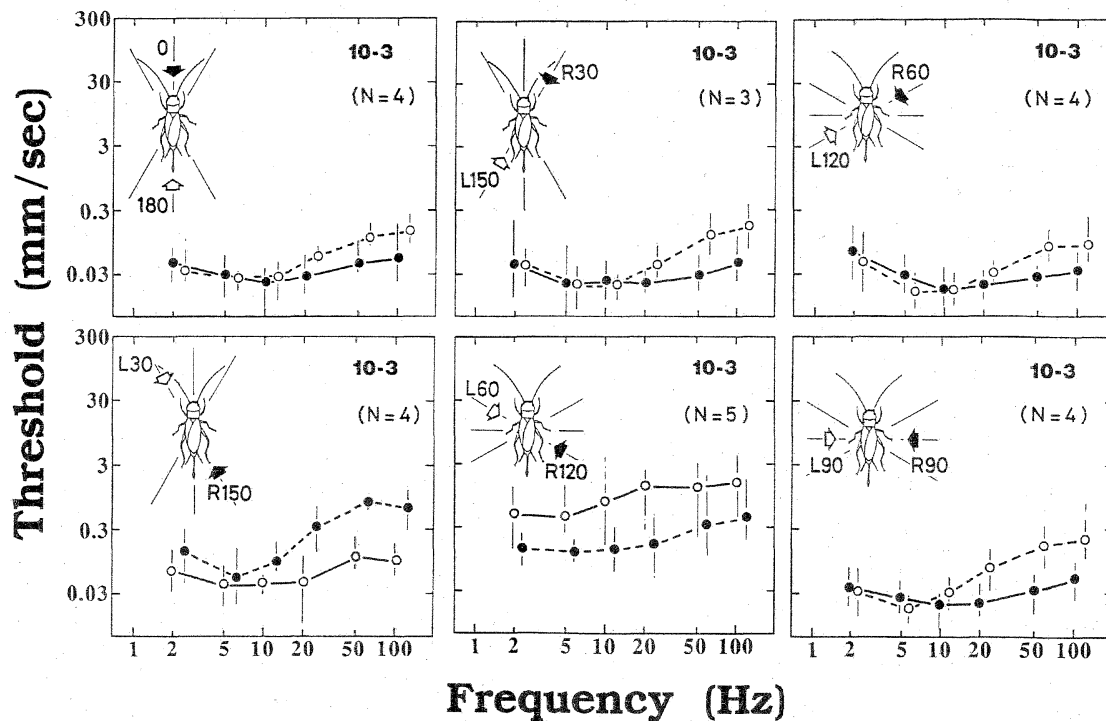


Fig. 11. Threshold curves of GI 10-3 measured by a unidirectional air-current from 12 different directions. Some of the threshold curves show positive slopes of ca. +20 dB/decade at higher frequencies as in GI 10-2. See text for details. See legends of Fig. 2 for other explanations.

characterized as acceleration sensitive GIs because they showed a high-pass characteristic to an alternating air-current stimulus applied parallel to the body axis (Kanou and Shimozawa, 1984). By an examination using a short duration unidirectional air-current (present study), it is revealed that the threshold curves of GIs 8-1 and 9-1 again showed negative slopes regardless of the stimulus direction. They were, therefore, still sensitive to the change of air-current velocity (acceleration) to some extent. However, the slopes of the threshold curves were different from those in the previous study, i.e. the slopes of the threshold curves of GIs 8-1 and 9-1 measured by an alternating air-current were steeper than those to the unidirectional air-current. Threshold curves of GIs 9-2 and 9-3 showed a high pass characteristic to the unidirectional air-current in most cases. However, for air-currents from some directions, threshold curves were parallel with the abscissa in the higher velocity range. Therefore, these GIs show acceleration- or velocity-sensitive characteristics depending upon the stimulus direction.

GIs 10-2 and 10-3 were roughly defined as velocity-sensitive neurons in the previous study using an alternating air-current stimulus applied parallel to the body axis (Kanou and Shimozawa, 1984). In the present study, the threshold curves measured by a unidirectional air-current from 0 or 180 directions (parallel to the body axis) again showed the velocity-sensitive features of these GIs (Figs. 10, 11). However, the threshold curves to air-currents from 0, L60, L90 in GI 10-2 (Fig. 6) and L90, L120, L150, R150 in GI 10-3 (Fig. 7) showed

+20 dB/decade slope above 5 or 10 Hz. The GIs were, therefore, sensitive to the amount of air-molecule displacement in such stimulus conditions. The frequency dependency of the threshold curves of these GIs change according to the change of stimulus direction as those of other GIs do.

#### *Directional sensitivity*

In the present study, directional characteristics of cricket GIs are expressed with two different parameters, i.e. velocity thresholds and response magnitudes. Threshold analysis is very useful for the investigation of information flow in a neural system (Kanou and Shimozawa, 1985; Shimozawa and Kanou 1984a). However, a neural system works not only at threshold level but also at supra-threshold level. Therefore, directional characteristics expressed by spike count function is also a very important clue to understand the role of the neural elements on the elicitation of an escape behavior as well as those expressed by velocity thresholds.

Directional characteristics of GI 8-1 have previously been reported by Tobias and Murphey (1979). In the study, the directionality of GI 8-1 was expressed by the number of impulses in response to a 380 mm/sec air-current stimulus. In the present study, however, it was revealed that the response magnitudes of GI 8-1 were almost saturated or started to decrease at 300 mm/sec air-current (Fig. 3). This suggests that the optimum air-current velocity for GI 8-1 is below 300 mm/sec. Therefore, 380 mm/sec air-current seems too strong for the investigation of response characteristics of

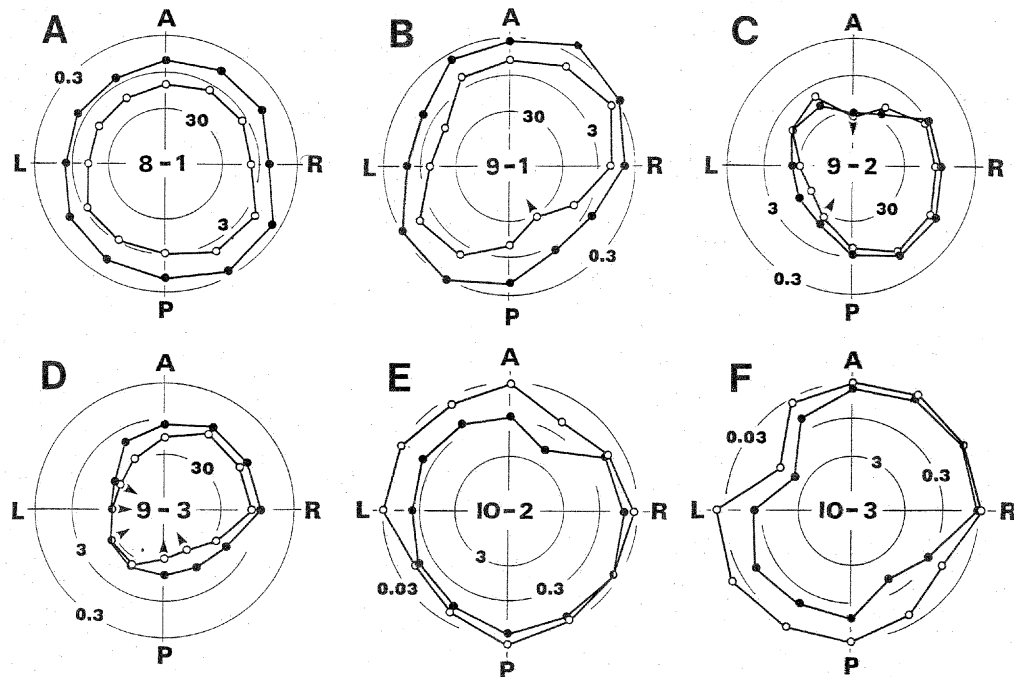


Fig. 12. Directionality of 6 GIs in terms of velocity thresholds plotted on polar coordinates. Air-current velocities (mm/sec) are expressed in logarithmic scale. Filled circles: thresholds at 50 Hz. Open circles: thresholds at 5 Hz. Thresholds with inward arrows were the tentative ones (see legends of Fig. 2). Note that the scales of the air-current velocity in the polar coordinates of GIs 10-2 and 10-3 are 10 times smaller than those of other GIs. A, P, R and L indicate anterior (0), posterior (180), right (R90) and left (L90), respectively. See text for details.

#### GI 8-1.

The present study revealed that the threshold of GI 8-1 is relatively low for air-currents from ipsilateral-rear directions (R120 and R150) (Fig. 12A). Directional characteristics expressed by the response magnitude differ from those expressed by the thresholds because intensity-response curves show different slopes depending upon the direction of the stimulus air-current (Fig. 3). However, in the supra-threshold level, GI 8-1 showed the largest response to air-currents from R60 or R90 in most cases (Fig. 3).

Directional characteristics of GI 9-1 of *Gryllus bimaculatus* have not been investigated before. The results of one earlier study of GI 9-1a (which seems to be homologous with GI 9-1 of *G. bimaculatus*) of *G. campestris* (Kämper, 1984) were different from those of the present study. In the earlier study, GI 9-1a of *G. campestris* showed the largest response to the stimulus applied parallel to the longitudinal axis of the body. However, in the present study, GI 9-1 of *G. bimaculatus* showed high sensitivity to air-currents from ipsilateral-front and contralateral-rear directions. The difference must be due to the difference of stimulation, i.e. a unidirectional air-current was used in the present study but a continuous 30 Hz air oscillation was used in the earlier study.

There are 2 main types of air-motion sensitive filiform hairs on the cricket cerci, i.e. L-hairs and T-hairs (Edwards and Palka, 1974). Lateral-T and anterior-L filiform hairs on ipsilateral cercus (referred to the axon of the GI) make

excitatory innervation on GI 9-1 (Bacon and Murphey, 1984). As filiform hairs are supported by the basal cuticular springs (Kanou *et al.*, 1988, 1989; Shimozaawa and Kanou, 1984b), those hairs move back toward resting positions and probably move beyond the resting position (opposite direction) when unidirectional stimulus air-current stops (rebound motion of filiform hairs). Therefore, the responses of GI 9-1 to the air-current from contralateral-rear directions may partially be due to the rebound motion of the filiform hairs on the ipsilateral cercus.

Intensity-response curves reveal that the directionality of GI 9-1 changes according to the change of the stimulus velocity (Fig. 5) as for GI 8-1. Therefore, it is difficult to express the directional characteristic of GI 9-1 by the response magnitudes. However, it can roughly be understood that GI 9-1 is more sensitive to air-currents from ipsilateral-front directions (0, R30 and R60) than from other directions, even when examined by response magnitudes (Fig. 5). On the other hand, although the thresholds of GI 9-1 to air-currents from contralateral-rear directions were as low as those to air-currents from ipsilateral-front directions (Fig. 12B), response magnitudes to air-currents from contralateral-rear directions reached to maximum at low velocities (Fig. 5). Therefore, GI 9-1 is not suitable for encoding higher velocity air-currents from contralateral-rear directions.

Most of the intensity-response curves of GI 8-1 reached maximal levels around air-current velocities between 100 and 300 mm/sec (Fig. 3), and all of those of GI 9-1 showed peaks

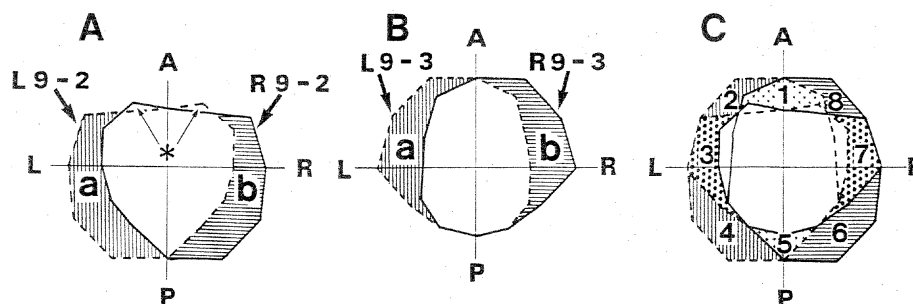


Fig. 13. Receptive fields of right and left GIs 9-2 and/or 9-3 are superimposed in order to illustrate the possible neural mechanism for encoding the air-current direction. A: receptive fields of right and left GI 9-2 (R9-2 and L9-2) are superimposed. a: only L9-2 is activated. b: only R9-2 is activated. Areas marked by asterisk are small exceptions. B: receptive fields of right and left GI 9-3 (R9-3 and L9-3) are superimposed. a: only L9-3 is activated. b: only R9-3 is activated. C: receptive fields of the 4 GIs (R9-2, L9-2, R9-3, L9-3) are superimposed. Directions of air-currents can possibly be coded by the combinational activities of the GIs. 1: R9-3 + L9-3. 2: L9-3. 3: L9-2 + L9-3. 4: L9-2. 5: R9-2 + L9-2. 6: R9-2. 7: R9-2 + R9-3. 8: R9-3.

between 10 and 300 mm/sec regardless of the stimulus direction (Fig. 5). On the other hand, response magnitudes of GIs 9-2 and 9-3 increase in proportion to the logarithm of the peak velocity of air-currents and are not saturated even at the highest velocity (300 mm/sec) used in the present study (Figs. 7, 9). These GIs can possibly encode air-current velocity higher than 300 mm/sec. As the thresholds of GIs 9-2 and 9-3 were higher than those of GIs 8-1 and 9-1, these two groups of GIs (GIs 9-2, 9-3 and GIs 8-1, 9-1) have different dynamic range for velocity encoding. This range fractionation makes the velocity range that the whole cercal sensory system can encode wider.

GIs 9-2 and 9-3 have distinctive directional characteristics (Figs. 12C, D). Their receptive fields are biased to one particular directions, i.e. the main receptive field of GI 9-2 was ipsilateral-rear and that of GI 9-3 is ipsilateral-front. Therefore, the receptive fields of these GIs are symmetrical with respect to the transverse axis of the insect body (front-rear symmetry). In other words, though there are some overlapping, front and rear receptive fields are shared by these GIs. As all cricket GIs investigated in the present study show rather broad receptive fields to a unidirectional air-current stimulus, activity of only one GI is not enough to encode the direction of an air-current. In the cricket nervous system, therefore, direction of an air-current must be encoded by combinational activities of two or more GIs. GIs 9-2 and 9-3 may be ideal candidates for encoding air-current direction in that way. Because, due to their biased receptive fields, combinational activities of the 4 GIs (2 bilateral pairs of GIs 9-2 and 9-3) can well encode the direction of the air-current. For example, although there were small exceptions, activity of the right or left 9-2 can discriminate the air-currents from right-rear or left-rear directions (Fig. 13A). Activity of right or left 9-3 can discriminate the air-currents from right-front or left-front directions (Fig. 13B). Furthermore, the combination of the 4 GIs can roughly encode the air-current from 8 different directions as shown in Figure 13C. Therefore, though I don't exclude the possibility of significance of other

GIs, GIs 9-2 and 9-3 must play an important role for encoding the direction of air-currents.

Directional characteristics of GIs 10-2 and 10-3 are symmetrical with respect to the transverse axis of the insect body (front-rear symmetry) as those of GIs 9-2 and 9-3. In addition to the similarities of the directional characteristics, 2 GIs in each pair show rather similar response characteristics to an air-current stimulus. Both GIs 9-2 and 9-3 show fast adaptation (phasic-tonic response), acceleration dependency (Kanou and Shimozaawa, 1984) and similar intensity-response characteristics (Figs. 7, 9). Both GIs 10-2 and 10-3 show slow adaptation (tonic response), velocity dependency and spontaneous activities (Kanou and Shimozaawa, 1984). The TAG of crickets results from fusion of some embryonic segmental neuromeres during development (Panov, 1966). Both GIs 9-2 and 9-3 belong to the 9th embryonic segment, and both GIs 10-2 and 10-3 belong to the 10th segment (Mendenhall and Murphey, 1974). The common history of embryonic development in the same embryonic segments may underlies the similarities of the response characteristics and the symmetrical receptive fields of the GIs.

Directional characteristics of GIs 10-2 and 10-3 of *Acheta domesticus* have been reported previously (GI 10-2: Miller *et al.*, 1991, GI 10-3: Jacobs and Miller, 1985; Jacobs *et al.*, 1986; Miller *et al.*, 1991). In terms of the most sensitive directions of the GIs, the results of the previous study (Miller *et al.*, 1991) showed good agreement with the result of the present study, although the crickets are from two different genera. However, the sensitivities to air-currents from opposite to the most sensitive directions of *Acheta* largely differ from those in *G. bimaculatus*, i.e. the earlier studies showed that the GIs were inhibited by such air-currents, whereas in the present study, the GI showed neural responses to the air-currents. Although the reason for this difference is not clear, aside from the difference in species, the following may be a possible reason. In the earlier study (Jacobs *et al.*, 1986), ipsps of the GI was detected from neural activities recorded

from soma, and neural activities within 100 msec time-window just after the onset of the air-current stimulus was only counted. On the other hand, in the present study, neural activities were recorded from axons at ventral nerve cord and no ipsp's was recorded. All the action potentials were counted as a response without setting any time-window. Because, cricket neural system can encode the direction of air-current by the action potentials of the GIs to elicit typical directional escape even if the activities contain action potentials caused by the rebound motion of filiform hairs.

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## REFERENCES

- Baba Y, Hirota K, Shimozaawa T, Yamaguchi T (1995) Differing afferent connections of spiking and nonspiking wind-sensitive local interneurons in the terminal abdominal ganglion of the cricket *Gryllus bimaculatus*. J Comp Physiol 176: 17–30
- Bacon JP, Murphey RK (1984) Receptive fields of cricket giant interneurons are related to their dendritic structure. J Physiol 352: 601–623
- Bentley D (1975) Single gene cricket mutations: Effects on behavior, sensilla, sensory neurons, and identified interneurons. Science 187: 760–764
- Bodnar DA (1993) Excitatory influence of wind-sensitive local interneurons on an ascending interneuron in the cricket cercal sensory system. J Comp Physiol A 172: 641–651
- Bodnar DA, Miller JP, Jacobs GA (1991) Anatomy and physiology of identified wind-sensitive local interneurons in the cricket cercal sensory system. J Comp Physiol A 168: 553–564
- Boyan GS, Ball EE (1989) The wind sensitive cercal receptor/giant interneuron system of the Locust, *Locusta migratoria*. II. Physiology of giant interneurons. J Comp Physiol A 165: 511–521
- Boyan GS, Williams JLD, Ball EE (1989) The wind sensitive cercal receptor/giant interneuron system of the Locust, *Locusta migratoria*. I. Anatomy of the system. J Comp Physiol A 165: 495–510
- Camhi JM, Tom W (1978) The escape behavior of the cockroach *Periplaneta americana*. I. Turning response to wind puffs. J Comp Physiol A 128: 193–201
- Camhi JM, Tom W, Volman S (1978) The escape behavior of the cockroach *Periplaneta americana*. II. Detection of natural predators by air displacement. J Comp Physiol A 128: 203–212
- Comer C, Camhi JM (1984) Behavioral compensation for altered cercal position in the cockroach. J Comp Physiol A 155: 31–38
- Daley DL, Camhi JM (1988) Connectivity pattern of the cercus-to-giant interneuron system of the american cockroach. J Neurophysiol 60: 1350–1368
- Dumpe K, Gnatzy W (1977) Cricket combined mechanoreceptors and kicking response. J Comp Physiol A 22: 1–8
- Edwards JS, Palka J (1974) The cerci and abdominal giant fibers of the house cricket, *Acheta domesticus*. I. Anatomy and physiology of normal adults. Proc R Soc Lond B 185: 83–103
- Gnatzy W, Heiße R (1986) Digger wasp against crickets. I. Receptor involved in the antipredator strategies of the prey. Naturwissenschaften 73: S.212
- Gnatzy W, Tautz J (1980) Ultrastructure and mechanical properties of an insect mechanoreceptor: Stimulus-transmitting structure and sensory apparatus of the cercal filiform hairs of *Gryllus*. Cell Tissue Res 213: 441–463
- Gras H, Hörner M (1992) Wind-evoked escape running of the cricket *Gryllus bimaculatus*. I. Behavioral analysis. J Exp Biol 171: 189–214
- Hirota K, Sonoda Y, Baba Y, Yamaguchi T (1993) Distinction in morphology and behavioral role between dorsal and ventral groups of cricket giant interneurons. Zool Sci 10: 705–709
- Jacobs GA, Miller JP (1985) Functional properties of individual neuronal branches isolated in situ by laser photoinactivation. Science 228: 344–346
- Jacobs GA, Miller JP, Murphey RK (1986) Integrative mechanisms controlling directional sensitivity of an identified sensory interneuron. J Neurosci 6: 2298–2311
- Kämpfer G (1984) Abdominal ascending interneurons in crickets: responses to sound at the 30-Hz calling-song frequency. J Comp Physiol A 155: 507–520
- Kanou M (1991) Threshold and directional sensitivity of air-current-sensitive giant interneurons of a cricket. Experientia 47: 447–448
- Kanou M, Ohshima M, Inoue J (1995) Analysis of air-current evoked escape behavior of the cricket. Zool Sci 12 (Suppl): 100. (Proceedings of the 66th annual meeting of the Zoological Society of Japan)
- Kanou M, Osawa T, Shimozaawa T (1988) Ecdysial growth of the filiform hairs and sensitivity of the cercal sensory system of the cricket *Gryllus bimaculatus*. J Comp Physiol A 162: 573–579
- Kanou M, Osawa T, Shimozaawa T (1989) Mechanical polarization in the air-current sensory hair of a cricket. Experientia 45: 1082–1083
- Kanou M, Shimozaawa T (1984) A threshold analysis of cricket cercal interneurons by an alternating air-current stimulus. J Comp Physiol A 154: 357–365
- Kanou M, Shimozaawa T (1985) Responses of cricket leg motoneurons to air-current stimuli: velocity dependent inhibition and acceleration dependent excitation. Zool Sci 2: 629–639
- Kobashi M, Yamaguchi T (1984) Local non-spiking interneurons in the cercus-to-giant interneuron system of crickets. Naturwissenschaften 71: 154–155
- Liebethal E, Uhlmann O, Camhi JM (1994) Critical parameters of the spike trains in a cell assembly: coding of turn direction by the giant interneurons of the cockroach. J Comp Physiol A 174: 281–296
- Levine RB, Murphey RK (1980) Pre- and postsynaptic inhibition of identified giant interneurons in the cricket (*Acheta domesticus*). J Comp Physiol A 135: 269–282
- Matsumoto SG, Murphey RK (1977) The cercus-to-giant interneuron system of crickets. IV. Patterns of connectivity between receptors and the medial giant interneuron. J Comp Physiol A 119: 319–330
- Miller JP, Jacobs GA, Theunissen FE (1991) Representation of sensory information in the cricket cercal sensory system. I. Response properties of the primary interneurons. J Neurophysiol 66: 1680–1689
- Mendenhall B, Murphey RK (1974) The morphology of cricket giant interneurons. J Neurobiol 5: 565–580
- Murphey RK, Palka J, Hustert R (1977) The cercus-to-giant interneuron system of crickets. II. Response characteristics of two giant interneurons. J Comp Physiol A 119: 285–300
- Panov AA (1966) Correlations in the ontogenetic development of the central nervous system in the house cricket *Gryllus domesticus* and the mole cricket *Gryllotalpa gryllotalpa* L. (Orthoptera, Grylloidea). Entomol Rev 45: 179–185
- Shen J-X (1983) The cercus-to-giant interneuron system in the bushcricket *Tettigonia cantans*: morphology and response to low-frequency sound. J Comp Physiol A 151: 449–459
- Shimozaawa T, Kanou M (1984a) Varieties of filiform hairs: range fractionation by sensory afferents and cercal interneurons of a

- cricket. J Comp Physiol A 155: 485–493
- Shimozawa T, Kanou M (1984b) The aerodynamics and sensory physiology of range fractionation in the cercal filiform sensilla of the cricket *Gryllus bimaculatus*. J Comp Physiol A 155: 495–505
- Tobias M, Murphey RK (1979) The response of cercal receptors and identified interneurons in the cricket (*Gryllus domesticus*) to airstream. J Comp Physiol A 129: 51–59
- Vardi N, Camhi JM (1982) Functional recovery from lesions in the escape system of the cockroach. II. Physiological recovery of the giant interneurons. J Comp Physiol A 146: 299–309
- Westin J, Langberg JJ, Camhi JM (1977) Responses of giant interneurons of the cockroach *Periplaneta americana* to wind puffs of different directions and velocities. J Comp Physiol A 121: 307–324