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Organization of Receptive Fields of Cricket Giant Interneurons Revealed by Cercal Ablations

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ABSTRACT—In order to determine the contribution of each cercus and its receptors in organizing the receptive field of four air-motion sensitive giant interneurons (GIs 8–1, 9–1, 9–2 and 9–3) of the cricket *Gryllus bimaculatus*, effects of removing the ipsilateral or contralateral cercus (referred to the side of the axons) on specific parameters of the wind-evoked responses of these neurons were investigated. All 4 GIs received only excitatory inputs from a group of filiform hairs on the ipsilateral cercus. In addition to the ipsilateral excitatory inputs, GIs 8–1 and 9–1 received weak excitatory and strong inhibitory inputs from a group of filiform hairs on the contralateral cercus. GI 9–2 received only inhibitory inputs from filiform hairs on the contralateral cercus. GI 9–3 received excitatory inputs from filiform hairs on the contralateral cercus and no inhibitory input was confirmed. In addition to such simple excitatory and inhibitory connections, the rebound motion of cercal filiform hairs had some role in organizing the receptive fields of GIs 9–2 and 9–3. Furthermore, the possibility of using a rebound depolarization of the membrane potential for mediating the long latency response in GIs 8–1 and 9–2 will be discussed.

INTRODUCTION

A large number of wind sensitive filiform hairs exist on the cerci of crickets. They are responsible for a wind-evoked escape behavior of the insects (Bentley, 1975; Gras and Hörner, 1992; Kanou *et al*., 1995). As giant interneurons (GIs) in the ventral nerve cord of crickets integrate the wind information from those cercal filiform hairs (Edwards and Palka, 1974; Tobias and Murphey, 1979; Kanou and Shimozawa, 1984), they are supposed to play a significant role in activating leg motoneurons (Kanou and Shimozawa, 1985) and mediating a wind-evoked escape behavior. Such wind-evoked escape behavior is highly directional, i.e. the insects almost always turn away from the stimulus source (Gras and Hörner, 1992; Kanou *et al*., 1995). Directional characteristics of GIs (Kanou, 1991, 1996; Levine and Murphey, 1980; Tobias and Murphey, 1979) must underlie such directional behaviors.

As information from cercal filiform hairs are main inputs to GIs, many studies have been focused on the physiological and mechanical features of the hair sensilla (Edwards and Palka, 1974; Gnatzy and Tautz, 1980; Kanou *et al*., 1988, 1989; Shimozawa and Kanou, 1984a, b). Traditionally, cercal filiform hairs have been classified into 2 major populations on the bases of their preferential plane for vibration; i.e. L-hairs (preferentially vibrate parallel to the longitudinal axis of a cercus) and T-hairs (preferentially vibrate parallel to the transverse axis of a cercus). They are further divided into two subpopulations from their directional sensitivities, i.e. anterior and posterior L-hairs, and lateral and medial T-hairs (Bacon and Murphey, 1984; Tobias and Murphey, 1979). Therefore, most studies dealing with the directional sensitivities of cricket GIs have been focused on the innervation with the 4 types of cercal filiform hairs (Bodnar *et al*., 1991; Jacobs *et al*., 1986; Levine and Murphey, 1980; Tobias and Murphey, 1979). However, it has been reported that obliquely oriented hairs exist in *Acheta domesticus* (Walthall and Murphey, 1986; Shepherd *et al*., 1988), and a recent study proved that the hairs can be divided into at least 8 subclasses (Landolfa and Jacobs, 1995). Although such detailed classification of cercal filiform hairs in *Gryllus bimaculatus* has not been as well confirmed as in *Acheta*, it has been reported that *Gryllus* is also equipped with D (diagonal) hairs other than L- and T-hairs (Gnatzy and Tautz, 1980).

Due to the variety of preferential directions in cercal filiform hairs, the receptive field of each GI in *Gryllus bimaculatus* must be complicated. The aim of the present study was to explore the receptive field organization of each GI in *Gryllus bimaculatus* by investigating the directional sensitivity of GIs after the unilateral cercal ablations. This process is essential for understanding the pattern of connection between a paticular GI and cercal filiform hairs with specific directionalities. More-

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over, such detailed investigation is also essential for the basis of more complicated studies of the neural network, e.g. an investigation of plastic natures (Matsuura and Kanou, 1998).

MATERIALS AND METHODS

Animals

Adult female crickets (*Gryllus bimaculatus*) reared in our laboratory were used. They were 1–2 weeks old after imaginal molt. The temperature of the culture room was kept at 28–30°C and the LD cycle was 14 : 10. Details were the same as in the previous paper (Kanou, 1996).

Air current stimulation

In the course of neurophysiological experiments, a unidirectional air current stimulus was given to a cricket in a wind-tunnel consisting of a pair of push-pull driven loud speakers (Kanou and Shimozawa, 1984). An electrical signal of a half cycle cosine wave was fed to the speakers so as to make a unidirectional air current in the tunnel. The frequency of the air-motion (although a whole cycle of cosine wave was not used, we use a term of "frequency" in order to express the rate of air-molecule displacement; ref. Kanou, 1996) was fixed at 50 Hz in all the experiments. The stimulus duration was, therefore, 10 msec (the time of a half cycle of 50 Hz). Experimental setups and stimulating methods are the same as in the previous studies (Kanou, 1991, 1996).

Air current velocities employed in the present experiments ranged from 0.095 mm/sec to 300 mm/sec. As the wind-tunnel could be rotated around the specimen in the horizontal plane, the direction of a stimulus air current could be changed even in the course of intracellular recordings. In the present study, air current stimuli were presented from 12 different directions (30° intervals) (ref. Fig. 1A). At each stimulus direction, 10 successive air current stimuli with a particular peak velocity were applied to an insect at 5-second intervals. An averaged response magnitude (number of action potentials) and an averaged response latency of the10 trials were calculated. The same measuring procedure was repeated at every 10 dB of the peak velocity of the stimulus air current. The averaged values were again averaged over the number of animals used. In order to avoid habituation, a weak stimulus was applied first, then the intensity (peak velocity of a stimulus air current) was increased stepwise.

Neural activity recordings

Activities of GIs were intracellularly recorded from axons in the right side connective at the point where it just left the 5th (terminal) abdominal ganglion. Recorded signals were fed into a microelectorode amplifier (Nihon Kohden; MEZ-8201) and displayed on a CRT (Tektronix; R5111A). Neural responses were stored on a magnetic tape (Maxell; DAT R-120DM) by a data recorder (TEAC; RD-111T) for the off-line analysis.

After the physiological investigation, Lucifer Yellow CH, which was placed in glass microelectrode beforehand, was iontophoretically injected into GIs with ±10 nA square pulses for more than 5 min for morphological identification. After the dye injection, the specimen was incubated in a refrigerator for about 1 hr before the observation.

Velocity thresholds (the lowest peak velocity of an air current that could elicit one action potential on a GI) were obtained by an interpolation of the intensity response curves. For each response property such as velocity threshold, response magnitude or response latency, significance of difference between the intact and the ablated animals were statistically examined with the t-test.

Ablation experiments

A cercus of a cricket was removed from the stump with a sharp razor blade. Special attention was paid so as to remove all the mechanoreceptive cercal filiform hairs at the basal part of a cercus.

Neurophysiological experiments on the unilaterally cercal ablated animals were carried out within 1 day (24 ± 5 hr) after the treatment. As the ventral nerve cords which contain GIs' axons are on the contralateral side of their somata, we defined "ipsilateral" or "contralateral" referred to the side of axons. For example, "ipsilateral cercus" means the cercus which is at the same side of the axon of a GI. As we made neural recordings from a right side connective, right cercus ablated crickets were called CCI (contralateral cercus intact) animals. In the same way, contralateral cercus ablated crickets were called ICI (ipsilateral cercus intact) animals. Normal intact crickets were called BCI (both cerci intact) animals.

RESULTS

An intensity-response relation of each GI was measured by using a unidirectional air current stimulus. Results were compared to those from animals with both cerci intact (BCI) (Kanou, 1996) in order to specify the role of ipsilateral and contralateral cercal inputs in organizing the receptive field of each GI (Fig. 1). Difference in response magnitudes between BCI and unilaterally cercal ablated animals (ICI or CCI) will show the amount of excitatory or inhibitory inputs from filiform hairs on the ablated cercus. Response latencies of GIs were measured in BCI animals and were also compared to those measured in either ICI or CCI animals. These were the clues to estimate the number of synaptic connections and to reveal other response properties of a GI such as "hair rebound".

Response properties of GI 8–1 in ICI animals

GI 8–1 in ICI animals showed excitatory responses regardless of the directions of a stimulus air current (Fig. 2). This suggests that all types of filiform hairs on the ipsilateral cercus have excitatory connections with the GI regardless of their directional sensitivities (Fig. 4, right). When a stimulus air current was applied from R60 direction, response magnitudes of GI 8–1 in ICI animals were significantly smaller than those in BCI animals ($P < 0.05$; Fig. 2). This suggests that the contralateral filiform hairs sensitive to the R60 air currents have excitatory connection with GI 8–1 (Fig. 4, left). On the other hand, response magnitudes of GI 8–1 in ICI animals were larger than those in BCI animals when the air currents were applied from L90, L60, L30, 0, R30, R120, R150, 180 and L150 directions (differences were statistically significant other than L60: Fig. 2). This suggests that the contralateral filiform hairs activated by those air currents have inhibitory connection with the GI (Fig. 4, left). Thresholds for the inhibition seemed to be higher than those for excitations in most cases, because most of the thresholds were not largely affected by the contralateral cercal ablation (Fig. 1A).

In BCI animals, response latencies of GI 8–1 changed depending upon both intensity and direction of a stimulus air current (Fig. 3). In most cases, response latencies of the GI became shorter with the increase of stimulus velocity. Response latencies of the GI were relatively long to the air currents from the contralateral-front directions. In ICI animals, some of the response latencies of GI 8–1 to the air currents from 0, R150 and L90 directions were significantly shorter than those in BCI animals ($P < 0.05$). As the contralateral filiform

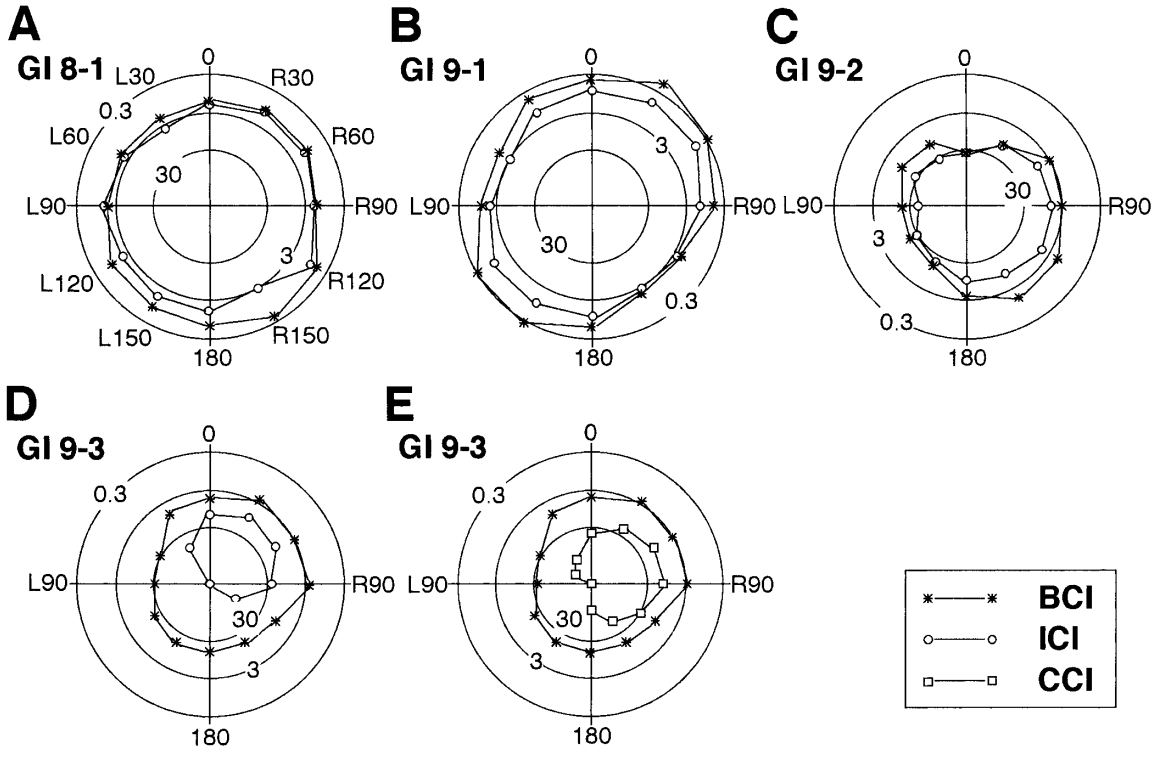

Fig. 1A–E. Receptive fields of 4 GIs running their axons in the right side ventral nerve cord. Velocity thresholds (mm/sec) of the GIs to an air current from 12 different directions were plotted on the polar coordinates. (**A**) GI 8–1. (**B**) GI 9–1. (**C**) GI 9–2. (**D**) and (**E**) GI 9–3. Crosses: BCI animals (data from Kanou, 1996). Open circles: ICI animals within 1 day after the cercal ablation. Open squares: CCI animals within 1 day after the cercal ablation. Responses of GIs 8–1, 9–1 and 9–2 in CCI animals were very small or nothing at all. Note that the most sensitive directions of all the GIs in unilaterally cercal ablated animals were almost the same with those in BCI animals.

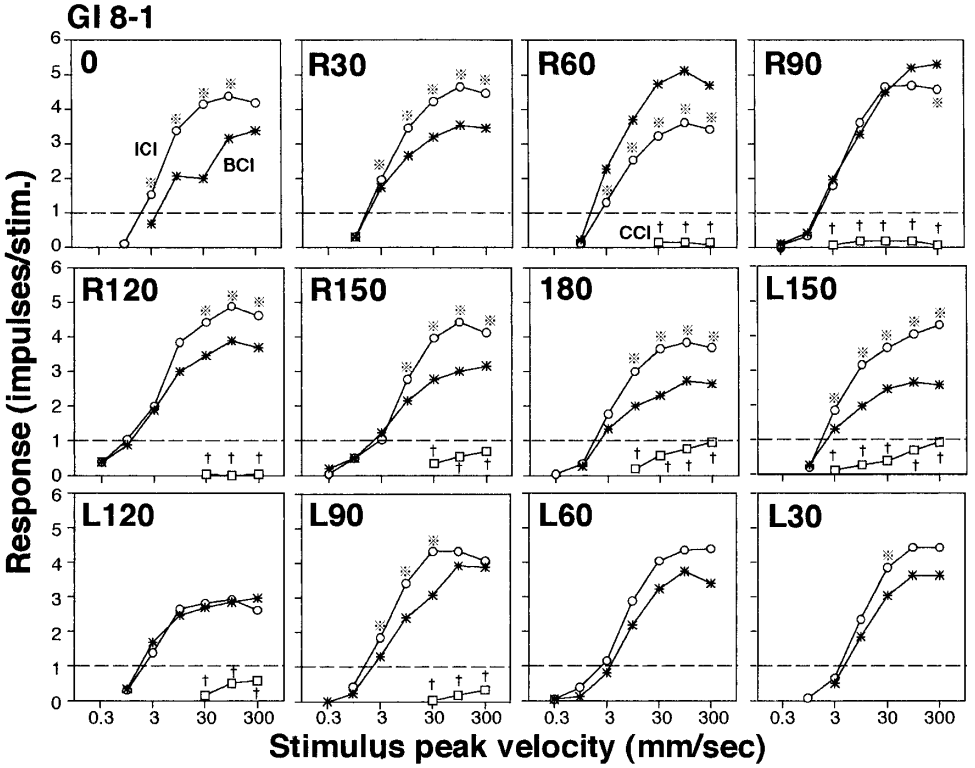

Fig. 2. Intensity-response curves of GI 8–1 measured with a unidirectional air current from 12 different directions. Response magnitudes (averaged number of action potentials elicited per stimulus) were plotted against the peak velocity of the stimulus air current. Symbols are the same as in Fig. 1. ※: statistically significant difference was confirmed between BCI and ICI animals ($P < 0.05$). †: statistically significant difference was confirmed between BCI and CCI animals ($P < 0.05$). N (number of animals) = 10 for open circles. N = 5 for open squares.

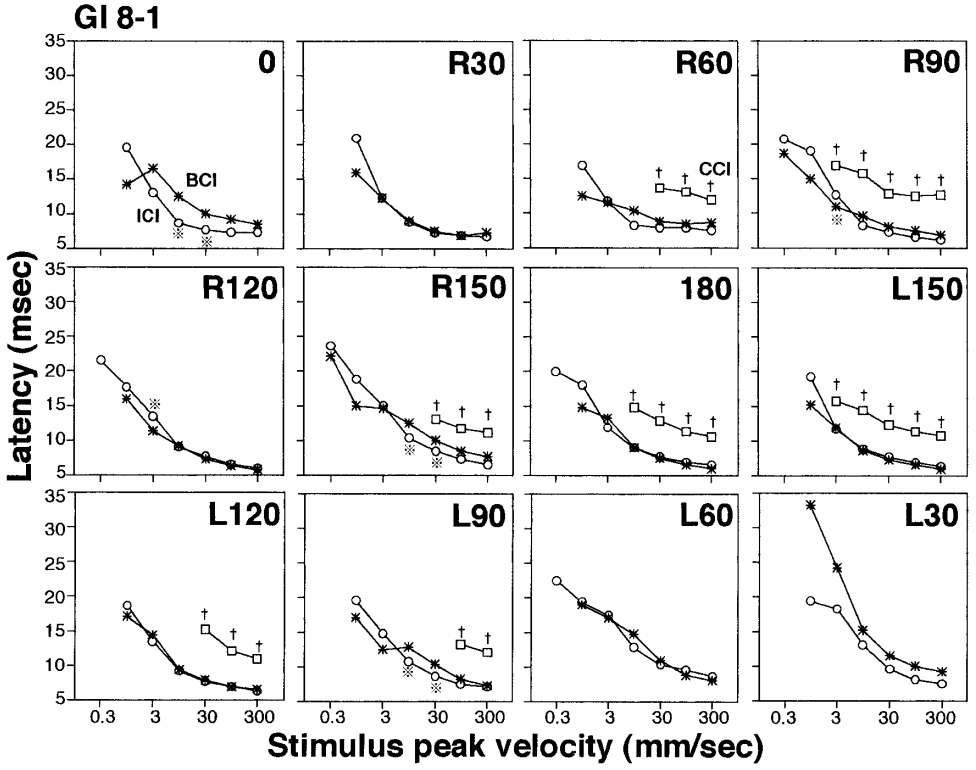

Fig. 3. Response latencies of GI 8–1. Symbols are the same as in Figs. 1 and 2. Response latencies of the GI became shorter with the increase of stimulus velocity in all the cases. All of the response latencies in CCI animals were significantly longer than those in BCI animals. Sample numbers are the same as in Fig. 2.

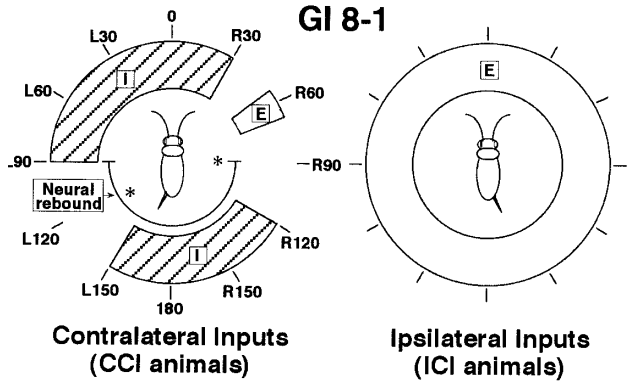

Fig. 4. Stimulus directions of excitatory and inhibitory inputs on GI 8–1 in ICI (ipsilateral inputs) and CCI (contralateral inputs) animals. Open area with "E": excitatory inputs. Hatched area with "I": inhibitory inputs. When an excitatory or inhibitory effect was ascertained in only one stimulus direction, the direction was shown a little bit wider in order to indicate the direction clearly, e.g. R60 in the left figure. Filiform hairs on the ipsilateral cercus have excitatory connection with GI 8–1 regardless of their directional sensitivities. Among filiform hairs on the contralateral cercus, only those sensitive to the air current from R60 direction have excitatory connection with the GI, and most of the other contralateral filiform hairs have inhibitory connections. Neural rebound: stimulus directions in which GI 8–1 showed responses that seemed to be caused by the rebound depolarization. Asterisk: neural depolarization was assumed only from long response latencies.

hairs sensitive to air currents from these directions have an inhibitory connection with the GI (Fig. 4, left), the inhibition from those hairs reaches GI 8–1 faster than or almost simultaneously with the excitatory inputs from the filiform hairs on the ipsilateral cercus, and delays the GI's membrane potential reaching threshold.

Response properties of GI 8–1 in CCI animals

GI 8–1 in CCI animals showed very weak responses only when the stimulus air currents were applied clockwise from R60 to L90 directions (Fig. 2). It seemed somewhat paradoxical with the observed differences in the response magnitudes between ICI and BCI animals (Fig. 2) because the results suggested that contralateral filiform hairs sensitive to the air currents from R120, R150, 180, L150 and L90 had inhibitory connection with the GI (responses to R60 air currents were caused by the excitatory inputs as mentioned before; Fig. 4). We assumed that the very weak responses (except R60) were caused by the rebound depolarization released from inhibitory hyperpolarizations (Fig. 4; Neural rebound), because latencies of those responses were significantly longer than those in BCI animals (longer than 10 msec, Fig. 3; see discussion).

Response properties of GI 9–1 in ICI animals

Like GI 8–1, GI 9–1 in ICI animals responded to air currents regardless of the stimulus direction (Fig. 5). It suggests that all types of filiform hairs on the ipsilateral cercus have

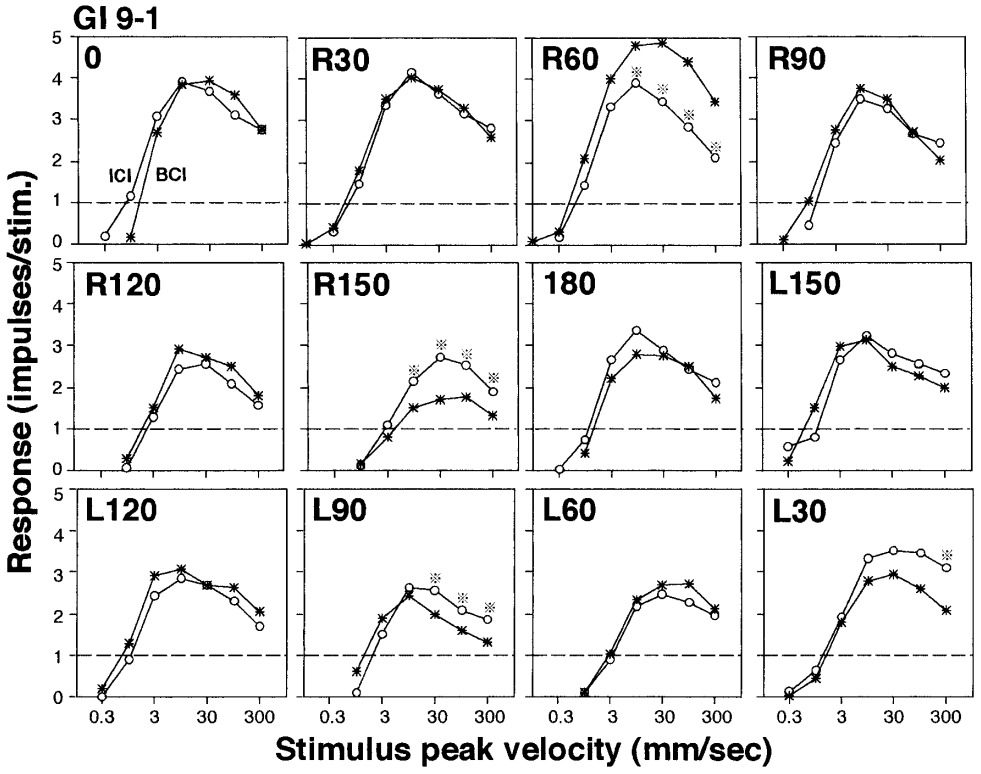

Fig. 5. Intensity-response curves of GI 9–1. Symbols are the same as in Figs. 1 and 2. In ICI animals, response magnitudes were larger than those in BCI animals when the air currents were applied from R150, L90 and L30 directions. On the other hand, response magnitudes in ICI animals were significantly smaller than those in BCI animals when the air current was applied from the R60 direction. Note that GI 9–1 in CCI animals showed no response regardless of the stimulus direction. N ≥ 9 for open circles.

excitatory connection with GI 9–1 regardless of their directional sensitivities (Fig. 7, right). Although a receptive field of the GI in ICI animals was slightly smaller than those in BCI animals, those shapes were very similar to each other (Fig. 1B).

Response magnitudes of GI 9–1 in ICI animals were significantly larger than those in BCI animals ($P < 0.05$) when the stimulus air currents were applied from R150, L30 and L90 directions (Fig. 5). It suggests that the contralateral filiform hairs activated by such air currents have inhibitory connection with the GI (Fig. 7, left). When the air currents were applied from the R150 and L90 directions, response latencies of the GI in ICI animals were significantly shorter than those in BCI animals ($P < 0.05$; Fig. 6). This suggests that the inhibitory information reaches the GI faster than or almost simultaneously with the excitatory ones and delays the GI's membrane potential reaching threshold.

Response latency of GI 9–1 in BCI animals became shorter with the increase of stimulus velocity in most cases, and was the shortest for air currents from mainly ipsilateral-front directions, i.e. L30, 0, R30, R60, R90 and R120 (Fig. 6). In ICI animals, response latencies of GI 9–1 to the unidirectional air currents were almost the same with those in BCI animals regardless of the stimulus direction except for the R150 and L90 directions as mentioned above (Fig. 6).

Response properties of GI 9–1 in CCI animals

In CCI animals, no response was evoked in GI 9–1 regardless of the direction of the stimulus air current (Fig. 5). It may suggest that no filiform hairs on the contralateral cercus have excitatory connection with the GI, i.e. excitatory inputs seemed to be restricted from filiform hairs only on the ipsilateral cercus. However, response magnitudes of GI 9–1 in ICI animals were significantly smaller than those in BCI animals when the air current was applied from R60 direction (Fig. 5). It suggests that filiform hairs on the contralateral cercus activated by the R60 air current provide excitatory but subthreshold inputs to GI 9–1 (Fig. 7, left; see discussion).

Response properties of GI 9–2 in ICI animals

Although velocity thresholds of GI 9–2 in ICI animals were relatively higher than those in BCI animals, especially when the air currents were applied from ipsilateral-rear (R120, R150 and 180) and contralateral-front (L30, L60 and L90) directions (Fig. 1C), the GI was still sensitive to the air currents from ipsilateral-rear directions like in BCI animals. Even after the unilateral cercal ablation, therefore, the preferred direction of the GI was kept constant.

GI 9–2 in ICI animals responded to air currents regardless of the stimulus direction (Fig. 8). It may suggest that all types of filiform hairs on the ipsilateral cercus have excitatory connection with GI 9–2 regardless of their directional sensi-

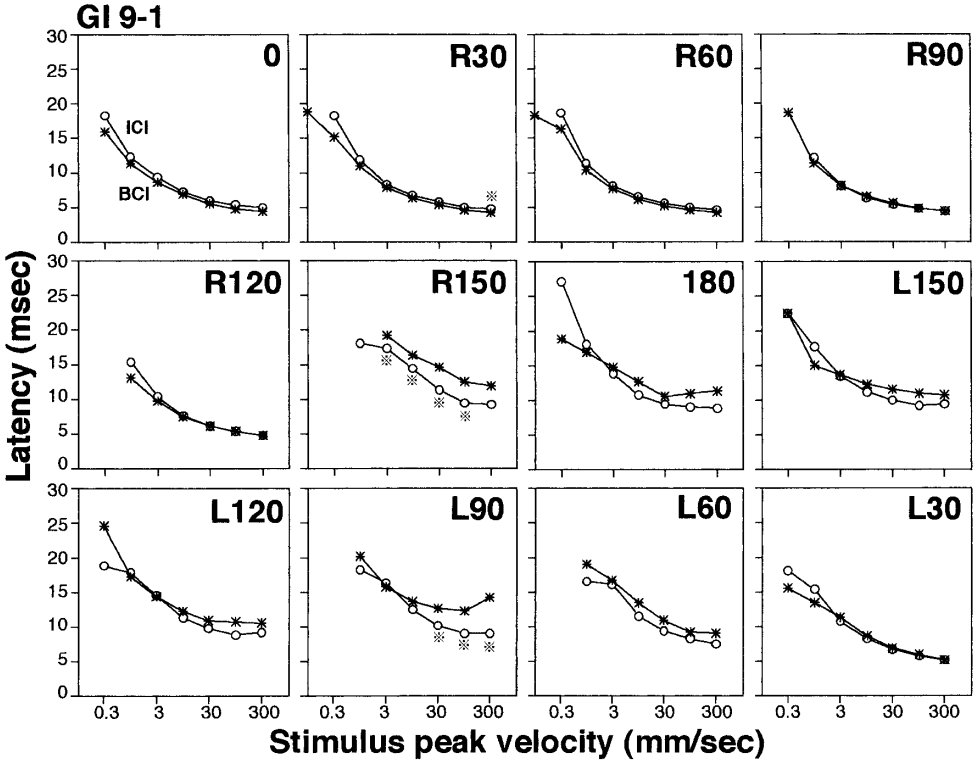

Fig. 6. Response latencies of GI 9–1. Symbols are the same as in Figs. 1 and 2. Response latencies of GI 9–1 became shorter with the increase of stimulus velocity in both BCI and ICI animals. In ICI animals, response latencies to the air currents from R150 and L90 directions were shorter than those in BCI animals. It suggests that inhibitory information from contralateral filiform hairs sensitive to those air currents reach the GI faster than or almost simultaneously with excitatory ones. Sample numbers are the same as in Fig. 5.

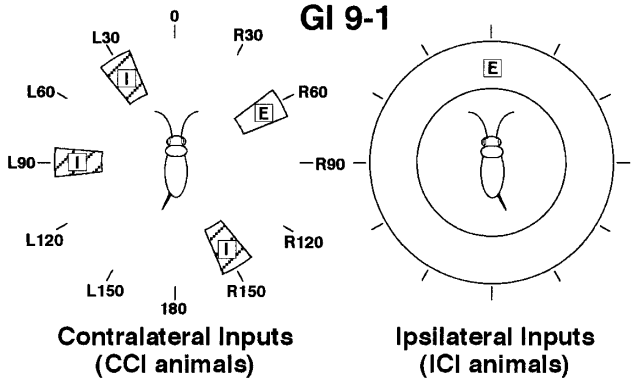

Fig. 7. Stimulus directions of excitatory and inhibitory inputs on GI 9–1. Like in GI 8–1, filiform hairs on the ipsilateral cercus have excitatory connection with the GI regardless of their directional sensitivities. Among filiform hairs on the contralateral cercus, only those sensitive to the air current from R60 direction have excitatory connection with the GI. Filiform hairs on the contralateral cercus sensitive to the air currents from R150, L90 and L30 directions showed inhibitory connections with GI 9–1. Refer to Fig. 4 for abbreviations.

tivities. However, when the air currents were applied from L90, L60 and L30 directions, response magnitudes were relatively smaller and response latencies were considerably longer (longer than 15 msec) than for other directions (Figs. 8, 9). A sensory afferent of a cercal filiform hair fires when the hair is deflected in one direction. Although the afferent shows short latency responses when the stimulus air current is applied from the preferred direction, the afferent shows long latency responses which correspond to the falling phase of the unidirectional stimulus when the air current is applied from the opposite direction (Tobias and Murphey, 1979). As GI 9–2 showed short latency responses when the stimulus was applied from R90, R120 and R150 directions, ipsilateral filiform hairs sensitive to such air currents must deliver excitatory inputs to the GI. Those afferents must evoke long latency responses to the unidirectional air current stimulus from L90, L60 and L30 directions (180° opposite to R90, R120 and R150, respectively), and resulted in the long latency responses on GI 9–2. We assumed that the long latency responses were caused by the rebound motion of filiform hairs (Fig. 10, right; Hair rebound; see discussion).

In the suprathreshold domain, response magnitudes of GI 9–2 in ICI animals were significantly larger than those in BCI animals when the air currents were applied from L30, 0, R30, R60, R150, 180, L150 and L120 directions (Fig. 8). It suggests that filiform hairs on the contralateral cercus sensitive to those air currents have inhibitory connection with GI 9–2 (Fig. 10, left). Among the cases, response latencies to the air currents from R150 and L120 directions were significantly shorter than those in BCI animals in most stimulus velocities ($P < 0.05$; Fig. 9). It suggests that the inhibition from filiform hairs on the contralateral cercus encoding those air currents

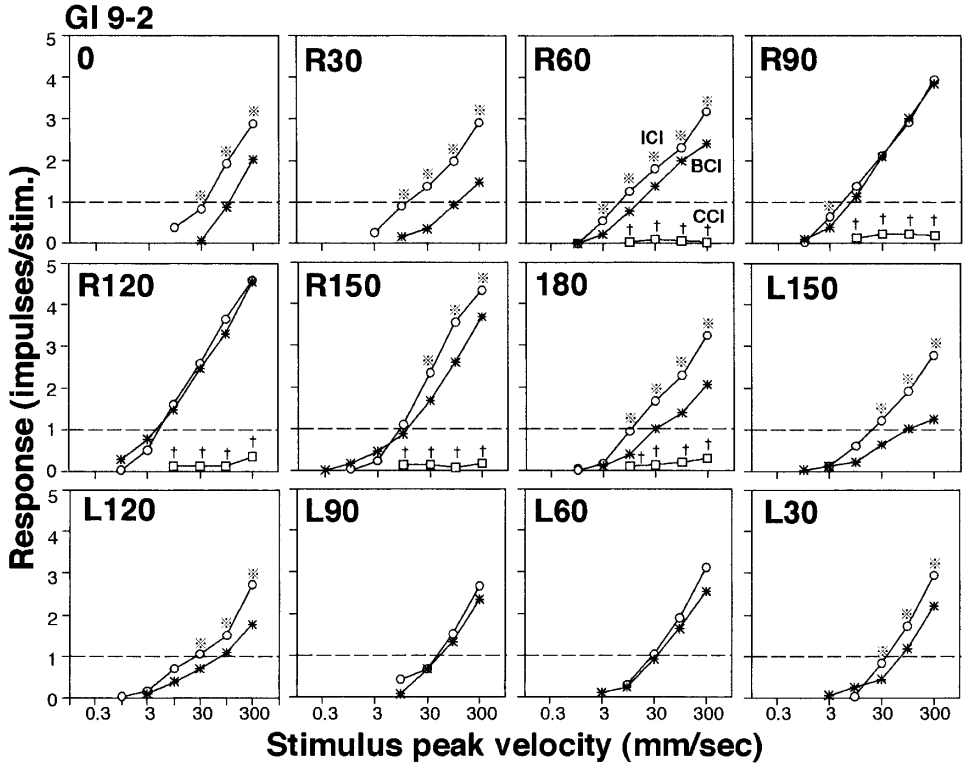

Fig. 8. Intensity-response curves of GI 9–2. Symbols are the same as in Figs. 1 and 2. Most of the response magnitudes in ICI animals were significantly larger than those in BCI animals. Note the very poor responses in CCI animals. N = 10 for open circles. N = 8 for open squares.

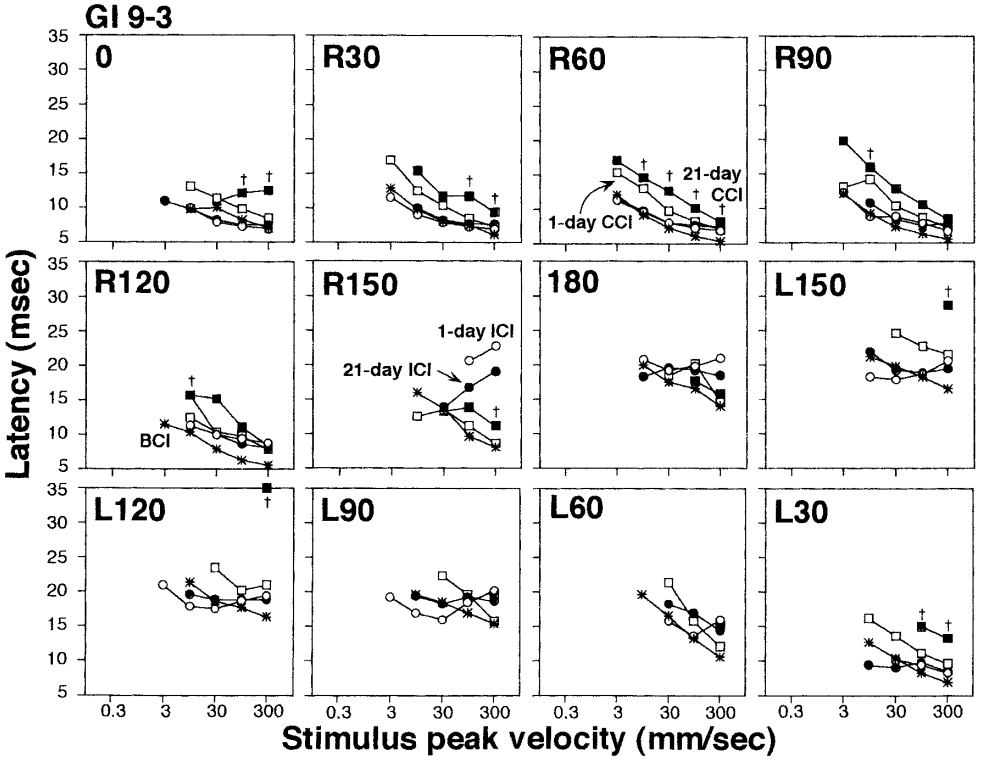

Fig. 9. Response latencies of GI 9–2. Symbols are the same as in Figs. 1 and 2. All of the response latencies in CCI animals were significantly different from those in BCI animals. Note the long response latencies in both BCI and ICI animals to the air currents applied from L90, L60 and L30 directions. Sample numbers are the same as in Fig. 8.

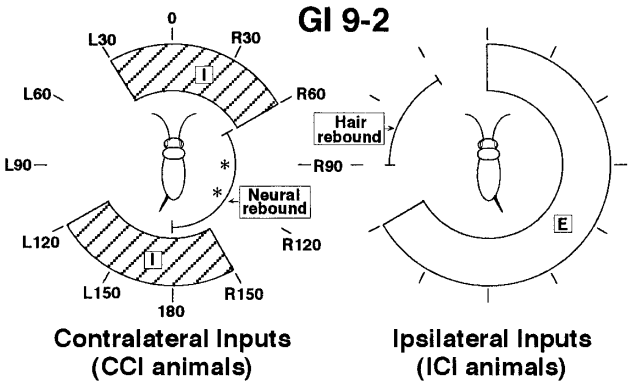

Fig. 10. Stimulus directions of excitatory and inhibitory inputs on GI 9–2. Most of the filiform hairs on the ipsilateral cercus showed excitatory connection with the GI. On the other hand, most of the filiform hairs on the contralateral cercus showed inhibitory connection with the GI. Hair rebound: responses were supposed to be caused by the rebound motion of cercal filiform hairs. See text for details. Refer to Fig. 4 for abbreviations.

reaches the GI earlier than or almost simultaneously with excitatory ones and delays the GI reaching threshold (see discussion).

Response properties of GI 9–2 in CCI animals

GI 9–2 in CCI animals showed responses only when the air currents were applied from R60, R90, R120, R150 and 180 directions (Fig. 8). These response magnitudes were considerably smaller than those in BCI animals. Moreover, latencies of all such responses were quite long (more than 10 msec; Fig. 9). As was suggested in ICI animals, filiform hairs on the contralateral cercus encoding the R60, R150 and 180 air currents have inhibitory connection with GI 9–2. Therefore, it is likely that the weak responses were caused by the rebound depolarization from the inhibition (Fig. 10, left) as was observed in GI 8–1 (see discussion).

Response properties of GI 9–3 in ICI animals

Although GI 9–3 in ICI animals showed responses to unidirectional air currents regardless of the stimulus direction, response magnitudes to the air currents applied from R150, 180, L150, L120, L90 and L60 directions were very small (Fig. 11), and the latencies of such responses were quite long (more than 15 msec; Fig. 12). On the other hand, response magnitudes to the air currents applied from 180° opposite directions (i.e. L30, 0, R30, R60, R90 and R120, respectively) were relatively large (Fig. 11) and the latencies of those responses were relatively short (Fig. 12). Therefore, the responses to the air currents from R150, 180, L150, L120, L90 and L60 directions were likely to have been caused by the rebound motion of cercal filiform hairs because of the same reason as in GI 9–2 (Fig. 13, right).

The receptive field of GI 9–3 in ICI animals was much smaller than that in BCI animals (Fig. 1D). However, the most preferred direction of the GI was remained constant even after the contralateral cercal ablation, i.e. ipsilateral-front direction.

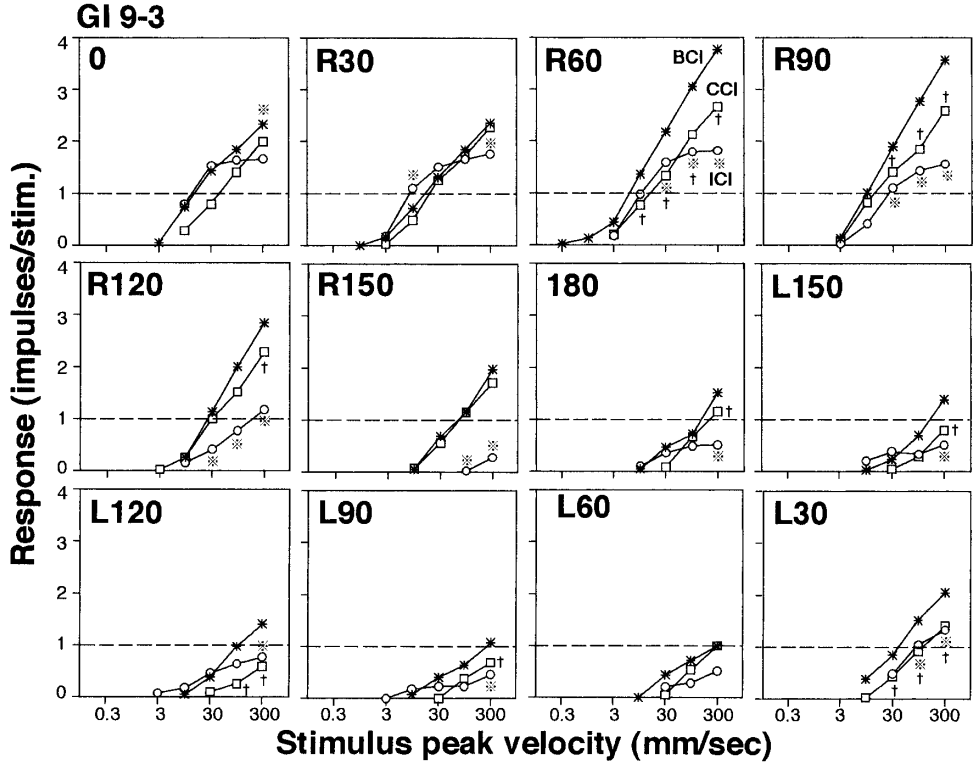

Fig. 11. Intensity-response curves of GI 9–3. Symbols are the same as in Figs. 1 and 2. Unlike other GIs, GI 9–3 in CCI animals showed relatively large response magnitudes. N ≥ 10 for all.

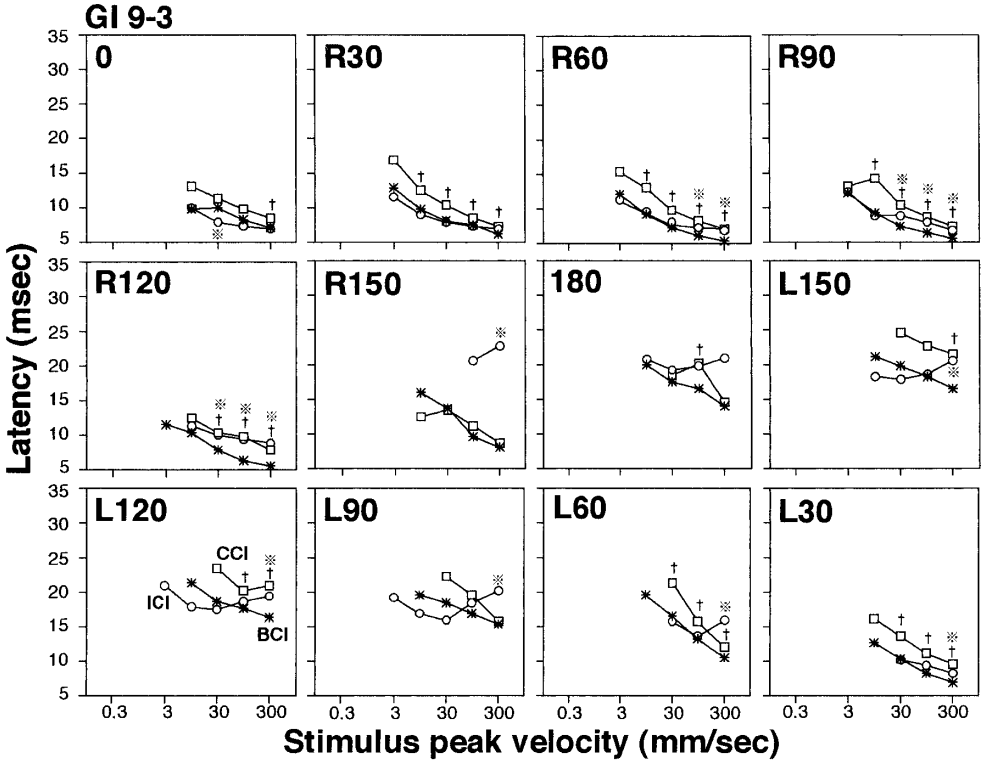

Fig. 12. Response latencies of GI 9–3. Symbols are the same as in Figs. 1 and 2. Latencies are relatively short to the air currents from the ipsilateral (right) side. Most of the directional air currents to which the GI showed large response magnitudes (Fig. 11) caused relatively short latency responses. Sample numbers are the same as in Fig. 11.

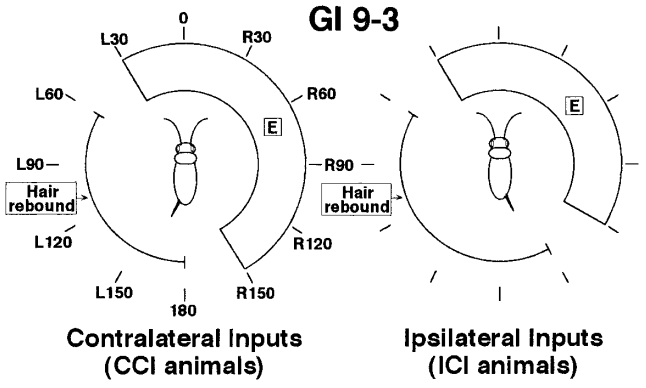

Fig. 13. Stimulus directions of excitatory inputs on GI 9–3. The directions of excitatory inputs in ICI and CCI animals are very similar to each other, i.e. the GI receives almost equivalent excitatory inputs from filiform hairs on both of the cerci. No inhibitory input was confirmed in GI 9–3. Refer to Figs. 4 and 10 for abbreviations.

Response properties of GI 9–3 in CCI animals

GI 9–3 in CCI animals showed responses to the unidirectional air currents regardless of the stimulus direction. However, responses to the air currents from180, L150, L120, L90 and L60 directions were very small (Fig. 11). Latencies of those responses were very long, i.e. most of them were more than 15 msec (Fig. 12). On the other hand, response magnitudes to the air currents from 180° opposite directions (0, R30, R60, R90 and R120, respectively) were relatively large (Fig. 11) and latencies of those responses were short (Fig. 12). These facts suggest that responses to the air currents from 180, L150, L120, L90 and L60 directions were caused by the rebound motion of cercal filiform hairs as in ICI animals. Therefore, excitatory inputs to GI 9–3 in CCI animals are from filiform hairs sensitive to the air currents from L30, 0, R30, R60, R90, R120 and R150 directions (Fig. 13, left).

The receptive field of GI 9–3 in CCI animals showed a cardioid shape but was much smaller than that in BCI animals (Fig. 1E). However, the preferred direction of the GI was still ipsilateral-front direction same as in BCI and ICI animals. The most preferred direction was thus kept constant even after the ipsilateral cercal ablation.

Unlike GIs 8–1 and 9–1, contralateral excitatory inputs on GI 9–3 seemed more powerful than those from ipsilateral ones (Fig. 11). Unlike other GIs (GIs 8–1, 9–1 and 9–3), any sign of inhibitory input was not confirmed on GI 9–3 by the spike count analysis in the present study.

DISCUSSION

Faint excitatory inputs for sharpening the directional characteristics

We showed that some GIs received very faint subthreshold excitatory inputs from cercal filiform hairs sensitive to the air currents from particular directions. For example, an air

current from R60 direction elicited significantly larger responses on GI 8–1 in BCI animals than those observed in ICI animals (Fig. 2). Such a difference must be due to the strong excitatory inputs from contralateral cercal filiform hairs sensitive to the R60 air current. However, the same air current given to CCI animals elicited only a few responses on the GI. This suggests that the contralateral excitatory inputs are not strong enough to elicit a neural response on GI 8–1 alone, but can make facilitative responses on the GI by working with the ipsilateral excitatory inputs. Similar weak excitatory inputs were also observed in GI 9–1. Although a response to R60 air currents was not observed in CCI animals, response magnitudes of the GI in BCI animals were significantly larger than those observed in ICI animals (Fig. 5). This also shows that contralateral filiform hairs encoding the R60 air current provide excitatory inputs to the GI for the facilitative responses. The facilitative responses caused by the weak contralateral inputs on both GIs 8–1 and 9–1 must have a significant role in sharpening the directional characteristic of these GIs.

Rebound motion of cercal filiform hairs

In addition to the simple excitatory and inhibitory connections, the rebound motion of cercal filiform hairs also have some role for making the neural responses of GIs 9–2 and 9–3 to an air current stimulus. As mentioned in the results, it has been reported that a sensory neuron of a filiform hair which responded to the rising phase of a unidirectional air-puff stimulus responded to the falling phase when the stimulus was delivered from 180° opposite directions (Tobias and Murphey, 1979). The authors of the previous study presumed that the response to the falling phase reflected the hair's return to rest position. However, we assumed that the response was due to the excess rebound motion of the hair after returning to the resting position because the structure of the basal part of a filiform hair indicated that the sensory neuron was activated only when the hair shaft was bent to one particular direction (Gnatzy and Tautz, 1980). As cricket filiform hairs show damped free oscillation after they are bent (Tautz, 1977; Shimozawa and Kanou, 1984b), a deflection angle of the filiform hairs during the excess rebound motion must be considerably smaller than that during the initial motion in rising phase. The small response magnitudes of GIs 9–2 and 9–3 during the rebound motion of filiform hairs must be due to such small deflection angles. As the rebound motion of a hair occurs after the motion in rising phase, a response latency of the sensory afferents to the rebound motion must be longer than that in the rising phase. This must be the reason why the response latencies of the GIs 9–2 and 9–3 were long in such conditions.

Neural rebound

GIs 8–1 and 9–2 in CCI animals showed very poor responses with appreciably long latencies (Figs. 2, 3, 8, 9) when the air currents were applied from particular directions (marked as "Neural rebound" in Figs. 4, 10). Those were somewhat paradoxical with the results obtained from the comparison of the response magnitudes between BCI and ICI animals, because such air currents applied to the contralateral cercus showed inhibitory or almost no effect on the GIs' responses (Figs. 4, 10). Evidently, those responses were not caused by the rebound motion of cercal filiform hairs because most of the filiform hairs sensitive to the air currents from 180° opposite to those directions showed inhibitory connections with the GIs (Figs. 4, 10). We hypothesized that those responses were caused by the rebound depolarization due to the release from an inhibitory hyperpolarization (neural rebound). The long latencies of the responses can be well explained by the hypothesis because the rebound depolarization, if any, occurs after a certain period of hyperpolarization caused by the air current stimulus. The poorness of the responses might also be explained by the hypothesis if the amplitude of rebound depolarization was smaller than the usual depolarization caused by the excitatory inputs. Although the hypothesis is based only on the circumstantial evidences, it is the most plausible interpretation for the paradoxical observations.

Significance of the neural responses evoked by the rebound depolarization has been suggested in some animals. In the central nervous system of the echolocating mustached bats (*Pteronotus parnellii*), for example, FM-FM neurons are created for encoding target range information (O'Neill and Suga, 1979). Such neurons show facilitative responses when an orientation sound (pulse) and a reflecting echo are presented with a particular time delay. That means the facilitation occurs when a long latency response to a pulse reaches an FM-FM neuron simultaneously with a short latency response to an echo. It has been thought that the long latency response to a pulse is formed by the rebound depolarization from inhibitions (Suga, 1990). Although we could not find any biological meaning of the rebound depolarization evoked responses in the cricket cercal sensory system, such responses must have some role for the processing of the air current information.

Polysynaptic inhibitory and excitatory neural pathways

As primary afferents in vertebrates or arthropods cannot mediate inhibition directly (Calabrese, 1976), inhibitory information from filiform hairs on the contralateral cercus to GIs 8–1, 9–1 and 9–2 must be relayed by inhibitory interneurons. Since directional characteristics of inhibition are different from GI to GI, different types and/or numbers of local interneurons in the cricket cercal system (Kobashi and Yamaguchi, 1984; Baba *et al*., 1995) must be recruited for mediating inhibitory information.

In addition to the polysynaptical inhibitory pathways, some of the excitatory information must also be polysynaptically relayed. For example, when the air currents were applied from R150 and L90 directions, response magnitudes of GI 9–1 became larger and response latencies of the GI became shorter than those in BCI animals after the ablation of the contralateral cercus (Fig. 6). Similar changes have been observed in GI 9–2 when the air currents were applied from R150 and L120 directions (Fig. 9). These facts suggest that the fili-

form hairs on the contralateral cercus sensitive to those air currents have inhibitory connection with each GI. Moreover, the inhibitory information must reach the GIs faster than or almost simultaneously with excitatory ones and affect the membrane potentials of the GIs because the inhibitory information apparently delays the GIs' membrane potential reaching a firing level. As inhibitory information must be relayed by at least one local interneuron as mentioned above, excitatory inputs to the GIs must also be polysynaptically relayed. The polysynaptical excitatory neural pathways in the cricket cercal system have also been suggested in the previous study dealing with local interneurons (Baba *et al*., 1995).

Consistency of GIs' preferred directions after the unilateral cercal ablations

In GIs 8–1, 9–1 and 9–2, excitatory inputs from contralateral cercal filfiorm hairs were considerably limited. Therefore, the receptive fields of the GIs were ascertained only in ICI animals. In GI 8–1, the receptive field of ICI animals was almost identical with that in BCI animals (Fig. 1A). In GIs 9–1 and 9–2, most velocity thresholds in ICI animals were different from those in BCI animals. However, the preferred direction of each GI was still consistent with that in BCI animals, i.e. ipsilateral-front and contralateral-rear directions for GI 9–1 and ipsilatral-rear direction for GI 9–2 (Fig. 1B, C). GI 9–3 received excitatory inputs from filiform hairs on both ipsilateral and contralateral cerci. Therefore, velocity thresholds of GI 9–3 in unilaterally cercal ablated animals were significantly higher than those in BCI animals. However, in spite of the shrinkage of the receptive fields, the most sensitive direction of GI 9–3 in the treated animals (ICI or CCI) was still ipsilateral-front directions as in BCI animals (Fig. 1D, E). Thus, the preferred direction (the most sensitive directions) of each GI was considerably well maintained even after a unilateral cercal ablation. We have observed that crickets still showed directional escape to an air current stimulus even after the unilateral cercal ablation, though the accuracy became worse to some extent (Kanou *et al*., in preparation). The consistency of GIs' directional properties must ensure the crickets keep the direction of the escape constant even after experiencing damages on cercal filiform hairs.

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