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Programmed Scale Detachment in the Wing of the Pellucid Hawk Moth, *Cephonodes hylas*: Novel Scale Morphology, Scale Detachment Mechanism, and Wing Transparency

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No scales of most lepidopterans (butterflies and moths) detach from the wings through fluttering. However, in the pellucid hawk moth, *Cephonodes hylas*, numerous scales detach from a large region of the wing at initial take-off after eclosion; consequently, a large transparent region without scales appears in the wing. Even after this programmed detachment of scales (d-scales), small regions along the wing margin and vein still have scales attached (a-scales). To investigate the scale detachment mechanism, we analyzed the scale detachment process using video photography and examined the morphology of both d- and a-scales using optical and scanning electron microscopy. This study showed that d-scale detachment only occurs through fluttering and that d-scales are obviously morphologically different from a-scales. Although a-scales are morphologically common lepidopteran scales, d-scales have four distinctive features. First, d-scales are much larger than a-scales. Second, the d-scale pedicel, which is the slender base of the scale, is tapered; that of the a-scale is columnar. Third, the socket on the wing surface into which the pedicel is inserted is much smaller for d-scales than a-scales. Fourth, the d-scale socket density is much lower than the a-scale socket density. This novel scale morphology likely helps to facilitate scale detachment through fluttering and, furthermore, increases wing transparency.

Key words: transparent moth wing, fluttering, pedicel, socket, scale development, conserved program

INTRODUCTION

In most lepidopterans (butterflies and moths), the wing mainly consists of a transparent cuticular membrane and numerous scales. Most scales are colored and petal-shaped, and entirely cover a transparent wing membrane. Thus, although being covered with scales produces a color pattern, it inhibits incident light from passing through the wing. However, a few lepidopterans have large transparent regions in their wings. In most of the transparent regions, the scales are small, slender, transparent, or erected, unlike the common colored, petal-shaped scales; consequently, a large region of the transparent wing membrane is exposed (Binetti et al., 2009; Goodwyn et al., 2009; Wanasekara and Chalivendra, 2011; Stavenga et al., 2012).

The pellucid hawk moth, *Cephonodes hylas* (Lepidoptera: Sphingidae), is diurnal and has a large transparent region in its wings (Fig. 1), unlike most other hawk moths. This moth flutters with high wingbeat frequency of approximately 70 Hz (Ando, 2005), and often hovers in the air as hummingbirds do (Warrick et al., 2005). Unlike the transparent wing regions described above, this transparent region has no scales; furthermore, it has remarkably high transparency because of an anti-reflective nanoprotuberance array on its surface (Yoshida et al., 1996, 1997). However, immediately after eclosion, the *Cephonodes* wing has no transparent region and is entirely covered with scales. Subsequently, numerous scales in a region of the wing are detached at initial take-off; consequently, this region becomes transparent (Inoue et al., 1959; Hennig, 1992). These scales appear to detach from *Cephonodes* wings through fluttering. This is unique because scales of most lepidopterans can only detach from their wings through accidental friction against solid objects, which can help them escape from predators in some cases (Eisner et al., 1964).

This programmed scale detachment from *Cephonodes* wings is rare in lepidopterans; therefore, it is possible that a novel mechanism is associated with this process. To investigate this scale detachment mechanism, we analyzed the process of scale detachment in detail using video photography and examined morphology of both the detached and attached scales (d- and a-scales, respectively) using optical and scanning electron microscopy. This study confirmed that the d-scales detach from the wing only through fluttering, and that d-scale morphology is novel among lepi-

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Fig. 1. *Cephonodes hylas*, hovering in the air and feeding on nectar.

dopteran scales. On the basis of these results, we suggest that this novel morphology likely helps to facilitate scale detachment and, furthermore, increases wing transparency.

MATERIALS AND METHODS

Animals

Cephonodes hylas eggs and larvae were collected from the flowers and leaves of gardenia, *Gardenia jasminoides*, that were planted in gardens and parks in Tokyo and neighboring prefectures in Japan. The larvae were reared on fresh leaves of *G. jasminoides* in plastic containers (11-cm diameter, 6-cm high) at approximately 20°C under a light:dark (L:D) cycle of 14L:10D–15L:9D. The pupae were kept at approximately 20°C under a 24-h L/D cycle: the L period started at 1:00 PM and the D period started at 5:00 AM. The pupal period was 11–13 days, and eclosion occurred from 7:00 AM to 9:00 AM. The freshly eclosed adults were examined by video photography, and their wings and scales were used for the examination by light, stereo, and scanning electron microscopy.

Video photography

Moths were video photographed on the tip of a wooden stick at approximately 25°C using a digital camera (TG-4, Olympus, Tokyo, Japan) while their wings quivered and fluttered. The still photographs at the critical phases of moth behavior were extracted from the video photographs.

Light microscopy

To examine scales by light microscopy, whole mounts of scales were prepared as follows. A small piece of wing was cut and placed on a glass slide. A small amount of 0.1% Triton X-100 solution was dropped onto the wing piece, and the scales were removed using forceps. After the scales were removed and dispersed, a cover slip was mounted and sealed with mounting medium (Entellan New, Merck, Damstadt, Germany). Each whole mount was examined with a light microscope (BX60, Olympus, Tokyo, Japan) equipped with Nomarski contrast optics. Fiji was used to measure scale area in each micrograph (Schindelin et al., 2012).

Stereo microscopy

After removal of most scales, each wing was examined with a stereo microscope (SZX16, Olympus, Tokyo, Japan) under incident light illumination and dark field illumination.

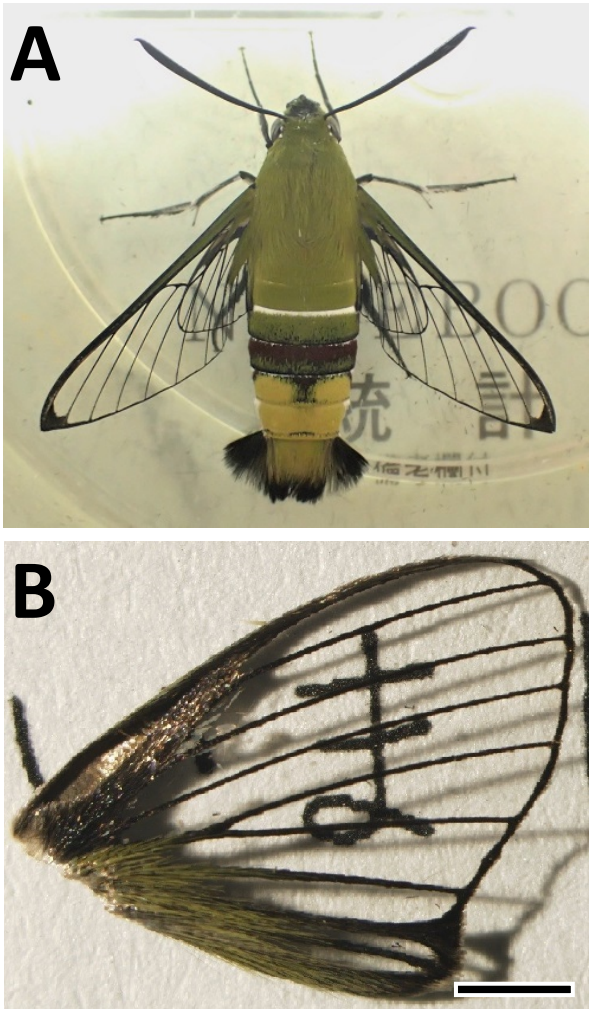

Fig. 2. *Cephonodes* wings with large transparent regions. **(A)** Adult at rest immediately after initial take-off. **(B)** The hindwing over a letter printed on copy paper. Scale bars: 2 mm.

Scanning electron microscopy

The scales attached to the wing, the scales detached from the wing, and the wing membrane after scale removal were examined by scanning electron microscopy. The dried specimens were coated with gold using an ion sputtering device (IB-3, Meiwafosis, Tokyo, Japan) and examined using a scanning electron microscope (JSM-6380LVN, JEOL, Tokyo, Japan).

Socket density on the wing membrane

The socket numbers were counted on the scanning electron micrographs of various sites on the wing membrane. The socket densities were calculated from the socket numbers and the areas of the examined sites on the wing membrane.

RESULTS

Large transparent regions in the *Cephonodes* wing

The wings after initial take-off had large transparent regions surrounded by colored wing margins and veins; these margins and veins were covered with scales (Fig. 2). Putting the excised wing over copy paper printed with a letter, the letter was clearly visible through the highly transparent region (Fig. 2B).

Behavior from eclosion to take-off

Immediately after eclosion, the moth had small opaque wings completely covered with scales (Fig. 3A). Subsequently, moths erected their wings over the dorsal surface of their bodies and their wings continued to expand for approximately 30 min. After the completion of wing expansion (Fig. 3B), moths kept this posture for approximately 30 min. Subsequently, the moths displaced their wings downward and spread them along their body sides (Fig. 3C), and kept this posture for approximately 30 min.

After the stage shown in Fig. 3C, the moths quivered their wings. From the onset of wing quivering, we observed the moths using video photography (see Supplementary Movie S1). From the video photographs, we extracted the still photographs of critical phases during the scale detachment process (Fig. 4). As shown in Fig. 4A, a small number of scales were detached from the wings during wing quivering for approximately 30 s. Subsequently, the moths stopped wing quivering and kept the same posture shown in Fig. 3C for approximately 10 s; meanwhile, they excreted meconium (Fig. 4B). Immediately after the excretion, their wings started to flutter intensely, and soon (within 1 s) the moths took off (Fig. 4C, D, E). Accompanying the intense fluttering, many scales were detached from the wings. The behaviors of the other two moths observed using video photography were very similar to that shown in Fig. 4.

Continuous observation of the moth behavior described above indicated that many scales were only detached from the wing through fluttering and not contact with a solid object.

Those scales that detached through fluttering were referred to as d-scales, whereas those that remained attached to the wing were referred to as a-scales.

Scale morphology

Figure 5 shows the wing excised from the bodies at the stage shown in Fig. 3C. The a-scales were localized along

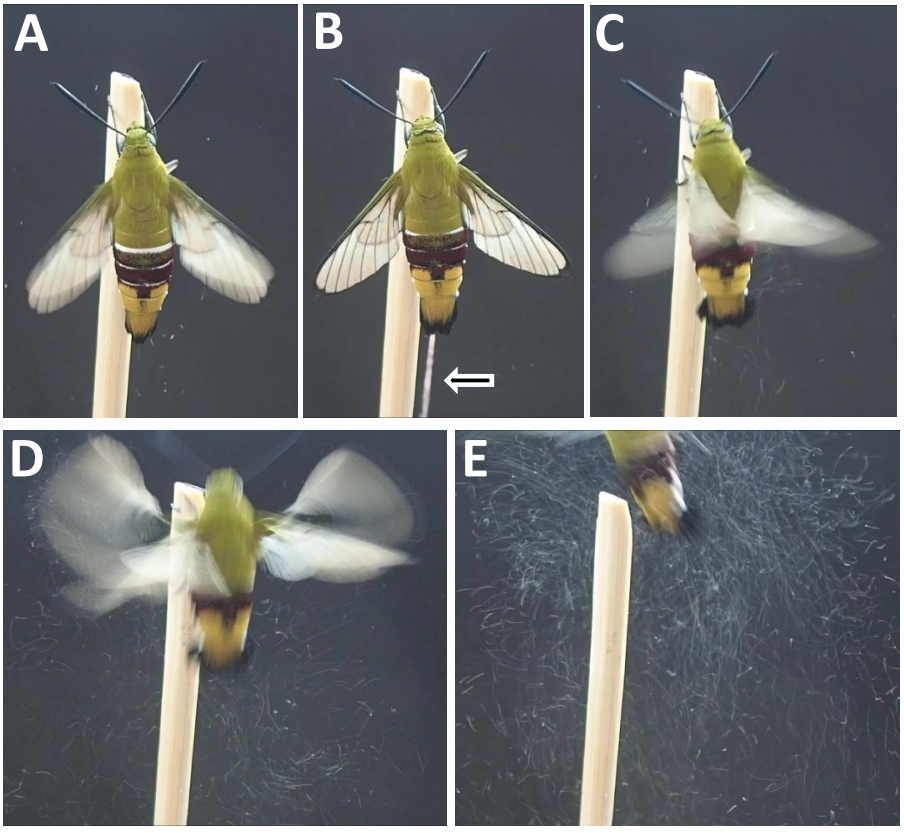

Fig. 4. Scale detachment process in the *Cephonodes* wing from wing quivering to take-off. **(A)** Wing quivering. A small number of the scales were detached from the wing and floated in air; the detached scales scattered the illuminated light. **(B)** Excretion of meconium (arrow). **(C–E)** From the onset of fluttering to take-off.

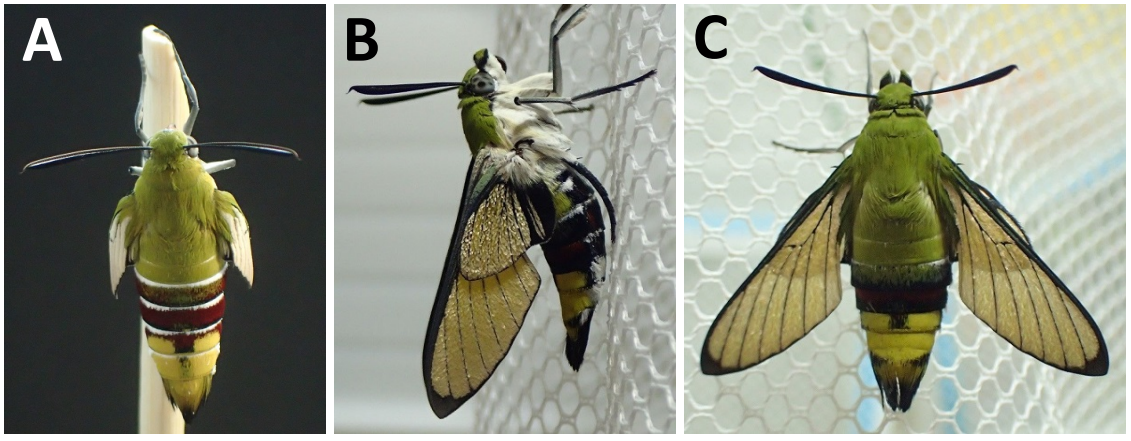

Fig. 3. *Cephonodes* behaviors after eclosion until wing spreading. **(A)** Immediately after eclosion. **(B)** After completion of wing expansion. **(C)** Immediately after wing spreading.

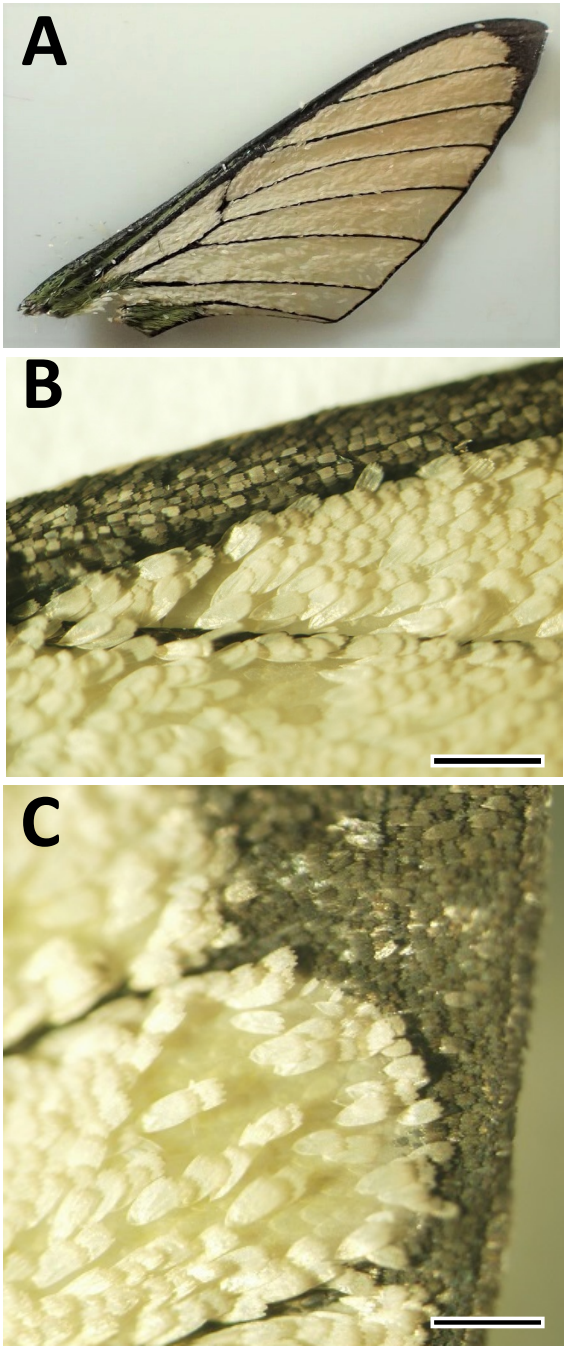

Fig. 5. Stereo micrographs of the ventral surface of the *Cephonodes* forewing covered with both d- and a-scales. **(A)** Whole view of the forewing. The length between the proximal and distal ends is approximately 30 mm. **(B)** Near the anterior wing margin. **(C)** Near the lateral wing margin. Scale bar: 500 μm.

the wing margins and veins as shown in Fig. 2, and the large regions occupied by the d-scales were surrounded by the wing margins and veins occupied by the a-scales (Fig. 5A). The d- and a-scales were clearly distinguished from each other by color and size: all d-scales were pale brown and larger, and a-scales were brown or dark brown and smaller (Fig. 5B, C).

To further study scale morphology, we pulled the scales from the wings at the stage shown in Fig. 3C and examined them using light microscopy. Figure 6 shows a typical d-scale and two typical a-scales that were selected among the majority of the scales, excluding the particularly small or long scales immediately near the wing margin, the wing vein, and the boundary between the d-scale and a-scale regions. The lengths and widths of d-scales were 364 ± 32 μm and 139 ± 10 μm (mean ± SD; n = 9), respectively; those of a-scales were 142 ± 23 μm and 47 ± 9 μm (mean ± SD; n = 9), respectively. The area of d-scales was $3.64 \times 10^4 \pm 0.40 \times 10^4$ μm^2 (mean ± SD; n = 9), and that of a-scales was $4.73 \times 10^3 \pm 1.26 \times 10^3$ μm^2 (mean ± SD; n = 9). Therefore, we estimate that d-scales are approximately eight times larger than a-scales. Immediately near the boundary between the d-scale and a-scale regions, several d-scales were smaller than the majority of d-scales. The minimum area of these smaller d-scales was approximately 3 times larger than the area of a-scales. Another distinctive feature of d-scales is their slender base or stalk, called a pedicel (Fig. 6 insets). The a-scale pedicel was generally columnar, which is common for scales (Downey and Allyn, 1975; Dinwiddie et al., 2014), whereas the d-scale pedicel was tapered.

Scanning electron microscopy confirmed that d-scales were much larger than a-scales (Fig. 7A, B). Additionally, the d-scale pedicel was tapered, whereas the a-scale pedicel was columnar (Fig. 7C, D). These results were consistent with those of light microscopy. The lengths and widths of a-scale pedicels were 9.7 ± 0.9 μm (mean ± SD; n = 9) and 3.2 ±. 0.5 μm (mean ± SD; n = 9), respectively. However, the length of d-scale pedicels was unable to be measured, because the boundaries between plates and pedicels of d-scales were obscure. The width of the ends of pedicels of d-scales was 2.5 ± 0.1 μm (mean ± SD; n = 4).

The microstructure of the upper surface of the scales is shown in Fig. 7E and F. Both the d- and a-scales had the four elements of scale microstructure that were described in the upper surfaces of common scales (Ghiradella, 1998): the longitudinal ridges run the length of the scale, the ridge lamellae are distally projecting spines located periodically along each longitudinal ridge, the transverse ribs connect adjacent longitudinal ridges, and the windows are the spaces surrounded by the longitudinal ridges and transverse ribs. However, the size and shape of each microstructural element were clearly different between d- and a-scales. Although the size of each a-scale microstructural element was similar to that of most lepidopteran scales (Ghiradella, 1998), those of the d-scale microstructural elements were not: the d-scale longitudinal ridge was more slender; the distances between adjacent ridges, adjacent spines, and adjacent transverse ribs were larger; and the area of the window was smaller than that of the a-scales, respectively.

The lower surface of the scale is shown in Fig. 7G and H. Large regions of the lower surfaces of both the d- and a-scales were fairly smooth, unlike their upper surfaces. This morphological feature of the lower surface is common to most lepidopteran scales (Ghiradella, 1998; Yoshida, 2002).

As described above, the scale surface microstructure is very similar between the d- and a-scales, except for the size and shape differences of the microstructural elements.

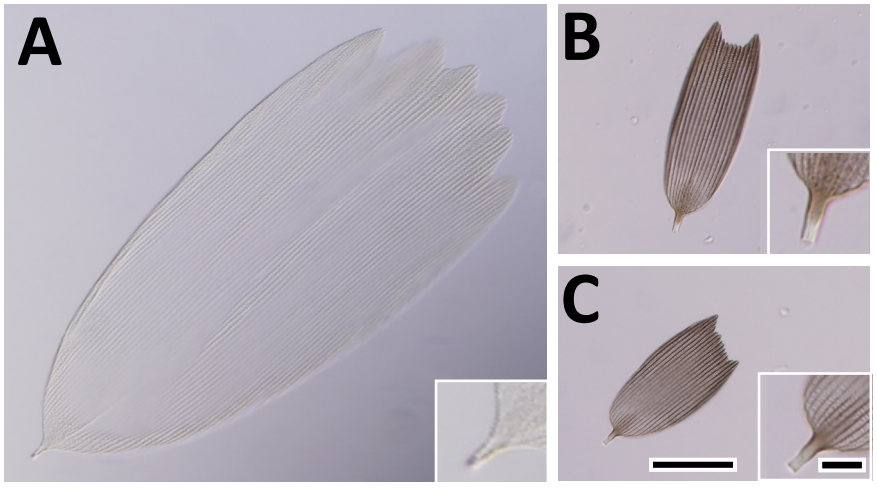

Fig. 6. Light micrographs of the scales pulled from the *Cephonodes* wing. **(A)** D-scales. **(B, C)** A-scales. The insets are the magnified views of the pedicels of each scale. Scale bar: 50 μm (10 μm in the inset).

Scale attachment sites on the wing surface

Figure 8 shows the scanning electron micrographs of the scale attachment sites on the wing surface. Both the d- and a-scales were attached to the wing through the attachment between the pedicel of the scale and the socket on the wing surface; the pedicel was inserted into the hole of the socket, which is common for most lepidopteran scales (Downey and Allyn, 1975; Ghiradella, 1998). The d-scale socket was much smaller than the a-scale socket. While the whole a-scale pedicel, approximately 10 μm in length, was inserted into the socket, only a small part of the d-scale pedicel was inserted into the socket, approximately 3 μm in length (Fig. 8C, D, G, H). Around the boundary between the d- and a-scale regions (Fig. 8I), some of the d- and a-scales were adjacent to each other, which clearly showed the morphological differences between the d- and a-scale attachment sites.

In the stereo micrograph (Fig. 5), the d-scales were raised from the wing surface, whereas the a-scales were in close contact with the wing surface. As shown in the lateral views of the scale attachment sites (Fig. 8G, H), the angle between the scale projection (or socket opening) and the wing surface was much larger in the d-scales (over 30°) than in the a-scales (approximately 0°), which is consistent with the view of the stereo micrograph (Fig. 5).

Socket density on the wing membrane

In the stereo micrograph (Fig. 5), the density of d-scales on the wing was much higher than that of the a-scales. The scale number was equal to the socket number, and although some scales may be lost, sockets are never lost. Thus, to quantitatively study scale density, we estimated the socket density on the wing membrane.

The d- and a-scale regions were readily distinguished from each other in the scanning electron micrograph of the wing membrane, even after removing scales (Fig. 9A); the d-scale regions are light and the a-scale regions are dark. The socket numbers were counted in magnified views of the d- and a-scale regions (Fig. 9B and C, respectively). The socket numbers were counted in four sites in the d-scale region and two sites in the a-scale region. The d-scale socket numbers were 18, 18, 19, and 21 (average, 19.0); the a-scale socket numbers were 187 and 189 (average, 188.0). The socket densities were estimated to be approximately 65/mm^2 and 644/mm^2 in the d- and a-scale regions, respectively. Therefore, the socket density in the d-scale region was approximately one-tenth of that in the a-scale region.

Wing membrane transparency, light scattering, and socket density

As shown in the micrograph of the wing membrane under incident light illumination (Fig. 10A), the letters on the copy paper were visible much more clearly through the d-scale region than the a-scale region; therefore, the transparency was much higher in the d-scale region.

As shown in the micrograph of the wing membrane under dark field illumination (Fig. 10B), there was much less scattered light in the d-scale region compared with the a-scale region. In the d-scale region, the dots of the scattered light were probably caused by the individual sockets that were dispersed against the dark background of the wing membrane. Alternatively, a large quantity of scattered light was observed throughout the a-scale region, which was likely because of the large amount of light scattering of the wing membrane itself, and the large size and high density of the sockets. Thus, the remarkably small size and low density of d-scale sockets likely contribute to the increased transparency of the wing membrane.

DISCUSSION

Moth behavior from eclosion to take-off

In this study, we observed a series of *Cephonodes* behaviors from eclosion to initial take-off, which is accompanied by scale detachment from the wing. After scale detachment, a large transparent region appears in the wing.

Unlike *C. hylas*, most other hawk moths have opaque wings that are entirely covered with scales. Truman and Endo (1974) described the wing expansion and spreading behaviors of *Manduca sexta*, a common hawk moth with opaque wings. The transition pattern of the wing position and duration of each wing position during *M. sexta* wing expansion and spreading are very similar to those of *C. hylas*. After wing spreading, *C. hylas* quiver their wings for approximately 30 s. This wing quivering, called pre-flight warm-up, was reported in many hawk moths; it increases the thoracic temperature to enable flying (Dorsett, 1962; Heinrich and Bartholomew, 1971). After wing quivering, *C. hylas* excreted meconium before initial take-off, as other lepidopterans do. Subsequently, these moths took off and concomitantly detached many scales from the wing.

As described above, the observed behaviors of *C. hylas* from eclosion to take-off were very similar to those of other

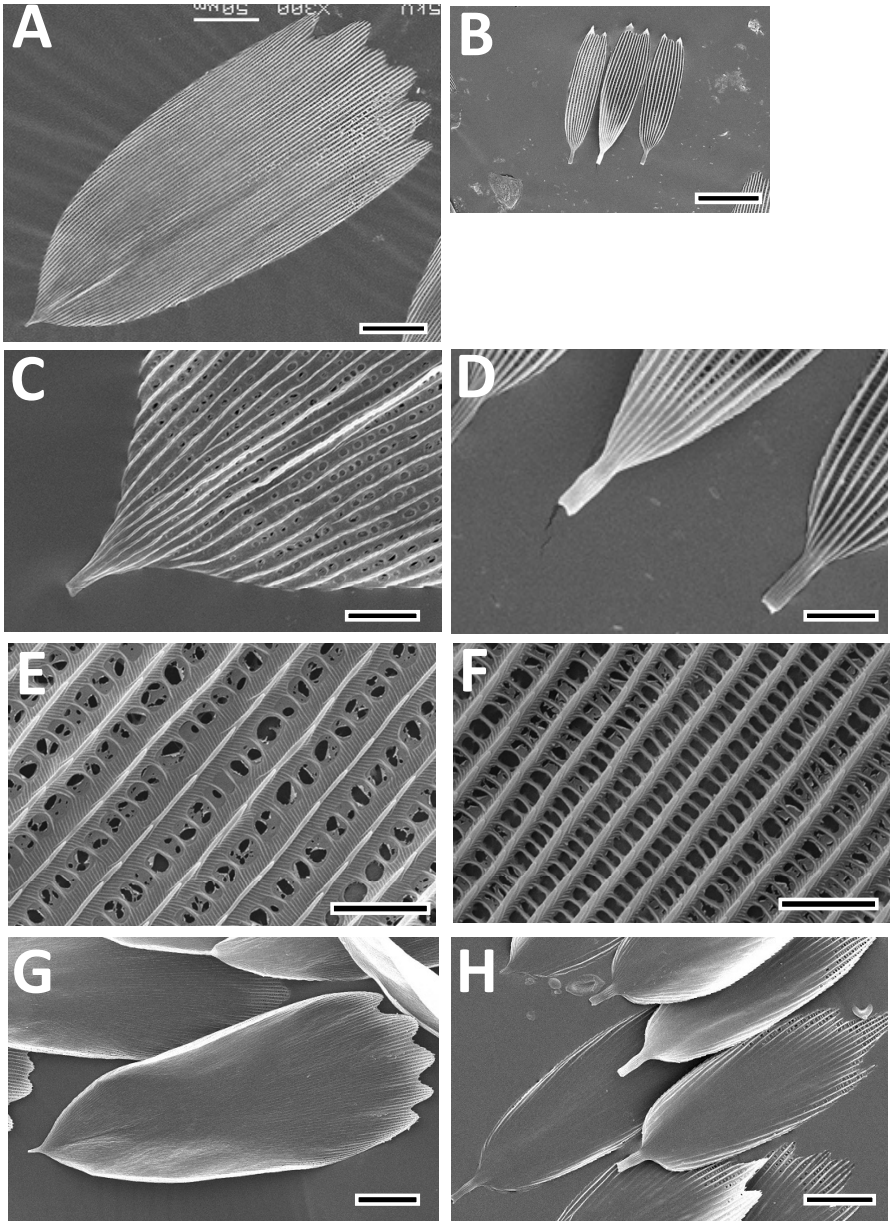

Fig. 7. Scanning electron micrographs of the d- and a-scales of the *Cephonodes* wing. **(A)** Upper surface of the d-scale. **(B)** Upper surface of the a-scale. **(C)** Magnified view around the pedicel of the d-scale in **(A)**. **(D)** Magnified view around the pedicel of the a-scale in **(B)**. **(E)** Microstructure of the upper surface of the d-scale. **(F)** Microstructure of the upper surface of the a-scale. **(G)** Lower surface of the d-scale. **(H)** Lower surface of the a-scale. Scale bars: 50 μm **(A, B, G)**, 10 μm **(C, D)**, 5 μm **(E, F)**, 20 μm **(H)**.

hawk moths with opaque wings; no specific behavioral events possibly related to scale detachment were observed. These findings showed that these scales detached from *Cephonodes* wings only through fluttering at initial take-off and without friction against solid objects. Thus, scale detachment from *Cephonodes* wings is programmed within the sequence of behaviors that is common to other hawk moths.

Novel scale morphology and its contribution to scale detachment

We reported three morphological features of the d-scale clearly distinguished from those of the a-scale. First, they are remarkably large. Simonsen and Kristensen (2003) reported that the average length of the wing scales of the Brahmin moth, *Brahmaea lucina*, is 322 μm, which is the largest among the scales attached to the wings, except for the long hair-like scales along the wing margins (Yoshida et al., 2017). The average length of the d-scale, 364 μm, is even larger than that of *Brahmaea lucina*. Second, only a small part of the scale pedicel is inserted into the remarkably small socket. Third, the scale pedicel is tapered; this pedicel morphology is the first report on lepidopteran scales. It is likely that all three features help facilitate scale detachment, as discussed below.

The first feature is that the d-scales are approximately eight times larger than the a-scales, which are a more common size. It is assumed that two kinds of forces are applied to the wing scale during fluttering: the viscous friction force generated by airflow, which is mainly applied to the wing scales during the upstroke and downstroke, and the inertial force generated by wing velocity change, which mainly occurs at stroke reversals. Because both the upstroke and downstroke are roughly circular motions centered on the wing base, both the friction and inertial forces are applied to the scales in the roughly distal direction of the wing. This direction corresponds to that needed for detachment of the distally projected scales.

The viscous friction force is roughly proportional to the area of the scale surface that is exposed to the airflow. Because the projecting angle of the a-scale to the wing surface is nearly 0°, the lower surfaces of the a-scales are almost unexposed to the airflow. However, that of the d-scale is over 30°; therefore, the lower and upper surfaces are more exposed to the airflow. Thus, the area effectively exposed to the airflow and therefore the viscous friction force applied to the d-scales was estimated to be over eight times larger in the majority of the d-scales, or over three times larger in several d-scales, compared with the a-scales.

The inertial force applied to the scales is directly proportional to the scale mass. Because the presumptive mass of

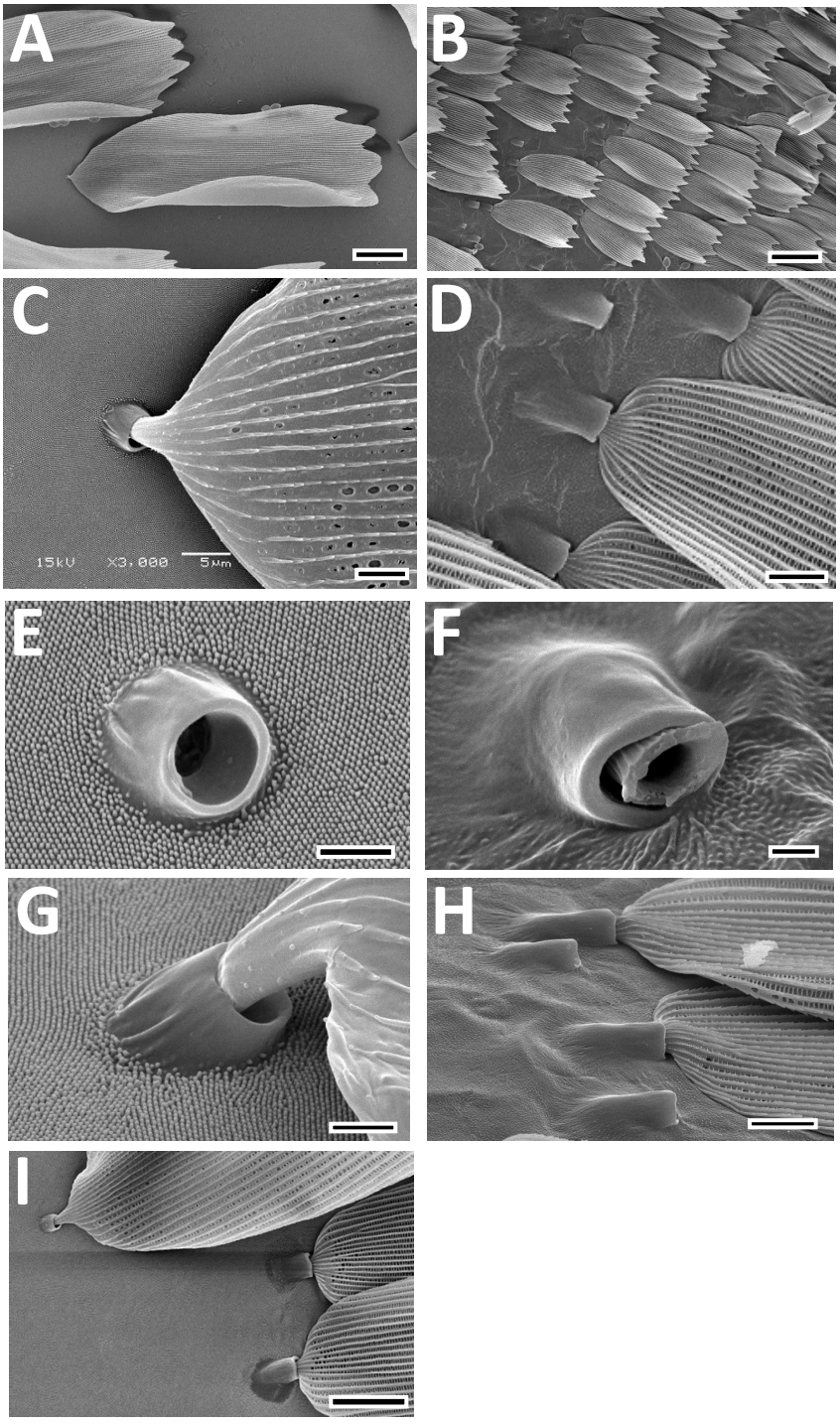

Fig. 8. Scanning electron micrographs of d- and a-scales attached to the *Cephonodes* wing. **(A)** D-scale. **(B)** A-scales. **(C)** Magnified view around the socket of **(A)**. **(D)** Magnified view around the socket of **(B)**. **(E)** D-scale socket with the nanoprotuberance array observed around the socket. **(F)** A-scale socket and torn pedicel. The nanoprotuberance array shown in Fig. 8E is not observed. **(G)** Lateral view of the d-scale base whose pedicel is inserted into the socket. **(H)** Lateral view of the a-scale base whose pedicel is inserted into the socket. **(I)** Scales around the boundary between the d- and a-scale regions before d-scale detachment. Scale bars: 50 μm **(A, B)**, 5 μm **(C)**, 10 μm **(D)**, 2 μm **(E, F, G)**, 10 μm **(H)**, 20 μm **(I)**.

the larger d-scales is larger than that of the a-scales, the inertial force applied to the d-scales is likely greater than that applied to the a-scales. Thus, it is likely that the total force applied to the d-scales during fluttering, which is in the direction that facilitates scale detachment, is much larger than that applied to the a-scales.

The second feature is that only a small part of the d-scale pedicel is inserted into the remarkably small socket, approximately 3 μm in length; in contrast, almost the entire a-scale pedicel, approximately 10 μm in length, is inserted into the socket. The scale is likely attached to the wing through the friction force generated between the outer surface of the pedicel and the inner surface of the socket. The friction force between two objects is roughly proportional to their contact area. Assuming that the contact area between the pedicel and socket is proportional to the contact length between them, the friction force between them is approximately three-tenths in the d-scale compared with the a-scales. Thus, the minimum force necessary for scale detachment is likely much smaller in the d-scales compared with the a-scales.

The third feature is that the d-scale pedicel is tapered; that of the a-scale is columnar, which is more common. A stopper inserted into a bottle neck is morphologically similar to the pedicel inserted into the socket. As schematically shown in Fig. 11, once the stopper starts to be displaced inside the bottle neck, the tapered stopper can be pulled out of the bottle neck more rapidly than the columnar stopper, because the contact area between the stopper and bottle neck decreases much more rapidly in the tapered stopper than in the columnar stopper. For the same reason, once the scale pedicel starts to be displaced, detachment occurs much more rapidly in d-scales than a-scales.

Methods for obtaining transparent wing regions

The large transparent region of *Cephonodes* wings develops through d-scale detachment. Because the remarkably large d-scales are eventually lost from the wing, *C. hylas* may waste energy on scale production. The production cost of d-scales can be estimated to be approximately the same as that of the a-scales in the wing because d-scales are approximately eight times larger than a-scales but the d-scale density is approximately one-tenth that of the a-scales. Thus, there does not appear to be an additional cost for producing the large d-scales in *C. hylas*.

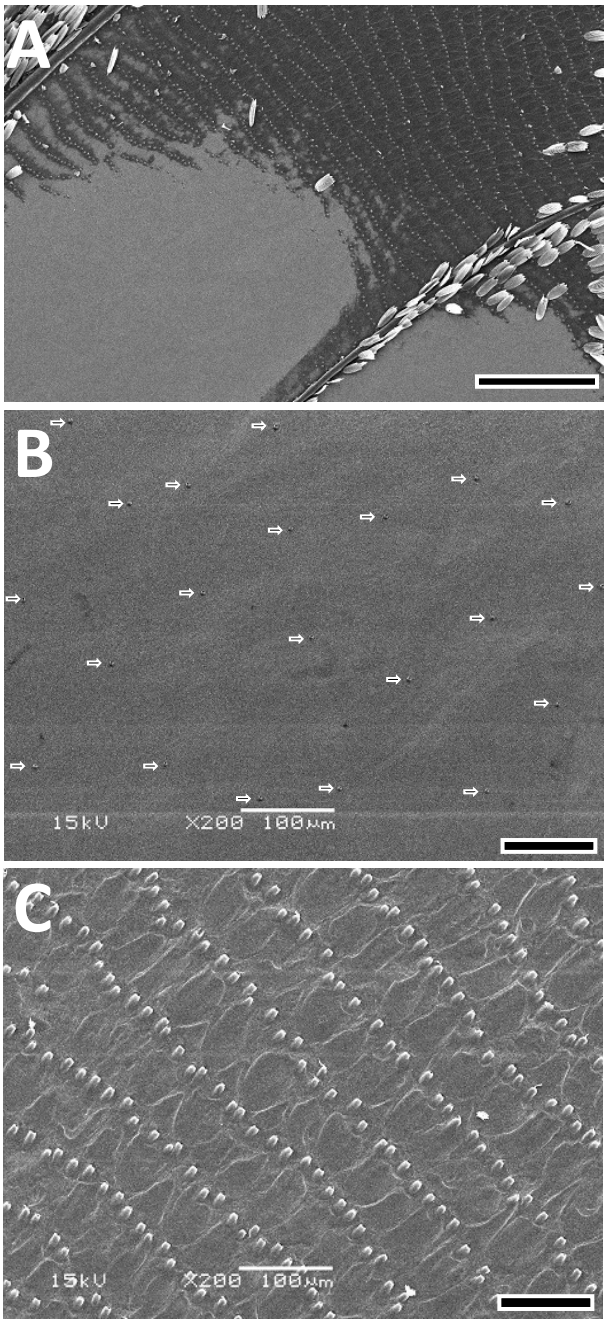

Fig. 9. Scanning electron micrographs of the sockets on the ventral surface of the *Cephonodes* wing. **(A)** Sockets around the boundary between the d- and a-scale regions. Most scales were removed, but some a-scales remained attached around the wing veins. **(B)** D-scale sockets, indicated by arrows; there are 21 sockets. **(C)** A-scale sockets; there are 189 sockets. Scale bars: 500 µm **(A)**, 100 µm **(B, C)**.

If a moth were able to make transparent wings without producing scales, there would be less of a cost than making wings with scales that are later detached, as in *C. hylas*. However, no lepidopteran wings have ever been reported that did not produce scales throughout their wings. Therefore, wing development without producing scales seems unlikely to have evolved in lepidopterans. As in the pro-

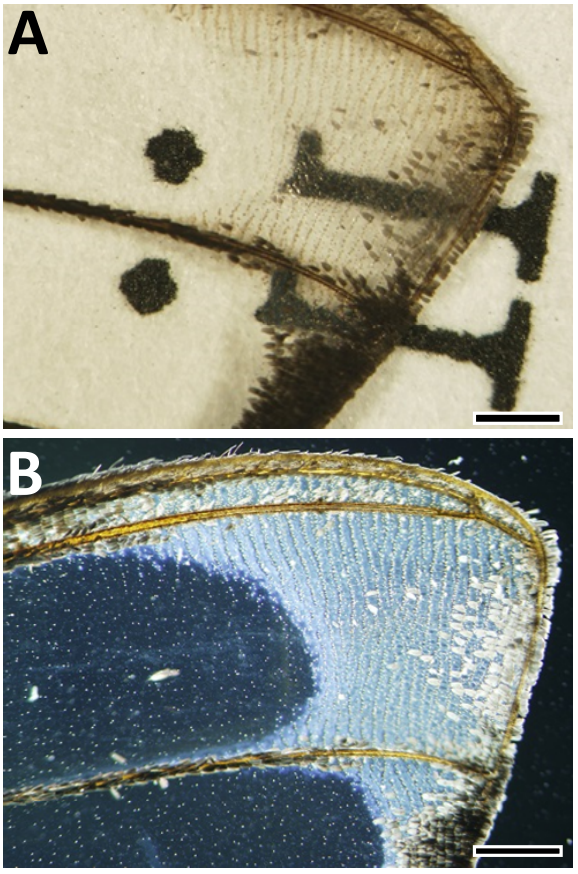

Fig. 10. Stereo micrographs of the distal part of the *Cephonodes* forewing membrane after removing many a-scales. **(A)** The wing over letters printed on copy paper under incident light illumination. **(B)** The wing under dark field illumination. Scale bar: 500 µm.

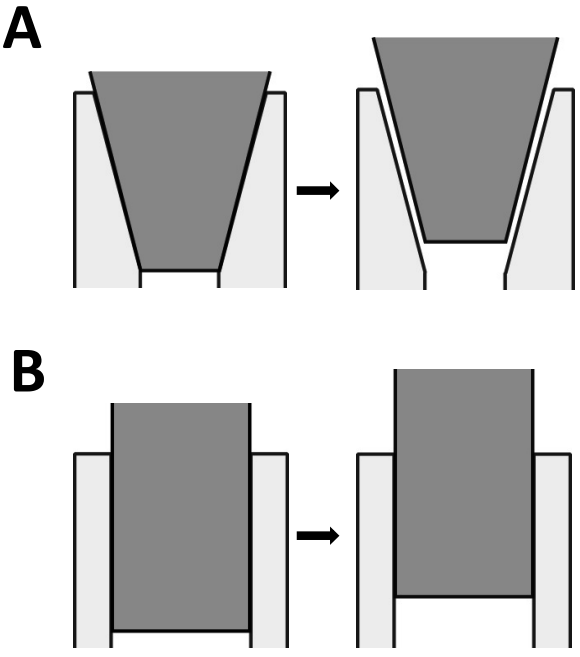

Fig. 11. Schematic diagram of stopper displacement inside a bottle neck. From left to right, the stopper starts to be displaced upwards. **(A)** Tapered stopper. **(B)** Columnar stopper.

grammed cell death in which once produced cells are later eliminated if unnecessary (Lockshin and Zakeri, 2001), the *Cephonodes* wing eliminates the unnecessary products, the d-scales, after completing scale cell development (Stossberg, 1937; Süffert, 1937). Although programmed scale cell development has been mostly conserved in lepidopteran wings, the scale morphology widely varies among regions within the wings, as in *C. hylas*, and among lepidopteran species (Downey and Allyn, 1975; Ghiradella, 1998).

Aside from *C. hylas*, transparent wing regions formed through scale detachment have been observed in several species of hawk moths (Sphingidae) and numerous species of clearwing moths (Sesiidae) (Hennig, 1992; Arita et al., 1994; Yoshida and Kato, unpublished). However, these other species have scales in the transparent regions (Binetti et al., 2009; Goodwyn et al., 2009; Wanasekara and Chalivendra, 2011; Stavenga et al., 2012). Moreover, their scales are small, slender, transparent, or erected, which allows exposure of the transparent wing membranes. In the glasswing butterfly, *Greta otto*, the wing membrane is covered with hair-like scales and has high transparency that is comparable to that of *C. hylas* (Yoshida et al., 1997; Siddique et al., 2015). Regardless of scale presence or absence, both types of wings have high transparency. However, whether the programmed scale detachment from wings provides moths with additional functions other than high wing transparency remains unknown.

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COMPETING INTERESTS

The authors have no competing interests to declare.

AUTHOR CONTRIBUTIONS

AY and YK designed the study. YK and HT collected and reared animals. AY and YK performed the experiments. AY, YK, and RK interpreted the data. AY prepared the draft, and all the authors contributed to revisions.

SUPPLEMENTARY MATERIAL

Supplementary material for this article is available online. (URL: https://doi.org/10.2108/zs210031)

Supplementary Movie S1. Scale detachment process in *C. hylas* from wing quivering to take-off.

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