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Novel wing display and divergent agonistic behaviors of two incipient *Psolodesmus* damselflies

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Abstract

Sexual selection via male competition is a strong evolutionary force that can drive rapid changes in competitive traits and subsequently lead to population divergence and speciation. Territorial males of many odonates are known to use their colorful wings as visual signals and to perform agonistic displays toward intruders. *Psolodesmus mandarinus dorothea* and Psolodesmus mandarinus mandarinus are two parapatrically distributed sister damselflies that share similar ecological characteristics but differ markedly in wing coloration. The wings of P. m. dorothea are mostly clear, whereas those of P. m. mandarinus have a large area of black pigmentation and a central white patch. We investigated whether territorial males of the two damselflies at breeding sites display distinct agonistic behaviors associated with their respective wing colors. Behavioral interactions between territorial and intruder males and their wing kinematics were filmed and analyzed for P. m. dorothea in Lienhuachih of central Taiwan, and P. m. mandarinus in Tianxiyuan and Fusan of northern Taiwan. We observed that the P. *m. mandarinus* males exhibited a novel set of perched wing displays, which was not only absent in its sister *P. m. dorothea* but also previously unknown in Odonata. At breeding sites, perched rival males of *P. m. mandarinus* with pigmented wings exhibited escalating agonistic wing-flapping and wing-hitting displays toward each other. In contrast, territorial males of P. m. dorothea with clear wings engaged only in aerial chase or face-to-face hovering when intruder males approached from the air. These results indicate that the two sister P. mandarinus damselflies diverged behaviorally in territorial contests and support the hypothesis of coadaptation on the basis of wing colors and types of wing movement in Odonata. Our findings further suggest that divergent agonistic wing displays may play a pivotal role in the speciation mechanism of P. mandarinus damselflies. The sequential analyses of behavioral characteristics and progression suggest that P. m. mandarinus damselflies likely use mutual assessment of rivals in territorial contests.

Keywords Calopterygidae \cdot Male competition \cdot Odonata \cdot *Psolodesmus mandarinus* \cdot Sexual selection \cdot Speciation \cdot Taiwan

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Introduction

Sexual selection via male competition for mates is a potent evolutionary force that drives population divergence and shapes speciation processes (Lackey et al. 2018; Tinghitella et al. 2018). In animals where direct combat between rival males mainly determines reproductive success (West-Eberhard 1983; Andersson 1994), agonistic interactions among males can produce strong selection that causes rapid evolutionary changes in traits used in competitive signaling and contests, such as coloration, body size, and weapons (Seehausen and Schluter 2004; Grether et al. 2013; McCullough et al. 2014). For example, aggression bias toward males with similar competitive traits may give males with a rare novel trait to have a fitness advantage because they receive less aggression from rival males (Seehausen and Schluter 2004). This negative frequency-dependent selection for rare competitive traits can permit the origin and maintenance of novel traits in the presence of multiple traits (Mikami et al. 2004; Dijkstra and Border 2018). This and other mechanisms of selection may facilitate reproductive isolation and subsequently lead to divergence of populations with different competitive male traits (Lackey et al. 2018; Tinghitella et al. 2018). Sexual selection via male competition often favors individuals engaging in direct combat for the fitness advantage (Andersson 1994). However, direct physical combat can be costly in terms of the time and energy trade-offs, risk of predation, and injury. Therefore, selection tends to favor males that can accurately assess their ability to win against rivals and reach contest resolution without engaging in injurious behaviors associated with physical combat (Arnott and Elwood 2009).

Dragonflies and damselflies (Insecta, Odonata) constitute a group of highly visual insects that exhibit a wide diversity of wing and body coloration (Corbet 1999; Silsby, 2001; Córdoba-Aguilar 2008). Males are often territorial and compete for food and mates at feeding and breeding sites (Corbet 1999; Suhonen et al. 2008). Territorial males are known to display various agonistic behaviors toward conspecific and heterospecific intruders when they are in perches, mate guarding, or patrolling flights (Corbet 1999). When approached by an intruder, a perched male can perform a simple threat display involving abrupt movement of the wings, abdomen, or body position (reviewed in Corbet 1999). However, more commonly, a perched male reacts to an intruder in its territory by quickly taking off, flying toward the intruder, and adopting various threat displays with diverse wing movements (e.g., Pajunen 1966; Corbet 1999; Hilfert-Rüppell and Rüppell 2013). The agonistic behavior of males in flight often features characteristic face-to-face hovering, circling and spiral flights, reciprocal lunges and vertical oscillations, and parallel flights (Corbet 1999). During perched and aerial threat displays, the coloration of wings can function as visual signals for recognizing competitors (Anderson and Grether 2010; Günther et al. 2014), assessing the rival's quality (Schultz and Fincke 2009; Guillermo-Ferreira et al. 2015a, b), and determining the contest outcome (Xu and Fincke 2015; Guillermo-Ferreira et al. 2019). When confronted with rivals, odonate species with colored wings were found to exhibit unique flying styles and have more wing pauses than the species with clear wings, suggesting that agonistic behavior involving wing movements in odonates is likely adapted to optimize the presentation of visual signals (Hilfert-Rüppell and Rüppell 2013; Günther et al. 2014).

Psolodesmus mandarinus (Calopterygidae) is a damselfly species endemic to Taiwan and consists of two closely related sister subspecies, *P. m. dorothea* Williamson 1904 (Fig. 1a and b) in central and southern Taiwan and P. m. mandarinus MacLachlan 1870 (Fig. 1c and d) in northern Taiwan (Lin et al. 2014). The two subspecies are distributed parapatrically and co-occur only near the boundaries of their ranges at approximately 24 degrees of north latitude in Taiwan. Another closely related species, P. kuroiwae Oguma 1913, is restricted to Yaeyama islands of the Ryukyu Archipelago. These two Taiwanese subspecies have similar ecological characteristics, and both inhabit shady, fast-flowing streams in lowland forests. Genetic analyses of the mitochondrial COI, 16S, nuclear ITS and 5.8S genes suggested that they are indistinguishable between these subspecies (Lin et al. 2012, 2014). The two subspecies are also identical in general morphology and genitalia (anal appendages and penile organs) but differ markedly in wing coloration (Lin et al. 2012, 2014). The fore- and hindwings of P. m. dorothea are mostly clear (hyaline) with a small amount of apical black pigmentation (Fig. 1 a and b), whereas P. m. mandarinus's wings have a large area of apical black pigmentation, a central white patch, and a subbasal brown pigmentation (Fig. 1c and d) (Lin et al. 2012). Females and males of both subspecies have similar wing colors, except for the colors of the pterostigma (Fig. 1). In odonates, wing color and movement can play important roles in visual signaling among males during territorial competition (Schultz and Fincke



Fig. 1 *Psolodesmus mandarinus dorothea* (**a**) male and (**b**) female with small black apical pigmentation and hyaline wings, and *Psolodesmus mandarinus mandarinus* (**c**) male and (**d**) female with larger pigmented area, conspicuous white patch, and brown basal coloration. The inset shows the pronounced pruinosity present in mature *P. m. mandarinus* males. (All images taken by LSB; a and b from Lianhuachih Forest Reserve, Nantou, central Taiwan; c and d from Fusan Botanical Garden, Yilan, northern Taiwan)

2009; Anderson and Grether, 2010; Günther et al. 2014; Xu and Fincke 2015; Guillermo-Ferreira et al. 2015a, b, 2019). The striking differences in wing colors between *P. m. doro-thea* and *P. m. mandarinus* raise questions of whether the two damselflies use wing displays as visual signals in male competition, and whether they display distinct agonistic behaviors associated with their respective wing colors.

The comparative study of agonistic behaviors in closely related odonate species with contrasting wing colors can lead to the identification of candidate traits targeted by sexual selection and provide insights into the mechanisms of how male competitive traits affect divergence of the species (Hilfert-Rüppell and Rüppell 2013; Svensson and Waller 2013; Günther et al. 2014; Cordero-Rivera and Zhang 2018). This study aimed to investigate how P. m. dorothea and P. m. mandarinus males behave and use their wings in territorial contests at breeding sites. Our main hypothesis is that the males of these two P. mandarinus subspecies use their wing colors and movement as visual signals during competition. Therefore, we predict that P. m. mandarinus, which has relatively more colorful wings (a central white patch and subbasal brown pigmentation) than P. m. dorothea, exhibits a distinct wing display involving characteristic wing movements to conspicuously present their wing ornaments toward rivals in territorial contests. The agonistic behaviors of P. m. dorothea and P. m. mandarinus in their natural habitats were characterized and compared using observations, video recordings, and sequential analyses of behavioral elements.

Materials and methods

Study sites, damselfly populations, and mating behaviors

A population of P. m. dorothea in the Lienhuachih Research Center, Nantou, central Taiwan (N 23° 55' 7.57", E 120° 53' 9.05") and a population of P. m. mandarinus in the Tianxiyuan Ecology Education Center, Taipei, northern Taiwan (N 25° 7' 57.16", E 121° 35' 33.40") were observed and video-recorded. These two populations represent the typical wing colors of the two subspecies (Lin et al. 2012) (Fig. 1). Additional observations were performed in a population of P. m. mandarinus in the Fushan Botanical Garden, Yilan, northern Taiwan (N 24° 45' 23.10", E 121° 35' 32.79"). The streams in Lienhuachih are approximately 2 m in width, with their banks covered by dense vegetation of secondary forests. The streams in Tianxiyuan and Fushan are wider, averaging approximately 3 m and 5 m in width, respectively. The stream banks comprised exposed rocks or were covered with the vegetation of disturbed primary forests in Fushan and secondary forests in Tianxiyuan. The

stream substrate at the three study sites was mostly gravels and cobbles with semisubmerged vegetation and woody debris.

During the breeding season between June and October, adults of both P. mandarinus subspecies frequently present in areas near the small streams of lowland forests. Mature males can be found perching on the vegetation and rocks by the streams and compete to defend mating territories in areas with rapid-flowing water. Females visit these territories to mate and oviposit in submerged vegetation and debris. In contrast to calopterygid damselflies with elaborate courtship behaviors (Córdoba-Aguilar and Cordero-Rivera 2005), males of both these P. mandarinus subspecies perform no apparent precopulatory courtship display and readily clasp arriving females for copulation (n=9). After copulation, females go to oviposition sites and search for submerged vegetation on which to lay eggs. Males guard females from harassment from other males by following and perching near egg-laying females at oviposition sites.

Behavioral recordings, wing kinematics, and statistical analyses

The observations of male interactions and mating behaviors at breeding territories took place between 9:00 AM and 3:00 PM from July to October 2017 (22 days, 132 h) in Tianxiyuan and from July to August 2018 (21 days, 126 h) in Fushan for P. m. mandarinus and from August to October 2018 (19 days, 114 h) in Lienhuachih for P. m. dorothea. Behavioral interactions between territorial and intruding males were observed and filmed using a digital camera (Casio EX-ZR1500, Casio Computer Co., Tokyo, Japan) at 30 fps for single frame analysis. Males in Fushan and Lienhuachih were color marked on the abdomen for individual identification. Males in Tianxiyuan were not marked. Territorial males were identified as those perched near ovipositional sites in a specific area for more than 5 min. Intruding males were identified as those that came close to the territory, and behavioral responses from the territorial males were observed. Video recordings were shot, as much as possible, from the lateral view of the contesting males. VSDC Video Editor v.5.8.7.830 (Flash-Integro LLC, New York, USA) was used to analyze the recordings. The wing beat frequency, WBF (Hz), was obtained by calculating the time (s) for one cycle of wing movement, from the start to the end, of a male in the resting position (folding above the thorax) (Fig. 2a). The stroke amplitude, A (°), was measured as the angle between the start and end of down- or upstrokes (Fig. 2b-d). The stroke angular speed, ω (rad/s), was calculated by converting the amplitude to radian (rad)

Fig. 2 Schematic view of the Psolodesmus mandarinus mandarinus wing position and stroke direction (arrows). Resting position (a) with wings closed along the sagittal plane and resting wing flap (RWF) (b) with ventral wing surface stroking along the frontal plane (y-axis). In wing flapping (c) and wing hitting (d) threat displays, the ventral surface of the forewing (FW) and hindwing (HW) stroke is anteriorly positioned (z-axis). Stroke amplitude, denoted by A (°), is defined as the angle between starting and ending positions of a wing relative to the stroke direction





divided by the time (s) for a complete down- or upstroke of the wing $[\omega = (A*0.174533)/s]$.

After testing the normality of the data by Shapiro–Wilk's tests, the data that were not normally distributed were analyzed using the Kruskal–Wallis H and Wilcoxon rank sum tests with RStudio v. 1.2.1335 (RStudio Inc., Boston, USA) to determine the differences in WBF, A and ω of the wing displays between behavioral stages. The significance levels of multiple comparisons were determined by Wilcoxon rank sum tests followed by Bonferroni correction. Because the forewings and hindwings of damselflies displayed distinct movements during agonistic behaviors, the analyses were performed separately for each pair of wings.

Sequential analyses of agonistic behaviors

The behavioral stages of the agonistic behavior were manually transcribed from the recordings using VSDC Video Editor. Following the method of Chen et al. (2020), the behavioral sequences were summarized into adjacency matrices using the igraph network analysis package (https://igraph.org) in R for sequential analysis (Green and Patek 2018). A permutation procedure of 10,000 replicates was conducted to examine whether a specific behavioral transition was more frequent than could be expected by chance.

Results

Agonistic behaviors

Psolodesmus mandarinus dorothea

A total of 22 agonistic interactions between males of P. m. dorothea were observed, and 19 of these interactions were video recorded. Territorial males guarded their breeding territories while perched on the vegetation or rocks by the sides of the stream. When an intruder male approached from the air (stage I), the territorial male directly took off (stage II) either to chase (stage III) the intruder (n = 11)or to engage in an aerial flight (stage IV) through faceto-face hovering (n = 8) (Fig. 3a) (Online resource 1). During face-to-face hovering, both males consistently faced directly toward each other, with an average WBF of approximately 8.22 ± 0.86 Hz (n = 8) (Online resource 2), while the orientation of the pairs drifted up, down, or sideways. The aerial flights terminated upon the escape of one of the two males. The aggression between territorial males and intruders lasted approximately 7.66 ± 4.31 s (n = 14) (Online resource 2), in which shorter interactions were mostly aerial fights involving face-to-face hovering Fig. 3 Ethograms of territorial contests of (a) *Psolodesmus mandarinus dorothea* and (b) *Psolodesmus mandarinus mandarinus*. *P. m. dorothea* has a simpler behavioral repertoire and performs direct aerial contests; *P. m. mandarinus*, on the other hand, shows more agonistic behaviors during perched displays prior to engaging in aerial contests



 $(3.93 \pm 6.32 \text{ s}, n = 8)$, whereas longer interactions were direct chases $(5.43 \pm 3.44 \text{ s}, n = 11)$ (Online resource 2).

Psolodesmus mandarinus mandarinus

When perched alone without any intruders, the territorial males flapped their wings at irregular intervals (Online resource 3). The forewings and hindwings moved downward to amplitudes of approximately 60° $(A = 60.3 \pm 16.34^\circ, n = 16 \text{ flaps/9 individuals})$ and 84° $(A = 88.75 \pm 3.31^\circ, n = 16 \text{ flaps/9 individuals})$ along the y-axis, respectively (Figs. 2b, 4a, and 5a and b; Online resources 3 and 4). Then, both wings paused for approximately 0.4 s (forewings, 0.41 ± 0.71 s; hindwings, 0.43 ± 0.73 s; n = 16 flaps/9 individuals) before returning to the folding position above the abdomen (Fig. 4a). These resting wing flaps (RWFs) had an average WBF of approximately 1.2 Hz (forewings, WBF = 1.17 ± 0.52 Hz; hindwings, WBF = 1.11 ± 0.46 Hz; n = 16 flaps/9 individuals) (Fig. 5c and d; Online resource 4), with characteristically slow downstrokes (forewings, $\omega = 1.99 \pm 0.62$ rad/s; hindwings, $\omega = 2.69 \pm 1.06$ rad/s; n = 16 flaps/9 individuals) and fast upstrokes (forewings, $\omega = 3.31 \pm 1.46$ rad/s; hindwings, $\omega = 4.82 \pm 2.93$ rad/s; n = 16 flaps/9 individuals) (Figs. 4a and 5e-h; Online resource 4).

Seventy-three agonistic interactions between perched P. m. mandarinus males were observed, and 24 of them were filmed for video analyses (Tianxiyuan, n = 44/14; Fushan, n = 29/10). All males involved in these agonistic interactions were mature and had pronounced pruinosity covering the black apical portions of ventral fore- and hindwings (Fig. 1c, inset). When approached by a flying intruder male, a perched territorial male often quickly chased the intruder, a behavior similar to that of P. m. dorothea males (Fig. 3a). When an intruder male approached a perched territorial male by landing near its perch in the breeding territory, a series of agonistic behaviors in the form of wing displays was performed by both males (n = 73)(Fig. 3b, Online resources 3 and 5). This series of agonistic behaviors consisted of eight distinct behavioral stages: (I) arrival display; (II) wing flapping (types 1 and 2); (III) wing hitting (types 1 and 2); (IVA) take-off/land; (IVB) changed perch; (V) aerial attack; (VI) escape/chase; and (VII) spiral flight (Fig. 3b).

Stage I. Arrival display

Upon the approach and before the landing of an intruder male, both pairs of wings of the territorial male quickly unfolded from their resting positions above the thorax



Fig. 4 Phase relationship of *Psolodesmus mandarinus mandarinus* perched displays. Wing beats are presented as mean values, but the time between cycles is based on an individual. The resting wing flap (RWF) (**a**) shows asynchronous movement with a sloping downstroke (i), peak pause (ii), and steep upstroke (iii). In stage II.1 (**b**) and stage II.2 (**c**), the nearly synchronous displays are differentiated by differences in duration, peak pause, and a secondary peak during stage II.2. Stage III.1 (**d**) and stage III.2 (**e**) asynchronous displays are similarly differentiated with differences in duration, peak pause, and secondary peak

(Fig. 2a) and moved anteriorly with the ventral side facing toward the direction of the incoming intruder (Figs. 2c and 3b) for an amplitude of approximately 90° along the z-axis (Fig. 2c; Online resource 4). With the ventral wing surface displayed toward the opponent, the hindwings' costal margins were aligned along the x-axis, and the forewings were positioned approximately 45° above the hindwings (Fig. 2c). This wing display was maintained for an average of approximately 1.06 ± 0.58 s (n = 45) until the intruder perched in front of or beside the territorial male. During the display, the territorial male continuously adjusted its body position to allow the ventral sides of the wings to face directly toward the direction of the approaching intruder (Online resource 4). When the intruder did not land, the territorial male either took off and chased the intruder or stayed on its perch.

Stage II. Wing flapping

Type 1: From the resting position of the wings above the abdomen (Fig. 2a), both forewings and hindwings started downstrokes anteriorly along the y- and z-axes (forewings, $\omega = 8.81 \pm 4.79$ rad/s; hindwings, $\omega = 8.61 \pm 1.09$ rad/s; n = 344) (Figs. 2c, 3b, and 5e and f; Online resources 3-5), which ended approximately 90° away from the resting position (forewings, $A = 84.8 \pm 16.1^{\circ}$; hindwings, A = $87.0 \pm 10.6^{\circ}$; n = 344) (Fig. 5a and b; Online resources 3-5). At the end of the downstrokes, the costal margins of hindwings were parallel along the x-axis with the ventral surface directed toward the opponents (Fig. 2c; Online resources 3 and 4). Then, both wings performed characteristic "bouncing pauses" (Fig. 4b; Online resource 5), in which they moved back and forth at approximately 90° from one to six times (pair 8, 2.64 ± 1.66 , n = 47) in 0.2 s (forewings, 0.20 ± 0.09 s; hindwings, 0.21 ± 0.10 s; n = 344) before starting the upstroke (forewings, $\omega = 4.67 \pm 3.02$ rad/s; hindwings, $\omega = 4.67 \pm 2.3.02$ rad/s; n = 344) (Fig. 5g and h; Online resource 4 & 5) and returning to the resting position. The WBF of the forewings and hindwings in the type 1 wing flapping was approximately 1.8 Hz (forewings, 1.88 ± 0.907 Hz; hindwings, 1.82 ± 0.88 Hz; n = 344) (Fig. 5c and d; Online resource 4). These agonistic wing flaps were different from the resting wing flaps in that the former move in the forward not the downward directions (agonistic: y- and z-axes; resting: y-axis) (Fig. 2b and c; Online resources 3 and 5).

Type 2: After the end of the first downstrokes (forewings, $\omega = 9.10 \pm 3.27$ rad/s; hindwings, $\omega = 8.75 \pm 3.32$ rad/s; n = 47) (Figs. 3b and 5e and f; Online resources 3–5) of both wings near 90°, the forewings and hindwings began the first upstrokes (forewings, $\omega = 10.3 \pm 3.56$ rad/s; hindwings, $\omega = 9.55 \pm 3.86$ rad/s; n = 47) (Fig. 5g and h; Online resources 4 and 5) until reaching within approximately 15° of the resting position (A = 14.04 ± 8.12°, n = 47) and 30° (A = $32.13 \pm 11.02^{\circ}$, n = 47) of the resting position, respectively, and then, both wings continued the second downstrokes (forewings, $\omega = 10.1 \pm 4.55$ rad/s; hindwings, $\omega = 10.4 \pm 4.08$ rad/s; n = 47) (Fig. 5e and f; Online resources 4 and 5) to approximately 75° (forewings, $A = 72.2 \pm 19.7^{\circ}$; hindwings, $A = 74.4 \pm 14.4^{\circ}$; n = 47) (Fig. 5a and b; Online resources 4 and 5) before both wings began the second upstrokes (forewings, $\omega = 3.81 \pm 2.03$ rad/s; hindwings, $\omega = 3.84 \pm 1.68$ rad/s; n = 47) (Fig. 5g and h; Online resources 4 and 5) back to the resting position (Fig. 4c; Online resource 3). The wing beat frequencies of the forewings and hindwings during type 2 wing flapping were approximately 1.1 Hz (forewings, 1.11 ± 0.18 Hz; hindwings, 1.09 ± 0.17 Hz; n = 47) (Fig. 5c and d; Online resource 4).

Fig. 5 Boxplots showing *Psolodesmus mandarinus mandarinus aggressive* behaviors and mean measurements for wing beat amplitude (A) (**a**, **b**), wing beat frequency (WBF) (**c**, **d**), and angular speed (ω) (**e**–**h**) for both forewings (FW) and hindwings (HW). Plots with different individual letters are significantly different, whereas those sharing the same letters indicate nonsignificant differences based on pairwise Wilcoxon rank sum test (see Online resource 7)



Stage III. Wing hitting

Type 1: The type 1 wing-hitting began with the downstrokes of both wings (forewings, $\omega = 19.4 \pm 11.0$ rad/s; hindwings, $\omega = 9.83 \pm 5.49$ rad/s; n = 286) from their resting positions forward along the y- and z-axes to approximately 170° (forewings, A = $168.25 \pm 20.8^{\circ}$, n = 286) and 90° away (hindwings, A = $85.3 \pm 13.7^{\circ}$, n = 286) (Figs. 2d, 3b, 4d, and 5a and b) (Online resources 4–6). At the end of the downstrokes, the tips of the forewings pointed directly toward the rival male, and the costal margins of the hindwings paralleled the x-axis with the ventral surface directed toward the opponents, similar to that of type 1 wing flapping in stage II (Fig. 2c) (Online resource 7). At this stage, the wings sometimes made contact with the opponent. After the end of the downstrokes, both wings quickly began upstroking (forewings, $\omega = 12.6 \pm 9.02$ rad/s; hindwings, $\omega = 5.60 \pm 4.43$ rad/s; n = 286) (Fig. 5g and h; Online resources 5 and 6) back to the resting position. The WBFs of the forewings and hindwings during type 1 wing hitting were approximately 2.26 and 2.13 Hz, respectively (forewings, 2.26 ± 1.13 Hz; hindwings, 2.13 ± 1.09 Hz; n = 347) (Fig. 5c and d; Online resources 2 and 5).

Type 2: After the first downstrokes (forewings, $\omega = 13.4 \pm 4.51$ rad/s; hindwings, $\omega = 9.38 \pm 4.10$ rad/s; n = 82) (Figs. 3b and 5 and f; Online resources 3–5), similar to that of type 1 wing hitting, both forewings and hindwings during type 2 wing hitting quickly began the first upstrokes (forewings, $\omega = 19.6 \pm 6.63$ rad/s; hindwings, $\omega = 7.15 \pm 2.79$ rad/s; n = 82) (Fig. 5g and h; Online resources 3–5) back to approximately 20° (forewings, A = 15.9 ± 8.76°, n = 82) and 40° (hindwings, A = 37.87 ± 15.05°, n = 82), respectively (Fig. 4e; Online resources 3–5). Then, both wings began the second downstrokes (forewings, $\omega = 12.4 \pm 9.34$ rad/s; hindwings, $\omega = 11.7 \pm 6.41$ rad/s; n = 82) (Fig. 5e and f; Online resources 3–5) to approximately 90° (forewings, A = 92.3 ± 34.3°; hindwings, A = 86.9 ± 10.3°, n = 82) (Fig. 5a and b; Online resources 3–5). Afterward, both wings started the second upstroke (forewings, $\omega = 4.60 \pm 2.55$ rad/s; hindwings, $\omega = 4.16 \pm 1.73$ rad/s; n = 82) back to the resting position. The WBF of type 2 wing hitting was approximately 1.1 Hz (forewings, 1.07 ± 0.21 Hz; hindwings, 1.04 ± 0.21 Hz; n = 82) (Online resources 3–5).

The main difference between stage II wing flapping and III wing hitting is the angle of the downstroke of the forewings (wing flapping, 90°; wing hitting, 170°; Fig. 2c and d). In stages II and III, the two contestant males gradually changed their body positions to have their wing flaps oriented toward each other (Online resource 5). At the beginning of the perched displays, the wing flaps (stages II and III) were performed in turn by the contestants (Fig. 6, 20–55 s), but they became almost simultaneous as the contests proceeded to later stages (Fig. 6, 110–115 s). However, there were cases when neither the intruder nor territorial male responded to the wing flap of each other (Fig. 6, 130–135 s), or an irregular alternation between stages II and III was observed (Fig. 6, 55–75 s).

Fig. 6 Sample pair of contesting *Psolodesmus mandarinus mandarinus* males showing the progression and synchronicity of aggressive behaviors



Stage IV_A. Take-off/land

This stage was initiated by the take-off of one of the two perched contestants (Fig. 3b). The flying male approached the perched male soon after take-off and landed again near his previous perch or a different position near the perched male (approximately 10.33 ± 4.97 cm; flight duration 1.95 ± 0.72 s, n=6) (Online resources 3 and 4). During this time, the perched male performed the arrival display toward the opponent in the air as described in stage I (Fig. 3b).

Stage IV_B. Change perch

In contrast with stage IVA, this stage was characterized by the take-off of one contestant away from its opponent and the landing on a different perching site. The male that tookoff was subsequently followed by its opponent to the new contest site (n=8) (Fig. 3b; Online resources 3 & 4). Upon landing, contestants resumed their display toward each other.

Stage V. Aerial attack

Similar to stage IVA, stage V started with the take-off of one of the two perched males (Fig. 3b; Online resources 3 and 4). Then, the flying male approached the perched male from the air within a close range (approximately 1.98 ± 1.87 cm; flight duration 1.29 ± 0.5 s, n = 23), while the perched male continuously adjusted its position toward the flying male and performed an arrival display as in stage I (Fig. 3b). The flying male sometimes used its wings to physically attack the wings of the rival or grasped and landed directly on the head of the perched male while continued performing wing flapping or repeated aerial attacks (n = 26) (Online resources 3 and 4).

Stage VI. Escape/chase

One of the two perched males took off and flew away from the perch (Fig. 3b; Online resources 3 and 4). Then, the other male took off and followed the escaping male from behind. Afterward, the winner males returned to the breeding territory.

Stage VII. Spiral flight

An ascending spiral flight was performed when both perched males took off into the air from their perches one after the other (Fig. 3b; Online resources 3 and 4). The aerial movement of one male was closely followed by the other male to maintain a face-to-face position. The two males often ascended spirally into the forest canopy at a high velocity (n = 12). The stage ended when the winner male performed a dive maneuver to land back in the territory (Online resource 3) and the loser male flew away. The distance between the two perched contestants started at 6.08 ± 3.48 cm and ended with a shortened distance of 5.04 ± 3.27 cm (n = 24) (Online resource 4).

Wing kinematics of agonistic behaviors in *P. m. mandarinus*

The results of Shapiro-Wilk's tests indicated that the distributions of A, WBF, and ω of both wings were all significantly deviated from normality (Online resource 6). The Kruskal-Wallis H tests for not normally distributed data showed that there were significant differences among the behavioral stages in all three kinematic parameters (Online resource 6). The Wilcoxon rank sum tests of pairwise comparisons revealed that, for the FWs, the A values in stages II and III were significantly larger than those in the RWF, except for the 2nd downstrokes of the type 2 wing flapping in stage II (II.2.2) (Fig. 5a; Online resource 7). In stage III, the FWs had significantly larger A than the FWs in stage II, except for the 2nd downstrokes of the type 2 wing hitting in stage III (III.2.2) (Fig. 5a; Online resource 7). For the HWs, the A of all stages did not significantly differ, except for a smaller A in the 2nd downstroke of the type 2 wing flapping during stage II (II.2.2) (Fig. 5b; Online resource 7). For the FWs, the type 1 wing hitting in stage III (III.1) had a significantly higher WBF than that of the type 1 wing flapping in stage II (II.1) and the remaining stages (Fig. 5c; Online resource 7). For the HWs, the type 1 wing hitting of stage III (III.1) had a significantly higher WBF than that of all other stages, except for the type 1 wing flapping of stage II (II.1) (Fig. 5d; Online resource 7). For the downstrokes of the FWs, in all II and III stages, ω was significantly higher than that of the RWF (Fig. 5e; Online resource 7). The type 1 and 2 wing hitting of stage III (III.1 and III.2.1) was associated with significantly higher ω than that of the type 1 wing flapping of stage II (II.1) (Fig. 5e; Online resource 7). For the downstrokes of the HWs, in all II and III stages, ω was significantly higher than that of the RWF (Fig. 5f; Online resource 7). For the upstrokes of the FWs, in stage III (III.1 and III.2.1), ω was significantly higher than that of the RWF (Fig. 5g; Online resource 7). The 1st upstroke of the type 2 wing hitting of the stage III (III.2.1) had a significantly higher ω than that of all the stage II behaviors. For the upstrokes of the HWs, ω in stage II and III was not significantly different than that of the RWF (Fig. 5h; Online resource 7). The latency (s) of wing displays (the duration between consecutive wing displays) decreased as the contests progressed, suggesting the escalating nature of the wing displays in most territorial contestants of P. m. mandarinus (Online resources 8 and 9).

In summary, for the FWs, stage III was associated with significantly larger A, WBF, and ω than those of the RWF (Fig. 5a, c, e and g). For the FWs in stage II, A and ω (down-strokes) were significantly larger than those of the RWF (Fig. 5a and e); in stage III, A and ω (downstrokes) were significantly larger than those in stage II (Fig. 5a and e). For the HWs, in stage III, the WBF and ω (downstrokes) were

significantly higher than those of the RWF (Fig. 5d and f), and in stage II, ω (downstrokes) was significantly higher than that of the RWF (Fig. 5f). In stages II and III, A, WBF, and ω did not significantly differ from each other. These results indicate that the wing movements in stages II and III are kinematically distinct from those of "normal" wing behaviors displayed by males perching alone in the breeding territory, and suggest that "wing hitting" of stage III is a more aggressive and energetically more demanding behavior than "wing flapping" of stage II, especially for the FWs.

Behavioral sequences of territorial contests

The behavioral sequences of 13 P. m. dorothea and 24 P. m. mandarinus contestant pairs were analyzed (Online resources 10 and 11). From the initial encounters of "approach" and "take-off," the majority of male P. m. dorothea entered "hovering flight" (Fig. 7a; transitional probability from I to IV, 0.67; II to IV, 0.64), with the others performing "escape/chase" (Fig. 7a; I to III, 0.33; II to III, 0.36). "Hovering flight" always ended up in "escape/chase" (Fig. 7a; IV to III, 1). In contrast, the contests of *P. m. man*darinus showed many behavioral elements and followed a more variable course of behavioral sequences (Fig. 7b). The contest started when the intruder approached the territorial male (percentage of "approach" in total behaviors, 2.3%), and in response, the territorial male performed an "arrival display" (6.3%). After the initial encounters, males began "wing flapping" (IB to II, 0.92), directly entered "escape/ chase" and "spiral flight" (IA to VI, 0.14; IA to VII, 0.16), or changed perch (IA to IVB, 0.05). "Wing flapping" was often repeated many times (II to II, 0.7; "wing flapping," 39.6%), or progressed to "wing hitting" (II to III, 0.19). Then, more aggressive "wing hitting" was repeated many times (III to III, 0.81; "wing hitting," 43.8%), or returned to "wing flapping" (III to II, 0.12). "Aerial attack" significantly led to "spiral flight" (V to VII, 0.38) but less often to "wing flapping" (V to II, 0.35), "wing hitting" (V to III, 0.12), or "arrival display" (V to IA, 0.12). However, "take-off/ land" proceeded to "wing hitting" (IVA to III, 0.29), "wing flapping" (IVA to II, 0.43), or "arrival display" (IVA to IA, 0.14). "Change perch" frequently led to "wing flapping" (IVB to II, 0.75) or sometimes back to "arrival display" (IVB to IA, 0.25).

Discussion

Novel agonistic wing display in Odonata

The results show that males of *P. m. mandarinus* exhibit a unique set of agonistic behaviors in elaborate forms of wing displays by perching close to each other in breeding

Fig. 7 Psolodesmus mandarinus dorothea (n = 13) (a) and Psolodesmus mandarinus mandarinus (n = 24) (b) sequential analysis of aggression behaviors of contesting males. Circles denote aggressive behaviors, with circle size denoting the corresponding percentage relative to the total number of behaviors performed and colors representing increasing aggression. Arrow thickness and direction indicate the probability of the transition from one behavioral stage to another. Solid lines indicate significant transitions (higher probability of observed transitions than expected), while dashed lines indicate nonsignificance. Transitions with probabilities < 0.1 were not included in the figure (see Online resources 10 and 11)



territories. Currently known agonistic behaviors of odonates consist of the arrival displays, face-to-face hovering, and various forms of aerial chasing and fights (Pajunen 1966; Corbet 1999; Córdoba-Aguilar and Cordero-Rivera 2005; Hilfert-Rüppell and Rüppell 2013). To our knowledge, this set of perched wing displays by male P. m. mandarinus is novel and previously unknown in Odonata (Corbet 1999). Even among winged insects, similar agonistic behavior of reciprocal wing waving of perched rivals is found only in a number of true fruit flies (Diptera, Tephritidae) (reviewed in Benelli 2014), suggesting a convergent evolution of the perched wing display in two evolutionarily distant insect lineages. For other winged animals, such as birds, the wing waving or flapping is common and often used together with vocal signals in courtship displays or begging for food (Andrew 1961), but less frequently used as agonistic behaviors between rivals in breeding territories (e.g., swamp sparrows, Anderson et al. 2013).

Several lines of evidence in our study demonstrate that the wing flapping (stage II) and wing hitting (stage III) of *P. m. mandarinus* are escalating agonistic displays, which likely function as visual signals by conspecific males for assessment and dominance in territorial contests. First, these two behaviors were observed in males only after intruders entered the territories of residents, and they were kinematically distinct from the RWF of males perching alone in their territories. Second, rival males continued adjusting and orienting their body axes toward the opponents while performing these behaviors, and the distance between them decreased as the contest progressed. Third, in the less aggressive wing flapping, both FWs and HWs turned perpendicularly to the body axis, with the ventral side facing toward rivals and performing characteristic "bouncing pauses," which likely function as essential visual signals at close range. Fourth, in the more aggressive wing hitting, the FWs moved forward to be parallel to the body axis with the tips of the FWs pointing toward the rival and sometimes appeared to be used as "weapons" to strike the wings or body of the rival male. Fifth, rival males often consecutively or reciprocally repeated wing flapping and wing hitting significantly many times in one contest series. Finally, the latency of wing displays in most contestant pairs shortened as contests progressed, suggesting that the territorial contests in P. m. mandarinus often escalated into more aggressive phases at later stages, where the wing displays were performed at a higher frequency.

The current work is significant in the context of the general view of territorial contests in Odonata. In particular, past and current research on odonate agonistic behavior and assessment strategies in breeding territories has mainly focused on aerial wing displays and fights (Corbet 1999), while perched wing displays have been overlooked,

especially among tropical species (however, see Cordero-Rivera and Zhang 2018). The perched wing display in P. m. mandarinus may be an energetically more efficient assessment strategy for contest resolution than aerial wing display, because rival males use less energy by perching rather than conducting energy-demanding behaviors such as hovering in the air or engaging in aerial chasing and spiral flight. The perched wing display may also be a more informative and effective assessment strategy in terms of signal quantity and quality due to the stationary nature and lower WBF of the wing displays at perch, as opposed to continuous position changes and higher WBF in most aerial wing displays. For an extreme example, in the face-to-face hovering of the phoenix damselfly Pseudolestes mirabilis (Pseudolestidae), rival males hover in the air using only the hyaline forewings while displaying motionless colored hindwings, which are potentially honest visual signals for male quality (Cordero–Rivera and Zhang, 2018).

The findings of this study are also important in documenting behavioral diversity in wild populations (Caro and Sherman 2012; Cordero-Rivera 2017). Psolodesmus is a monotypic damselfly genus with only two species, P. mandarinus of Taiwan and P. kuroiwae of Yaeyama Islands, which are phylogenetically distantly related to all other members of the Calopterygidae (Lin et al. 2014; Svensson and Waller 2013). The closest extant sister genus of *Psolodesmus* is *Mnais*, which has nine Asian species with intraspecific male wing polymorphisms that are associated with alternative reproductive strategies of orange-winged territorial fighters and clear-winged nonterritorial sneakers (Tsubaki et al. 1997). The wing polymorphism and pigmented wing are probably ancestral traits in this damselfly lineage (Svensson and Waller 2013). The common ancestor of Mnais and Psolodesmus likely has male wing polymorphism at the species or intraspecific level. However, a perched wing display similar to male P. m. mandarinus has not been found in Mnais or other related calopterygid damselflies with colorful wings. Therefore, the perched wing display in P. m. mandarinus may represent an evolutionarily novel and derived behavior of Odonata, which has evolved only in this geographically and phylogenetically isolated damselfly. Future research on understudied tropical damselfly species is needed to document the diversity of male-male agonistic behavior in Odonata, and to fully assess the novelty and antiquity of the perched wing display in P. m. mandarinus.

It is interesting to consider the reasons that perched wing display in breeding territories has evolved in *P. m. man-darinus*, and the ecological and evolutionary factors that may have driven and maintained this agonistic behavior. The evolution of wing display is correlated with the origin of the colored wing in Odonata, as both traits are under intense intraspecific sexual selection (Svensson and Waller 2013; Wellenreuther and Sánchez-Guillén 2016). However,

the colored wing by itself cannot explain the origin of the perched wing display in P. m. mandarinus since numerous odonate species are equipped with colorful wings (Corbet 1999). More specifically, we propose that the unique combination of wing colors of P. m. mandarinus, in which the white patch between black and brown pigmentation and the apical pruinosity on the ventral side of wings possibly play important roles in the origin of perched wing display. The white wing patch and pruinosity of P. m. mandarinus may function to scatter broadband, UV, or polarized light and produce high intensity, contrasting or flashing signals against the relatively dark forest background in the breeding territory, similar to the colored wings of the helicopter damselfly, Megaloprepus caerulatus (Pseudostigmatidae) (Schultz and Fincke 2009; Xu and Fincke 2015), and phoenix damselfly, Pseudolestes mirabilis (Pseudolestidae) (Nixon et al. 2017; Orr et al. 2017; Cordero-Rivera and Zhang 2018). The effect of white wing patch and pruinosity as visual signals in territorial contests may be more pronounced in P. m. mandarinus, whose habitats are densely covered, shady streams of tropical forests.

Divergent agonistic behavior of *P. m. dorothea* and *P. m. mandarinus*

The results support the hypothesis that males of the two P. mandarinus subspecies use their wing colors and movement as visual signals in male competition. We confirm the hypothesis suggesting that males of P. m. mandarinus with conspicuous wings exhibit a unique set of agonistic behaviors in the form of wing displays by perching close to each other in breeding territories. This set of agonistic behaviors is absent in its sister subspecies P. m. dorothea with mostly hyaline wings. The results further indicate that males of both subspecies diverged behaviorally during aerial fights in territorial contests, in which P. m. dorothea engages only in face-to-face hovering, whereas only P. m. mandarinus performs spiral flights. These findings are significant not only because they show the functional link between wing colors and agonistic behavior in P. mandarinus damselflies but also because they provide evidences to support the hypothesis of coadaptation between wing traits and styles of wing movement in Odonata (Hilfert-Rüppell and Rüppell 2013; Günther et al. 2014). Although our results emphasize the importance of functional coadaptation between conspicuous wing color and unique perched wing display in P. m. mandarinus, the hyaline wing in P. m. dorothea also possibly functions as a visual signal during face-to-face hovering flight or at perch, especially against dark forest backgrounds. The interference pattern of transparent wings was found to be a critical visual cue in species recognition and mate choice of small winged insects (Shevtsova et al. 2011; Katayama et al. 2014; Hawkes et al. 2019). A recent study

showed that the optical signature of the wing interference pattern is likely to be perceived by conspecifics of odonates (Brydegaard et al. 2018), suggesting that the wing interference pattern of *P. m. dorothea* may play a role in male competition.

The findings of this study are also important in suggesting the potential role of divergent agonistic behavior in the speciation mechanism of P. mandarinus damselflies. Sexual selection via male competition for mates can be a strong driving force in the adaptive divergence and speciation of odonates (Svensson et al. 2006; Svensson and Waller 2013; Iyengar et al. 2014; Wellenreuther and Sánchez-Guillén 2016). The most closely related species of P. mandarinus is P. kuroiwae, which has hyaline wings, suggesting that P. kuroiwae may have agonistic behaviors similar to P. m. dorothea, and the colored wing and perched wing display of P. m. mandarinus is likely a derived trait in P. mandarinus damselflies. The evolutionary origin of the colored wing in P. m. mandarinus may give males with this novel trait a fitness advantage in territorial contests because they face less aggression from rivals with hyaline wings. The perched wing display may function to optimize the presentation of visual signals to males with similar wing colors. The divergence of wing traits and agonistic behavior may later facilitate reproductive isolation and subsequently lead to the speciation of P. m. dorothea and P. m. mandarinus. Further tests of these hypotheses require additional experimental and field studies.

Implication of the contest assessment strategy in *P*. *m. mandarinus*

Three major models (self-, cumulative, and mutual assessment) were proposed to clarify the assessment strategies used by the rivals in animal contests (Arnott and Elwood 2009). These models differ in how information about the resource holding potential (RHP) of rivals is obtained. In self-assessment, each contestant has only information about its own RHP. In the cumulative assessment, the contestants gather information regarding their own RHP and the cost (damage, injury) accrued from rivals. In mutual assessment, both contestants obtain information on the rival's RHP relative to their own. Assessment models can be tested by analyzing the pattern of behavior progression throughout contests (Briffa et al. 2013) and the correlation between contest duration and RHP in random and RHP-matched contests (Briffa and Elwood 2009). Because behaviors in self- and cumulative assessment are not used to compare rival's RHP but only to inflict costs on rivals, contest behaviors can progress in any direction and without phases (Payne 1998; Payne and Pagel 1996). In contrast, contestants using mutual assessment assess both their own and the other's RHP; therefore, contest behaviors are expected to proceed in phases of increasing intensity that offer more accurate information on RHP and are not expected to de-escalate to levels of the previous uninformative behaviors (Enquist et al., 1990).

The behavioral observations and sequential analyses indicate that the territorial contests in P. m. mandarinus can be characterized as few physical contacts with rare injuries, directional behavioral progression in phases, and escalation from low to high aggression with rare de-escalation (Fig. 7b). A significantly large number of behavioral transitions occurred within the same behavioral elements (in stages II and III). There was frequent behavioral matching between P. m. mandarinus males in the contests (Fig. 6). The distance between the rival males decreased as contests progressed. These patterns of behavioral characteristics and progression closely match the predictions derived from mutual assessment strategies (Payne 1998; Payne and Pagel 1997; Enquist et al. 1999; Green and Patek 2018). Therefore, our findings suggest that P. m. mandarinus damselflies engage in mutual assessment of rivals in territorial contests. Earlier studies of odonate contests showed that they can use self-assessment (energetic wars of attrition; e.g., ebony jewel-wing damselfly *Calopteryx maculata*, Calopterygidae; Marden and Waage 1990), mutual assessment (e.g., skimmer dragonfly Diastatops obscura, Libellulidae; Junior and Peixoto 2013), or both self- and mutual assessment strategies depending on contestant's RHP (red-winged damselfly Mnesarete pudica, Calopterygidae; Guillermo-Ferreira et al. 2015a, b). To further test the assessment strategy of territorial contests in P. m. mandarinus damselflies, a combined approach of integrating both tests of behavioral progression and the correlation of contest duration and RHP is needed (Taylor and Elwood RW. 2003; Briffa and Elwood 2009).

Because the agonistic display in territorial contests of animals generally conveys information about a male's RHP such as body size and strength (Andersson 1994; Arnott and Elwood 2009), the results in this study raise important questions regarding which specific aspects of wing traits and movement constitute signals of the fighting ability (i.e., RHP; Parker 1974), which may influence the outcome of male-male contests. Based on the current work, we hypothesize that wing size and coloration (black pigmentation, white patch, and pruinosity) are visual signals and indicators of male RHP in perched contests of P. m. mandarinus. Alternatively, the number and kinematics of the wing flapping and wing hitting may also be used as condition-dependent signals for males' RHP (i.e., energetic war of attrition, Marden and Waage 1990; Plaistow and Siva-Jothy 1996; Suhonen et al. 2008).

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Data and code availability All data used in this study are available on Figshare: https://figshare.com/s/6be8e699b8d09097fda9; https://doi. org/10.6084/m9.figshare.16417926.

References

- Anderson CN, Grether GF (2010) Interspecific aggression and character displacement of competitor recognition in Hetaerina damselflies. Proc R Soc London, B 277:549–555. https://doi.org/10. 1098/rspb.2009.1371
- Anderson RC, DuBois AL, Piech DK, Searcy WA, Nowicki S (2013) Male response to an aggressive visual signal, the wing wave display, in swamp sparrows. Behav Ecol Sociobiol 67(4):593–600. https://doi.org/10.1007/s00265-013-1478-9
- Andersson M (1994) Sexual Selection. Princeton University Press, Princeton (NJ)
- Andrew RJ (1961) The displays given by passerines in courtship and reproductive fighting: a review. Ibis 103(4):549–579. https://doi. org/10.1111/j.1474-919X.1961.tb02452.x
- Arnott G, Elwood RW (2009) Assessment of fighting ability in animal contests. Anim Behav 77:991–1004. https://doi.org/10.1016/j. anbehav.2009.02.010
- Benelli G (2014) Aggression in Tephritidae flies: where, when, why? Future directions for research in integrated pest management. Insects 6(1):38–53. https://doi.org/10.3390/insects6010038
- Briffa M, Elwood RW (2009) Difficulties remain in distinguishing between mutual and self-assessment in animal contests. Anim Behav 77:759–762. https://doi.org/10.1016/j.anbehav.2008.11.010
- Briffa M, Hardy ICW, Gammell MP, Jennings DJ, Clarke DD, Goubault M (2013) Analysis of animal contest data. In: Hardy ICW, Briffa M (eds) Animal contests. Cambridge University Press, Cambridge, UK, pp 47–85
- Brydegaard M, Jansson S, Schulz M, Runemark A (2018) Can the narrow red bands of dragonflies be used to perceive wing interference patterns? Ecol Evol 8(11):5369–5384. https://doi.org/10. 1002/ece3.4054
- Caro T, Sherman PW (2012) Vanishing behaviors. Conserv Lett 5(3):159–166. https://doi.org/10.1111/j.1755-263X.2012.00224.x
- Corbet P (1999) Dragonflies, behavior and ecology of Odonata. Cornell University Press, Ithaca, NY
- Chen ZY, Hsu Y, Lin CP (2020) Allometry and fighting behaviour of a dimorphic stag beetle Cyclommatus mniszechi (Coleoptera: Lucanidae). Insects 11(2):81. https://doi.org/10.3390/insects11020081
- Cordero-Rivera A (2017) Behavioral diversity (ethodiversity): a neglected level in the study of biodiversity. Front Ecol Evol 5:7. https://doi.org/10.3389/fevo.2017.00007
- Cordero-Rivera A, Zhang HM (2018) Ethological uniqueness of a damselfly with no near relatives the relevance of behaviour as part of biodiversity. Anim Biodivers Conserv 41(1):161–174. https://doi. org/10.32800/abc.2018.41.0161
- Córdoba-Aguilar A (Ed.) (2008). Dragonflies and damselflies: model organisms for ecological and evolutionary research. OUP Oxford
- Córdoba-Aguilar A, Cordero-Rivera A (2005) Evolution and ecology of Calopterygidae (Zygoptera: Odonata): status of knowledge and research perspectives. Neotrop Entomol 34(6):861–879. https:// doi.org/10.1590/S1519-566X2005000600001

- Dijkstra PD, Border SE (2018) How does male-male competition generate negative frequency-dependent selection and disruptive selection during speciation? Curr Zool 64(1):89–99. https://doi. org/10.1093/cz/zox079
- Enquist M, Leimar O, Ljungberg T, Mallner Y, Segerdahl N (1990) A test of the sequential assessment game: fighting in the cichlid fish Nannacara anomala. Anim Behav 40:1–14. https://doi.org/ 10.1016/s0003-3472(05)80660-8
- Green PA, Patek SN (2018) Mutual assessment during ritualized fighting in mantis shrimp (Stomatopoda). Proc R Soc London, B 285:20172542. https://doi.org/10.1098/rspb.2017.2542
- Grether GF, Anderson CN, Drury JP, Kirschel AN, Losin N, Okamoto K, Peiman KS (2013) The evolutionary consequences of interspecific aggression. Ann NY Acad Sci 1289:48–68. https:// doi.org/10.1111/nyas.12082
- Guillermo-Ferreira R, Bispo PC, Appel E, Kovalev A, Gorb SN (2015) Mechanism of the wing colouration in the dragonfly Zenithoptera lanei (Odonata: Libellulidae) and its role in intraspecific communication. J Insect Physiol 81:129–136. https://doi.org/10.1016/j.jinsphys.2015.07.010
- Guillermo-Ferreira R, Gorb SN, Appel E, Kovalev A, Bispo PC (2015) Variable assessment of wing colouration in aerial contests of the red-winged damselfly Mnesarete pudica (Zygoptera, Calopterygidae). Sci Nat 102(3–4):13. https://doi.org/10.1007/ s00114-015-1261-z
- Guillermo-Ferreira R, Bispo PC, Appel E, Kovalev A, Gorb SN (2019) Structural coloration predicts the outcome of male contests in the Amazonian damselfly Chalcopteryx scintillans (Odonata: Polythoridae). Arthropod Struct Dev 53:100884. https://doi.org/10.1016/j.asd.2019.100884
- Günther A, Hilfert-Rüppell D, Rüppell G (2014) Reproductive behaviour and the system of signalling in Neurobasis chinensis (Odonata, Calopterygidae) – a kinematic analysis. Int J Odonatol 17(1):31–52. https://doi.org/10.1080/13887890.2014.881305
- Hawkes MF, Duffy E, Joag R, Skeats A, Radwan J, Wedell N, Sharma MD, Hosken DJ, Troscianko J (2019) Sexual selection drives the evolution of male wing interference patterns. Proc R Soc London, B 286:20182850. https://doi.org/10.1098/rspb. 2018.2850
- Hilfert-Rüppell D, Rüppell G (2013) Do coloured-winged damselflies and dragonflies have flight kinematics different from those with clear wings? Int J Odonat 16(2):119–134. https://doi.org/10.1080/ 13887890.2013.763332
- Iyengar VK, Castle T, Mullen SP (2014) Sympatric sexual signal divergence among North American Calopteryx damselflies is correlated with increased intra- and interspecific male-male aggression. Behav Ecol Sociobiol 68:275–282. https://doi.org/10.1007/ s00265-013-1642-2
- Junior RSL, Peixoto PEC (2013) Males of the dragonfly Diastatops obscura fight according to predictions from game theory models. Anim Behav 85(3):663–669. https://doi.org/10.1016/j.anbehav. 2012.12.033
- Katayama N, Abbott JK, Kjærandsen J, Takahashi Y, Svensson EI (2014) Sexual selection on wing interference patterns in Drosophila melanogaster. Proc Natl Acad Sci USA 111(42):15144–15148. https://doi.org/10.1073/pnas.1407595111
- Lackey A, Martin MD, Tinghitella RM (2018) Male competition and speciation: expanding our framework for speciation by sexual selection. Curr Zool 64(1):69–73. https://doi.org/10.1093/cz/ zoy009
- Lin S-C, Chen Y-F, Shieh S-H, Yang P-S (2014) A revision of the status of Psolodesmus mandarinus based on molecular and morphological evidence (Odonata: Calopterygidae). Odonatologica 43(1):51–66
- Lin S-C, Chen Y-F, Shieh S-H, Yang P-S (2012) Patterns of mitochondrial and wing morphological differentiation in Taiwanese

populations of Psolodesmus mandarinus McLachlan (Zygoptera: Calopterygidae). Odonatologica 41:109–121

- Marden JH, Waage JK (1990) Escalated damselfly territorial contests are energetic wars of attrition. Anim Behav 39:954–959. https:// doi.org/10.1016/S0003-3472(05)80960-1
- McCullough EL, Tobalske BW, Emlen DJ (2014) Structural adaptations to diverse fighting styles in sexually selected weapons. Proc Natl Acad Sci USA 111:14484–14488. https://doi.org/10.1073/ pnas.1409585111
- Mikami OK, Kohda M, Kawata M (2004) A new hypothesis for species coexistence: male-male repulsion promotes coexistence of competing species. Popul Ecol 46:213–217. https://doi.org/10. 1007/s10144-004-0189-5
- Nixon MR, Orr AG, Vukusic P (2017) Covert linear polarization signatures from brilliant white two-dimensional disordered wing structures of the phoenix damselfly. J R S Interface 14(130):20170036. https://doi.org/10.1098/rsif.2017.0036
- Orr AG, Nixon MR, Vukusic P (2017) The nature and structure of the white-reflecting underside 'scales' on the hind wing of Pseudolestes mirabilis (Odonata: Pseudolestidae). Odonatologica 46(1/2):83–97
- Pajunen, V. I. (1966). Aggressive behaviour and territoriality in a population of Calopteryx virgo L. (Odonata: Calopterygidae). Ann Zool Fenn 3 201–214. http://www.jstor.org/stable/23731277
- Parker GA (1974) Assessment strategy and evolution of fighting behavior. J Theor Biol 47:223–243. https://doi.org/10.1016/0022-5193(74)90111-8
- Payne RJH (1998) Gradually escalating fights and displays: the cumulative assessment model. Anim Behav 56:651–662. https://doi.org/ 10.1006/anbe.1998.0835
- Payne RJH, Pagel M (1996) Escalation and time costs in displays of endurance. J Theor Biol 183:185–193. https://doi.org/10.1006/ jtbi.1996.0212
- Payne RJH, Pagel M (1997) Why do animals repeat displays? Anim Behav 54:109–119. https://doi.org/10.1006/anbe.1996.0391
- Plaistow SJ, Siva-Jothy MT (1996) Energetic constraints and male mate securing tactic in the damselfly Calopteryx splendens xanthostoma (Charpentier). Proc R Soc London, B 263:1233–1238. https://doi.org/10.1098/rspb.1996.0181
- Schultz TD, Fincke OM (2009) Structural colours create a flashing cue for sexual recognition and male quality in a Neotropical giant damselfly. Funct Ecol 23:724–732. https://doi.org/10.1111/j.1365-2435.2009.01584.x
- Seehausen O, Schluter D (2004) Male-male competition and nuptialcolour displacement as a diversifying force in Lake Victoria cichlid fishes. Proc R Soc London, B 271:1345–1353. https://doi.org/ 10.1098/rspb.2004.2737

- Shevtsova E, Hansson C, Janzen DH, Kjærandsen J (2011) Stable structural color patterns displayed on transparent insect wings. Proc Natl Acad Sci USA 108(2):668–673. https://doi.org/10.1073/ pnas.1017393108
- Silsby J (2001) Dragonflies of the world. CSIRO Publishing, Collingwood, Australia
- Suhonen J, Rantala MJ, Honkavaara J (2008) Territoriality in odonates. In: Córdoba-Aguilar A (ed) Dragonflies and damselflies: model organisms for ecological and evolutionary research. Oxford University Press, New York, pp 203–217
- Svensson EI, Eroukhmanoff F, Friberg M (2006) Effects of natural and sexual selection on adaptive population divergence and premating isolation in a damselfly. Evolution 60:1242–1253. https://doi.org/ 10.1554/06-036.1
- Svensson EI, Waller JT (2013) Ecology and sexual selection: evolution of wing pigmentation in calopterygid damselflies in relation to latitude, sexual dimorphism and speciation. Am Nat 182(5):E174-195. https://doi.org/10.1086/673206
- Taylor PW, Elwood RW (2003) The mismeasure of animal contests. Anim Behav 65:1195–1202. https://doi.org/10.1006/anbe.2003. 2169
- Tinghitella RM, Lackey AC, Martin M, Dijkstra PD, Drury JP, Heathcote R, Keagy J, Scordato ES, Tyers AM (2018) On the role of male competition in speciation: a review and research agenda. Behav Ecol 29(4):783–797. https://doi.org/10.1093/beheco/ arx151
- Tsubaki Y, Hooper RE, Siva-Jothy MT (1997) Differences in adult and reproductive lifespan in the two male forms of Mnais pruinosa costalis Selys (Odonata: Calopterygidae). Res Popul Ecol 39:149–155. https://doi.org/10.1007/BF02765260
- Wellenreuther M, Sánchez-Guillén RA (2016) Nonadaptive Radiation in Damselflies. Evol Appl 9(1):103–118. https://doi.org/10.1111/ eva.12269
- West-Eberhard MJ (1983) Sexual selection, social competition, and speciation. Q Rev Biol 58:155–183. https://doi.org/10.1086/ 413215
- Xu M, Fincke OM (2015) Ultraviolet wing signal affects territorial contest outcome in a sexually dimorphic damselfly. Anim Behav 101:67–74. https://doi.org/10.1016/j.anbehav.2014.12.018

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