J Insect Behav (2025) 38:21
https://doi.org/10.1007/s10905-025-09885-y

RESEARCH

Warty Birch Caterpillar, *Falcaria bilineata* (Drepanidae): Ontogenetic Changes in Morphology and Behavior

Leonardo M. Turchen · Jayne E. Yack

Received: 10 January 2025 / Revised: 23 June 2025 / Accepted: 15 July 2025

Abstract Drepanidae caterpillars exhibit vibratory communication and variability in social behaviors within and between species, offering an excellent opportunity to test hypotheses on the function and evolution of vibratory communication and sociality in juvenile insects using comparative methods. Critical for conducting such studies is understanding morphological and behavioral traits across ontogeny. Here, we examine *Falcaria bilineata* to identify the number of developmental stages and their durations, as well as to characterize and distinguish their morphological and behavioral changes throughout development. Seven immature stages were identified: egg, five larval instars, and pupa, with a development time of ~ 45 days from egg to pupation. Five instars were confirmed by counting head capsules. Head width showed a geometric increase between instars, consistent with Dyar's rule, thus confirming its reliability as an indicator of the instar. Morphological traits in head capsule shape and tubercles also help to distinguish instars. Feeding patterns transitioned from early instar leaf skeletonization to later instar leaf cutting. No group formation was observed, suggesting a lack of sociality. All instars lay silk mats, but no shelters were observed. Caterpillars rested in an arched position. Notably, all five instars generated vibratory signals. Fifth instars construct silk cocoons and remain inside until pupation. Pupae are covered by powdery white bloom in their cocoons and may emerge in two weeks or after overwintering. Our findings advance the understanding of life history and ontogenetic changes in *F. bilineata*, laying the groundwork for comparative studies into the mechanisms of social behavior and communication in larval insects.

Keywords Larvae · Development · Immature stages · Juveniles · Life-history · Vibroacoustic

Introduction

Caterpillars of the two-lined hook tip moth, *Falcaria bilineata* (Packard 1864) (Lepidoptera: Drepanidae), commonly known as the warty birch caterpillar due to their 'warty' appearance in the late instars (Mosher 1917), are subjects of scientific interest for a few reasons. First, *F. bilineata* belongs to the family Drepanidae, which encompasses approximately 122 genera and 660 species (Minet and Scoble 1999; Kristensen et al. 2007; van Nieukerken et al. 2011). Larvae within this family exhibit intriguing inter- and intraspecific variability in social behavior (Yack et al. 2001; Bowen et al. 2008; Scott et al. 2010a,b; Guedes et al. 2012; Scott and Yack 2012; Yadav et al. 2017; Yadav and Yack 2018). Second, Drepanidae

---

L. M. Turchen · J. E. Yack (✉)
Department of Biology, Carleton University, Ottawa, ON K1S 5B6, Canada
e-mail: jayneyack@cunet.carleton.ca

L. M. Turchen
e-mail: leonardoturchen@cunet.carleton.ca

caterpillars have been reported to produce vibratory signaling on plants, which allows them to communicate with each other (Yack et al. 2001; Bowen et al. 2008; Scott et al. 2010a,b; Guedes et al. 2012; Scott and Yack 2012; Yadav et al. 2017; Yadav and Yack 2018). While vibratory sensing and communication is widespread among insects (Cocroft and Rodríguez 2005; Turchen et al. 2022; Virant-Doberlet et al. 2023), our understanding of its precise mechanisms and functions in juveniles remains limited (Yack 2016; Turchen et al. 2022). Third, larvae of many species reside on the leaf surface or in open leaf shelters (Bryner 1999), providing an ideal setting for studying natural behaviors while simultaneously recording vibratory signals. For these reasons, comparative studies on Drepanidae caterpillars, within and between species, offer unique opportunities to test hypotheses on the evolution of communication (e.g., Scott et al. 2010a) and underlying proximate mechanisms of social behavior (e.g., Yadav et al. 2020, 2022). For example, species such as the warty birch caterpillar, *F. bilineata*, that exhibit solitary behavior, can be compared to social species, such as *Drepana arcuata* Walker, 1855 (Lepidoptera: Drepanidae) to test hypotheses explaining the genetic, sensory, and behavioral mechanisms of sociality. A crucial prerequisite for such comparative studies involves detailed documentation of the developmental stages of larvae (e.g., Yadav and Yack 2018), including their morphological features for instar identification purposes, as well as their developmental time, and behavioral traits. For most Drepanidae species, this information is lacking. Our study aims to address this gap in the warty birch caterpillar.

Falcaria bilineata is widely distributed throughout deciduous woodlands of northeastern and western North America (Rose and Lindquist 1997; Powell and Opler 2009), exhibiting two broods per year in some regions (Handfield 1999). Adults are medium-sized and recognized by the double lines across the forewings and scalloped wing tips (Fig. 1a). *Betula papyrifera* Marshall (Betulaceae) is frequently reported as the primary host plant for the larvae (Handfield 1999; Wagner 2010). While previous reports have provided brief descriptions of various morphological, behavioral, and life history traits of immature stages (e.g., Dyar 1894a,b, 1895; Beutenmuller 1898; Mosher 1917; Daviault 1937; Bowen et al. 2008), these accounts often lack comprehensive information, with only one feature or stage described, and sometimes conflicting data. Notable exceptions with respect to larval ontogeny and behavior are Dyar (1894b) and Bowen et al. (2008). Specifically, Dyar (1894b) provided a more comprehensive study of instar-specific morphological descriptions of juvenile stages. Bowen et al. (2008) contributed anecdotal observations of morphological and behavioral aspects of larvae, and reported, for the first time, vibratory signaling. However, the focus was on late (4th and 5th) instars. Despite these valuable contributions, there remains a

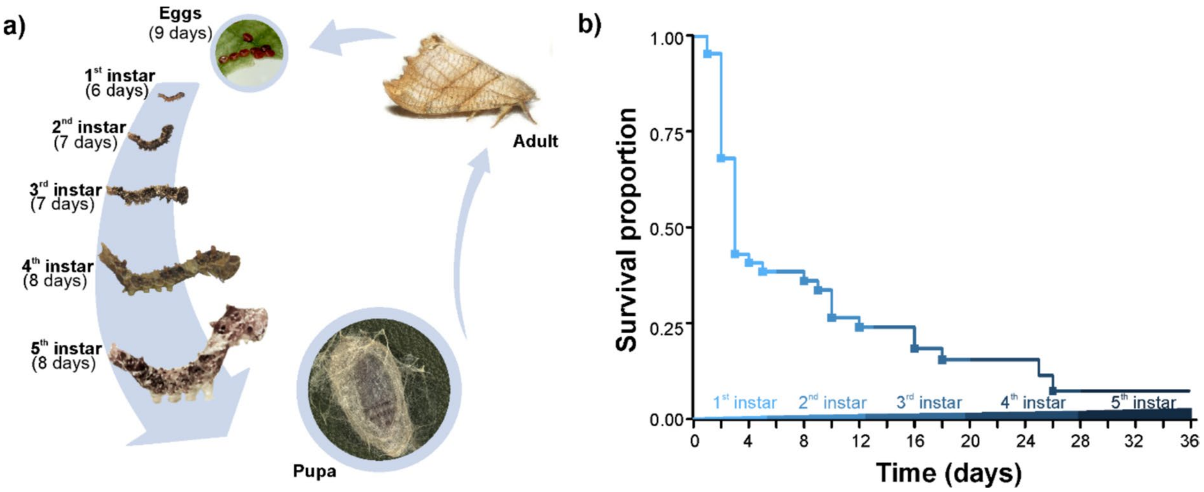

Fig. 1 Life cycle of *Falcaria bilineata*: (**a**) Illustration of the general life cycle stages. (**b**) Proportion of caterpillar survival over time, with the duration of each instar color-coded

need for formal documentation of instar-specific morphological and behavioral traits. Our study aims to fill critical knowledge gaps by documenting the number and duration of each immature stage, delineating morphological criteria for distinguishing between instars, and through careful documentation of behavior using video and vibratory recordings.

Materials and Methods

Insect Rearing and Plant Material

Adults of two-lined hook tip moths (*F. bilineata*) were collected at the Queen's University Biology Station (Chaffey's Lock, ON, Canada, 44.5788° N, 76.3195° W) between May and June 2023, using a combination of mercury vapor lights bulbs, ultraviolet lights, and LepiLED Maxi Switch equipped with four LEDs with UV radiation emittance (peak at 365 nm), two in the blue range (peak at 450 nm), one in the green range (peak at 520 nm), and one cool-white LED (Brehm 2017). Gravid females were held in glass jars (5L) where they deposited eggs on paper birch cuttings or brown paper bag clippings. To document the egg-laying dates and monitor the time of egg development, females were transferred to new jars every 24 h. Eggs were collected from jars, transferred to Petri dishes, and monitored daily until the day of hatching. On the day of hatching, neonates were used for experiments. Paper birch (*B. papyrifera*), a known host plant of the warty birch caterpillar (Handfield 1999), was the plant species utilized in rearing and experiments. Fresh twig cuttings were gathered daily from trees near Carleton University, Ottawa, ON, Canada (45.3896° N, 75.7014° W), and the twig cuttings were utilized for rearing and in the trials.

Tracking Individuals across Ontogeny

To determine the number and duration of stages, individuals were tracked from hatching to pupation following the methodology outlined by Yadav and Yack (2018) with some modifications. Birch cuttings containing a leaf were inserted into the lids of water-filled plastic vials, and the bases of the cuttings where they were inserted into the vial were sealed using reusable adhesive putty (Staples®). Each cutting with vial was then enclosed within 1.5L plastic containers (13.8 cm diameter × 13 cm height) lined with moistened paper towels to maintain humidity levels. Subsequently, one neonate larva was carefully transferred to the center of the leaf in a birch cutting. This procedure was replicated for a total of 52 larvae. Neonate larvae were observed hourly during the first 4 h to document behavioral activities such as wandering, feeding, and silk deposition. A caterpillar was considered "established" by the amount of time it spent in one leaf location and the presence of a feeding scar and silk mat. Once established, the caterpillar's position on the leaf (base, middle, lateral edge, or tip) and the side of the leaf (abaxial or adaxial) was recorded. Daily inspections were conducted to collect head capsules, capture photographs, monitor feeding and shelter-building activities, and refresh food supplies. Two representative individuals of each developmental instar with less than 24 h following molting were preserved in 80% alcohol for further morphological examination. Data collected from individuals tracked from hatching to pupation were used to document the life cycle of *F. bilineata,* and to assess each instar's survival and duration. Survival of individuals and median duration of each instar were computed using the Kaplan–Meier estimators using the 'survival' package in R.

Morphology

Morphological features of the head capsule, setae, and tubercles were observed from live and ethanol-preserved individuals, using photographs [light and scanning electron micrographs] and shed head capsules. Live individuals from each stage were examined within 24 h of molting, and the final larval instar was examined for an additional time period to document the pre-pupal stage. In addition to counting the number of shed head capsules by following individuals, we also measured the head capsule width and body length parameters. Shed head capsules were measured across the widest part for each larval stage following Dyar (1890). As head capsules were deformed following ecdysis from the last instar, these measurements were taken directly from photographs of live larvae. Body length was measured from the head to the end of the terminal abdominal segment (A10) in a dorsal view. Head capsule width and body length measurements across larval instars served a dual purpose: (1) to confirm if parameters follow a predicted rate across instars, and (2) to determine if they are useful for distinguishing between larval stages. To achieve this, the natural log of the head capsule width and body length for each instar was plotted against the number of instars, and a linear regression analysis was conducted to assess if larvae followed a regular geometric growth progression (Dyar 1890).

Photographs were captured using a stereo microscope (Leica M205 C, Leica Microsystems, Germany) equipped with a camera (Leica DMC4500, Leica Microsystems, Germany), and a Canon camera coupled with a 24 mm lens (EOS Rebel T6, Canon, Japan). Measurements and *z*-stacked images were acquired using Leica application suite X (LAS X, version: 3.7.1.21655, Leica Microsystems, Germany). Scanning electron micrographs (SEM) were generated by air-drying specimens, mounting on aluminum stubs with carbon tape, and sputter-coating with gold–palladium. Specimens were examined using a Tesca Vega-II XMU scanning electron microscope (XMU VPSEM; Czech Republic). Identification and naming of morphological traits followed the nomenclature of Stehr (1987).

Behaviors

Behaviors of *F. bilineata* caterpillars were systematically monitored to describe the following traits: resting, crawling, feeding patterns, silk deposition, vibratory signaling, frass deposition, and propensity for social grouping. Two experimental approaches were used. In the first experimental approach, we recorded established individuals using high-definition audio–video recordings using a camcorder (Canon XA11, Canon Corp. Japan) with a macro close-up lens (Raynox, DCR-150, Yoshida Industry Co., Japan) coupled with a laser-Doppler vibrometer (PVD-100; Polytec, Inc., Germany) to record leaf vibrations simultaneously. The laser was set at a velocity of 20 mm/s; high pass filters off; low pass filter 20 kHz and the laser beam focused on reflective tape (5.5 mm diameter) affixed to the upper (adaxial) leaf surface 2–3 cm from the caterpillar. Undisturbed, established individuals were recorded for a minimum of 1 h during each larval instar. The audio–video recordings were conducted in an acoustic chamber (model C-14A MR, Eckel Industries Ltd., USA) containing two LED pad lights (model LG-E268C, Ledgo Technology Ltd., China), configured with a brightness level of L 050 and a color temperature of 4800 K. Recordings were always carried out between 7:00 AM and 6:00 PM at room temperature. A total of 19 h of video recordings were examined across all instars, and video files (.mov) were analyzed using the Behavioral Observation Research Interactive Software (BORIS) (Friard and Gamba 2016). Audio files were extracted from videos (waveform audio, '.wav') using the software Audacity [version 3.0.2] with the FFmpeg library (Audacity Team 2024). Audio files (.wav) were imported to R-software [version 4.3.3] run in the RStudio interface [version 2023.12.1+402] (R Core Team 2024; RStudio Team 2024) and waveforms were generated using the 'seewave' package. Graphical illustrations were produced with Wacom creative tablet (Intuos S, Tokyo, Japan) using Corel Draw 2021 (Corel, Ottawa, ON, Canada). In addition to conducting video recordings, each day we observed live specimens for approximately 10 min per individual, documenting their behavior through manual notes and photographs (EOS Rebel T6 coupled with a Canon EF-S 24 mm STM Lens; Canon, Japan).

A second experiment investigated the potential for group formation among early instars. The rationale for focusing on early instars is based on observations in other Drepanidae species, such as *D. arcuata*, where early instars form groups actively, whereas later instars live solitarily (Yack et al. 2001; Yadav et al. 2017). In contrast, *F. bilineata* seems to differ in their potential to form groups (Bowen et al. 2008), although this has not been formally tested. The experimental design included the following: one birch cutting containing three leaves was placed in a water-filled plastic vial, as described above. Six early instar (I and II) individuals were randomly selected from rearing jars and positioned on the center of one of the three leaves, and then allowed to move freely on the birch cutting. The larvae were monitored at hourly intervals (1, 2, 3, 4, 5, 6, and 7 h) to note whether group formations occurred. Three experimental trials were conducted per day over a period of three days, totaling nine replicas.

Results

Falcaria bilineata develops through seven distinct immature stages: eggs, five larval instars, and pupa (Fig. 1a). Morphological and behavioral traits (where applicable) for each stage are described below.

General Comments on Immature Stages and Ontogeny Tracking

Overall, the developmental time from hatching to pupation ranged from 34–45 days, with an average duration of 35.6 ± 3.04 days (Fig. 1a). To follow the ontogeny, 52 first instars were initially tracked. However, not all these individuals were followed through to pupation, for two reasons. First, high mortality occurred in early instars (e.g., ~ 56% of individuals died in the first instar [N = 29/52 individuals], with additional mortality at subsequent stages (Fig. 1b)). Second, two representatives per instar (10 individuals in total) were preserved in 80% ethanol for morphological analysis. Due to these factors, the number of individuals studied at each developmental stage declined throughout the study. Consequently, the maximum sample sizes available for study for each instar are as follows: 52 first instars, 21 second instars, 16 third instars, 10 fourth instars, 7 fifth instars and 5 pupae. However, not all these individuals may have been examined for any particular behavioral or morphological trait. Further details on mortality per instar and sample sizes reported for different traits are provided below for each developmental stage.

Eggs

Females usually lay 2–7 eggs in a non-overlapping row (Fig. 1a), aligned along their longest diameter and often touching each other. Eggs were laid on both the upper and lower surfaces of leaves, plant twigs, brown paper bag clippings, and the sides of the glass jars.

Morphology: Eggs are smooth, polished, flattened, and oval with diameters ranging from 0.79–0.95 mm (0.88 ± 0.04 mm; N = 28 eggs). The color of fertilized eggs changed from yellow when laid, to orange (~ 4 days after hatching), reddish-brown (~ 7 days after hatching), and then to black as they neared hatching. Eggs hatch in approximately 9–11 days at room temperature (Fig. 1a). Neonates from the same row hatched at different times, with the exit holes oriented away from adjacent eggs. While synchronous hatching within the same egg mass was not observed, all individuals hatched within 24 h following the first hatching.

Body Plan of *F. bilineata* Larvae

Falcaria bilineata larvae exhibit a conserved body plan across five instars. They possess a sclerotized head with a striated surface texture (Fig. 2a-e) and six lateral stemmata arranged in a characteristic pattern: five forming a semicircle (stemmata 1–4, and 6), with the fifth being more ventral and posterior to the antenna, (with the third stemma being the largest; Fig. 3a). The rest of the body consists of 13 segments—three thoracic (T1-T3) each bearing a segmented leg with terminal claw, and ten abdominal (A1-A10) segments, four of which (i.e., A3-A6) that bear thick prolegs with crochets (Fig. 3a-f). Spiracles are located on T1 and A1-A8 (slightly larger on T1, A7 and A8). The terminal segment (A10) lacks prolegs (Fig. 3a-f), which is common in other Lepidoptera (Stehr 1987) and forms a suranal plate with a conical projection (called a "knob" or "process" by Stehr 1987) covered with spinules in the cuticle, and two setae emerging from the tip (Fig. 3). All instars possess on their anal segment a pair of PP1 seta (the paraproct seta Stehr 1987) that are thickened and oar-shaped (see Fig. 3g-j). These structures are used in producing vibrations and have been termed 'anal oars' in this context in this species and other Drepanidae (Yack et al. 2001; Bowen et al. 2008; Scott et al. 2010a). The body of both early instars is rusty brown with white bands, then transitioning to a creamy color with irregular brown and white patches in later instars (Figs. 1a; 4a). The body surface is somewhat rough with small mound-like projections of the body wall, frequently bearing a seta (i.e., named "tubercles" by Stehr 1987). The most prominent setae include two dorsal (D1, D2) and subdorsal (SD1, SD2) setae, and three lateral (L1, L2, L3) and subventral (SV1, SV2, SV3) setae. An illustrative general map of these setae is presented in Fig. 3a, and details for each instar are described below.

First Instar

Morphology: Head capsule width ranges from 0.32–0.36 mm (0.35 ± 0.01 mm, N = 21 head capsules) (Fig. 2a, f), and the body length from 1.45–2.56 mm (2.01 ± 0.33 mm, N = 21 individuals) (Figs. 2g; 3b; 4a). The head capsule is rounded with a slightly notched dorsal margin at the epicranial suture. The head colors range from dark brown

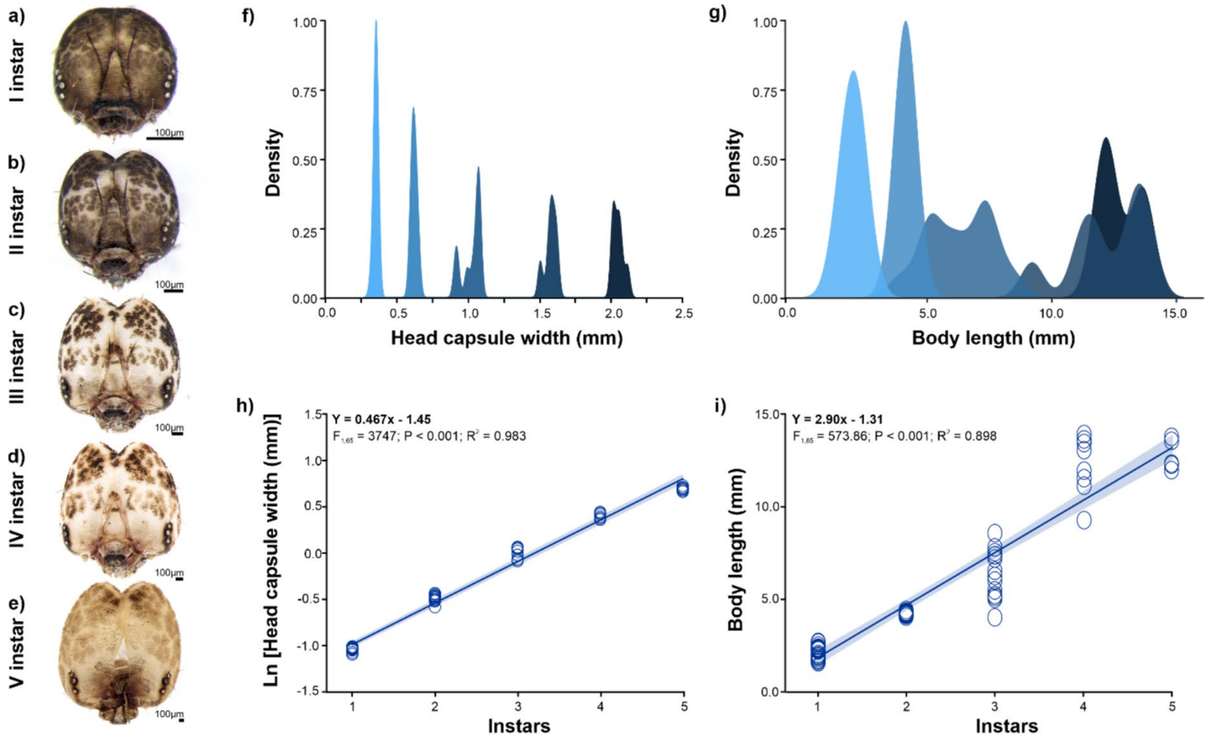

Fig. 2 Ontogeny of growth patterns in *F. bilineata* larvae. Light micrographs of *F. bilineata* head capsules at different instar stages: (**a**) first instar; (**b**) second instar; (**c**) third instar; (**d**) fourth instar; (**e**) fifth instar. Scale bar: 100 µm. Density plots showing the frequency distribution of head capsule width (mm) (**f**) and body length (mm) (**g**), color-coded by instar stages. Linear regression illustrating the growth trend in head capsule width (mm) (**h**) and body length (mm) (**i**) across the instars

to black, with a single faint white transverse band across the head positioned above the frons (Fig. 2a). The body is primarily rusty brown with conspicuous light color bands prominent on dorsal and subdorsal regions of the thoracic segment T1 and abdominal segments A1, A4, A5, A7, and A10 (Fig. 3b, 4a). Prolegs are brown, slightly lighter in color than the body. Dorsal setae (D1, D2) of thoracic and abdominal segments are located in tubercles, except A10. Dorsal tubercles are very similar in size in all segments. At the thoracic segment (T1), the pairs of tubercles containing the D1 and D2 setae are located in the prothoracic shield (Fig. 3a-b). Subdorsal setae (SD1 and SD2) are present on T1-T3, while only SD1 is visible on abdominal segments. Importantly, during the first instar, the subdorsal setae are not located in tubercles. Lateral setae (L1, L2, L3) are present in T1, but L2 and L3 are absent (or not visible) in T2-T3 thoracic segments. In abdominal segments, only L1 and L2 setae are visible, while L3 is absent or not visible. Subventral setae (SV1 and SV2) are present on segments T1, A8, A9, and A10. Only SV1 is present (or visible) in T2-A2 segments. In A3-A7, no subventral setae are visible (Fig. 3a-b). SV3 is lacking in this instar stage. The conical projection at the end of the suranal plate is brown with short spinules in the cuticle, and two small setae at its tip (Fig. 3a,g).

Behavioral observations: Neonate (first instar) larvae exhibit an initial exploratory phase where they wander across the leaf surface until they locate a suitable site to establish themselves. Our observations revealed a strong preference among first-instar larvae for the leaf tip. Once established at the leaf tip, a subset of established larvae (N = 3 individuals) was monitored using audio–video recordings for a total of five hours. They spent an average of 60.13% of the time resting (~ 36.08 min h^{-1}), 18% of the time feeding (~ 10.80 min h^{-1}), 11.97% of the time crawling (~ 7.18 min h^{-1}), 7.48% of the time laying silk (~ 4.49 min h^{-1}), and in the remaining 2.42% of the

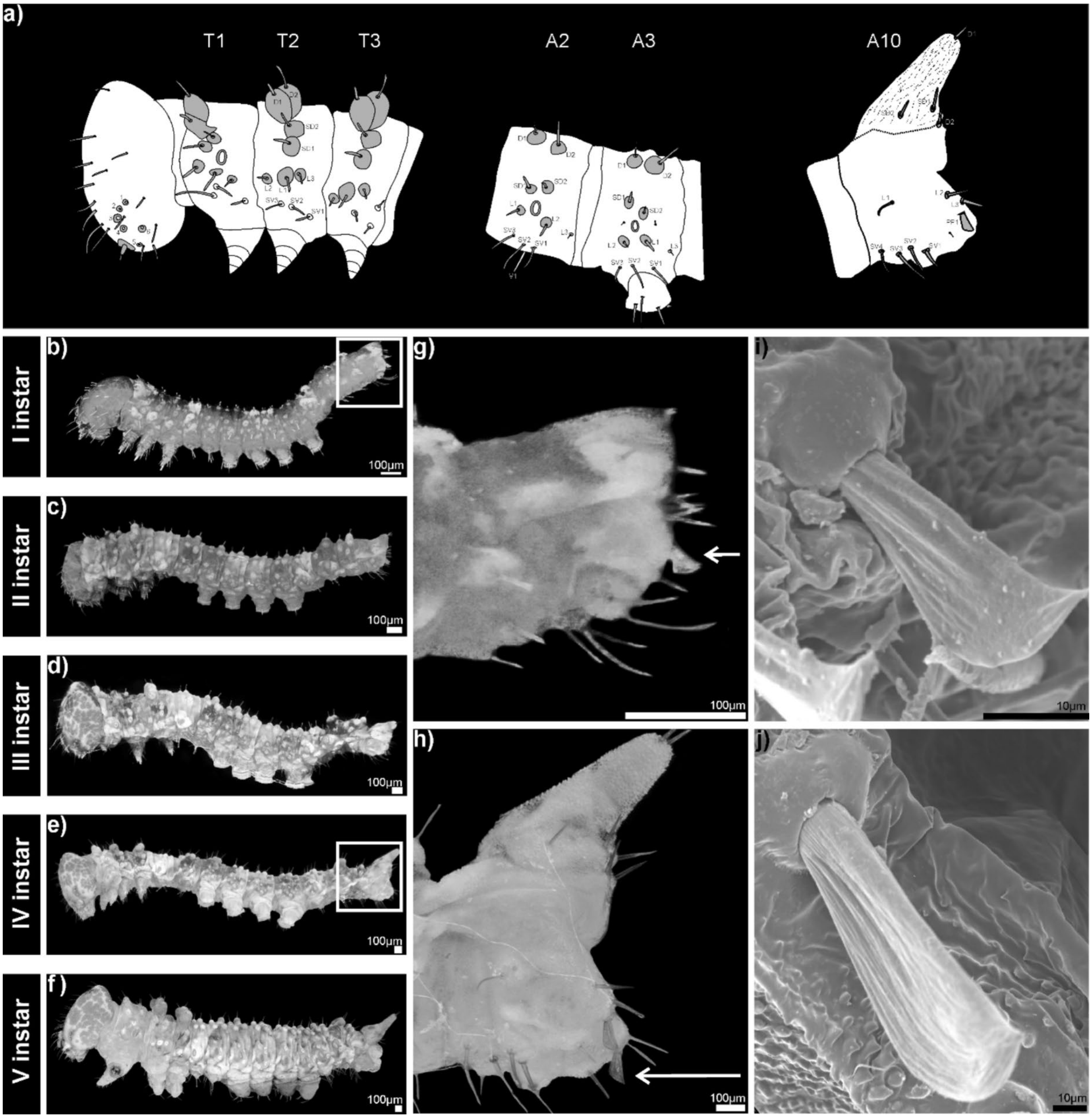

Fig. 3 Morphology traits in *F. bilineata* larvae. Body plan of *F. bilineata* larvae illustrating the map of setae in thoracic and abdominal segments (**a**). Light micrographs of *F. bilineata* body showing a lateral view of first (**b**), second (**c**), third (**d**), fourth (**e**), and fifth (**f**) instars. Scale bar: 100 µm. Close-up of the abdominal segment (A10) highlighting the suranal plate and the location of the anal oar (PP1 setae) in early (**g**) and later (**h**) instars. Scanning electron micrographs of *F. bilineata* anal-oar (PP1 setae) in early (**i**) and later (**j**) instars. Scale bar: 10 µm

time (~ 1.45 min h^{-1}) they produced occasional point events, such as signaling (n = 402 signals) and frass disposal (n = 2 frass events). Behavioral transitions were concentrated mainly between Resting – Crawling – Feeding. Feeding involves skeletonizing the leaf by consuming the area between veins and trichomes (Fig. 4c). Larvae tend to feed away from the leaf tip (~ 1 cm from the extreme leaf tip), leaving the tip unconsumed (Fig. 4c). The feeding scar tends to extend in the opposite direction from the leaf tip,

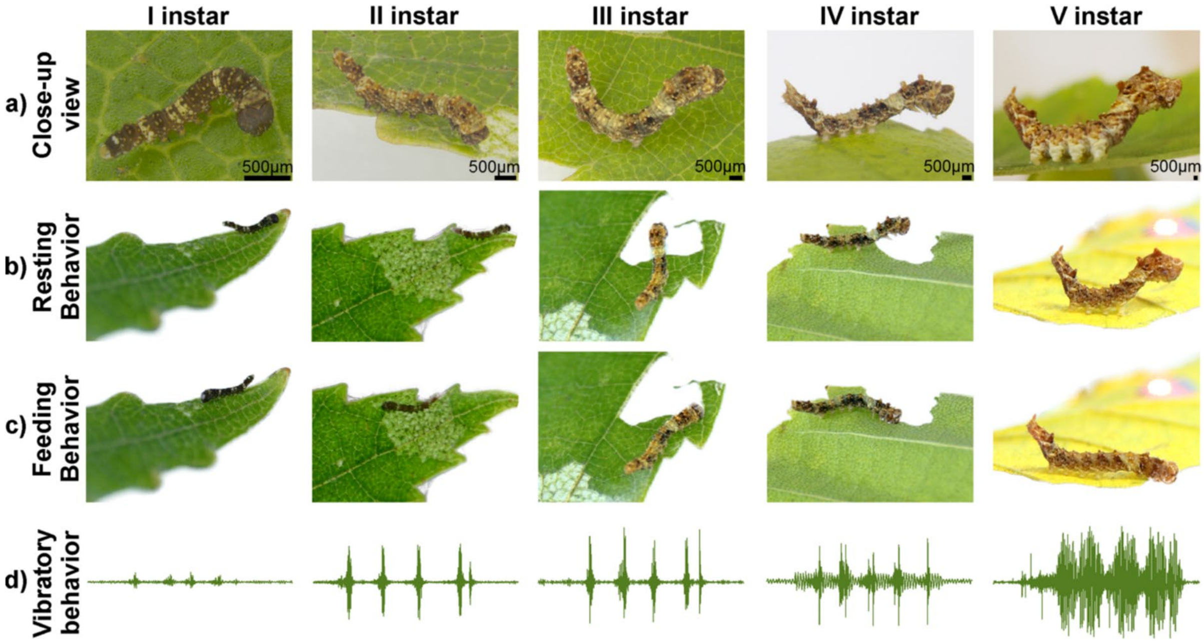

Fig. 4 Photographs of *F. bilineata* showing a close-up view of the caterpillar on its natural substrate (**a**), along with characteristic behaviors of resting (**b**) and feeding (**c**). Waveforms (five seconds) at the bottom show vibrational signals generated by rapid head and body movements (**d**). Each column represents an instar (I-V), with early instars on the left and later instars on the right

progressing laterally across the scars. Larvae also lay silk on the leaf surface. This process involves repetitive back-and-forth movements of the anterior body, with a silk thread emerging near the mouth from silk-producing glands. Each movement lays down another strand, gradually forming a silk mat on the leaf surface. Neonates frequently lay silk at the tip (as a silk mat) and edges of the leaf but do not construct tents or shelters. Notably, first-instar larvae exhibit a preference for resting at the leaf tip (Fig. 4b). During rest, larvae maintain a characteristic posture, with their anterior (Head, T1-A2) and terminal abdominal segments (A7-A10) held up at a ~ 45-degree angle, supported by their abdominal prolegs (A3-A6) on the silk mat (Fig. 4b). While resting, larvae deposit frass away from the leaf by turning the anal part outward towards the leaf edge and dropping frass away from the edge. If frass drops on the leaf, the caterpillar removes it by flicking it over the edge with its terminal abdomen or picking it up in their mandibles, walking to the edge, and dropping it. Larvae also generate rapid head and body movements, creating leaf vibrations with a high signal-to-noise ratio (Fig. 4d). Signaling in solitary established individuals occurs 1.46 events per minute and primarily happens at the leaf tip (75% of cases; n = 302/402 events; N = 3 individuals). However, it can also occur at the feeding scar (12% of cases n = 48/402 events; N = 3 individuals) or leaf edge (13% of cases; n = 52/402 events; N = 3 individuals). Crawling is primarily observed when the larvae move between the leaf tip and their feeding site, or along the leaf edge while laying down silk.

Development time: Of the 52 individuals hatched from eggs, 29 died during the first instar and 2 were preserved, resulting in 21 progressing to the second instar. The duration of the first instar is 5–9 days with an average of 6.57 ± 0.98 days (N = 21 individuals) (Fig. 1b). The molting process occurs at the leaf tip, commonly where silk is present (95.24% N = 20 individuals).

Second Instar

Morphology: Head capsule width ranges from 0.60–0.65 mm (0.62 ± 0.02 mm, N = 19 head capsules) (Fig. 2b, f), and the body length from 3.94–4.20 mm (4.13 ± 0.08 mm, N = 19 individuals) (Figs. 2g; 3c; 4a). The head capsule becomes bilobed with the notched dorsal margin at the epicranial suture more pronounced in comparison with the first instar. The head color is dark brown, but the white transverse band above the frons is more pronounced in comparison with the first instar (Fig. 2b). The body color is lighter rusty brown compared with the first instar, and the light color bands follow the same pattern as the first instar. Prolegs are dark brown, slightly darker than the body. Dorsal setae (D1, D2) of thoracic and abdominal segments are located in tubercles, except A10. Prominent dorsal tubercles are present in the first three thoracic segments (T1-T3) and specific abdominal segments (A2, A3, A8, and A9) (Fig. 3a,c). Subdorsal setae (SD1 and SD2) are present on both thoracic and abdominal segments, except A9 where SD2 is absent. On the thoracic segments, the subdorsal setae are present in tubercle projections, while on the abdominal segments, they are simple setae. Lateral setae (L1 and L2) are present in the thoracic and abdominal segments, but L3 is absent (or not visible) in segments T2-T3 and shortened in the abdominal segments. Subventral setae (SV1, SV2, and SV3) are present in the thoracic and abdominal segments (Fig. 3a,c). The conical projection at the end of the suranal plate is more prominent than in the first instar. This projection is light brown at the base with spinules in the cuticle transitioning to black near the tip where two small setae are present (Figs. 3c; 4a).

Behavioral observations: Following the molt from the first instar, larvae consume their exoskeleton in > 90% of the cases, while the head capsule is often on the leaf surface or the floor of the plastic rearing container. Behavioral patterns are quite similar to those observed in the first instar. Larvae (N = 4 individuals) were monitored using audio–video recordings for a total of five hours. They spend on average 55.01% of the time resting (~ 33.01 min h^{-1}), 37.33% of the time feeding (~ 22.40 min h^{-1}), 5.45% of the time crawling (~ 3.27 min h^{-1}), 0.41% of the time laying silk (~ 0.25 min h^{-1}) and in the remaining 1.80% of the time (~ 1.08 min h^{-1}) they produce occasional point events, such as signaling (n = 256 signals; N = 4 individuals) and frass disposal (n = 14 frass events; N = 4 individuals). Feeding behavior and resting posture observed in the first instar are also seen in the second instar (Fig. 4b,c). Larvae continue extending the same first instar feeding scar by skeletonizing the leaf and depositing silk on the leaf surface and edges (Fig. 4c). Signaling in solitary established individuals occurs 0.80 events per minute (Fig. 4d) and primarily at the extreme leaf tip (67% of cases; n = 171/256 events; N = 4 individuals) and feeding scar (33% of cases; n = 85/256 events; N = 4 individuals). Crawling is primarily observed when the larvae move between the leaf tip and their feeding site.

Development time: Of the 21 individuals that reached the second instar, 2 were preserved and 3 died, resulting in 16 individuals progressing to the third instar. The duration of the second instar is 5–8 days with an average developmental time of 6.13 ± 0.81 days (N = 16 individuals) (Fig. 1b). The molting process occurs at the leaf tip, commonly where silk is present (100%; N = 16 individuals).

Third Instar

Morphology: Head capsule width ranges from 0.90–1.08 mm (1.02 ± 0.07 mm, N = 14 head capsules) (Fig. 2c, f), and the body length from 3.90–8.50 mm (6.30 ± 1.32 mm, N = 14 individuals) (Figs. 2g; 3d; 4a). The head capsule is bilobed with the notched dorsal margin at the epicranial suture, as in the second instar. The head is yellowish in color with one white transverse band above the frons (as in the first and second instar) and an additional (i.e., not present in the first and second instar) white band appearing across the frons. Stemmata are surrounded by brown-black markings (Fig. 2c). The body color is yellowish-brown and light-colored dorsal bands follow the pattern of earlier instars (Fig. 3d, 4a). Prolegs are light brown following the same body color. Dorsal (D1, D2) and subdorsal setae (SD1, SD2) occur in tubercle projections in the thoracic and abdominal segments, except A10. Dorsal tubercles on T1-T3 and A8 are particularly conspicuous (Fig. 3a,d; 4a). Additionally, all body segments display lateral (L1, L2, L3) and subventral (SV1, SV2, SV3) setae, with the third lateral setae (L3) being generally shorter. The conical projection on the terminal segment is more prominent than in the second instar, brown with spinules in the cuticle, and two setae at its tip (Fig. 3d; 4a).

Behavioral observations: Following the molt from the second instar, larvae consume their exoskeleton in > 80% of the cases, while the head capsule is often on the leaf surface or the floor of the plastic rearing container. Third instar larvae (N = 3 individuals) were monitored using audio–video recordings for a total of five hours. They spent on average 62.83% of the time resting (~37.70 min h^{-1}), 29.33% of the time feeding (~17.6 min h^{-1}), 6.13% of the time crawling (~3.68 min h^{-1}), and in the remaining 1.71% of the time (~1.03 min h^{-1}) they produced occasional point events, such as signaling (n = 162 signals; N = 3 individuals) and frass disposal (n = 18 frass events; N = 3 individuals). Third instar larvae continue to feed by skeletonizing the leaf tissue but then transition to cutting the leaf edges (Fig. 4c). Additionally, third instar larvae move away from the tip. At this point, they no longer skeletonize the leaf and begin cutting it from the edge. After eating the entire leaf, or the leaf becomes dry, the larva moves to a new leaf and lays down a mat of silk on the upper leaf surface typically at the lateral part of the leaf near the feeding site rather than specifically at the leaf tip (Fig. 4c). Interestingly, they clip out the non-eaten parts such as the veins and petiole. The resting posture is similar to earlier instars, but the location of resting may be anywhere on the leaf, not just the tip as for previous instars (Fig. 4b). Signaling occurs at a rate of 0.52 events/minute and is primarily concentrated at the lateral part of the leaf (55% cases; n = 89/162; N = 3 individuals) and tip (41% cases; n = 66/162; N = 3 individuals) on the silk mat and a lower proportion of signals are observed near where the feeding occurs (4% of cases; n = 7/162; N = 3 individuals) (Fig. 4d).

Development time: Of the 16 individuals that reached the third instar, 2 were preserved and 4 died, resulting in 10 individuals progressing to the fourth instar. The duration of the third instar is 4–9 days with an average developmental time of 6.40 ± 1.71 days (N = 10 individuals) (Fig. 1b). The molting process occurs at the lateral part of the leaf, commonly where silk is present (100%; N = 10 individuals).

Fourth Instar

Morphology: Head capsule width ranges from 1.46–1.63 mm (1.56 ± 0.07, N = 8 head capsules) (Fig. 2d, f), and the body length from 9.21–13.91 mm (12.21 ± 1.60 mm, N = 8 individuals) (Figs. 2g; 3e; 4a). Head capsule is bilobed with the notched dorsal margin at the epicranial suture, as in the second and third instar. The head is yellowish in color with two white transverse bands, and the stemmata are surrounded by brown-black markings similar to the third instar (Fig. 2d). Body color and light color bands remain in the same pattern as the third instar (Figs. 3e; 4a). Prolegs are pale brown, distinguishing them from the yellowish-brown in the body color. Dorsal (D1, D2) and subdorsal (SD1, SD2) setae occur in tubercle projections in the thoracic and abdominal segments, except A10. Dorsal tubercles on T1-T3 and A8 are particularly conspicuous. Additionally, all body segments display lateral (L1, L2, L3) and subventral (SV1, SV2, SV3) setae, with the third lateral setae (L3) being generally shorter. The conical projection is more prominent than in the third instar, being light brown with spinules in the cuticle, and has two setae at its tip (Figs. 3a, d; 4a).

Behavioral observations: Following the molt from the third instar, larvae consume their exoskeleton in 80% of the cases, while the head capsule is often on the leaf surface or the floor of the plastic rearing container. Fourth instar larvae (N = 4 individuals) were monitored using audio–video recordings for a total of three hours. They alternated primarily between resting (55.52% of the time; [~33.31 min h^{-1}]) and feeding (43.27% of the time [~25.96 min h^{-1}]) with lower proportions of the time spent crawling (0.85% of the time [~0.51 min h^{-1}]) mainly between the silk mat and the feeding location. In the remaining 0.36% of the time (~0.22 min h^{-1}) they produced occasional point events, such as signaling (n = 11 signals; N = 4 individuals) and disposing of frass (n = 14 frass events; N = 4 individuals). Larvae lay a silk mat on the leaf surface as do third instars. Like late third instars, fourth instars feed by cutting the leaf (Fig. 4c). Once the leaf is consumed, or becomes dry, a larva wanders in search of a new fresh leaf. The resting posture remains as in the earlier instars (Fig. 4b). Resting occurs most of the time on the silk mat deposited in the intact leaf area at the lateral part of the leaf near where the feeding occurs (Fig. 4b). Signaling is also observed in the fourth instar, occurring at a rate of 0.056 events per minute and predominantly at the lateral part of the leaf (100% of cases; n = 11 events; N = 4 individuals) (Fig. 4d).

Development time: Of the 10 individuals that reached the fourth instar, 2 were preserved and 1 died, resulting in 7 individuals progressing to the fifth instar. Duration of the fourth instar is 6–9 days with an average developmental time of 7.00 ± 1.15 days (N = 7 individuals) (Fig. 1b). The molting process occurs at the lateral part of the leaf, commonly where silk is present (100%; N = 7 individuals).

Fifth Instar

Morphology: Head capsule width ranges from 2.02–2.11 mm (2.06 ± 0.03 mm, N = 5 head capsules) (Fig. 2e, f), and the body length from 11.94–13.76 mm (12.76 ± 0.83 mm, N = 5 individuals) (Figs. 2g; 3f, 4a). The head capsule is bilobed with the notched dorsal margin at the epicranial suture, as in the second, third, and fourth instars. The head remains yellowish in color with stemmata surrounded by brown-black markings similar to the third and fourth instars, but the two white transverse bands are fainter (Fig. 2e). The body color is a lighter yellowish-brown compared to the third and fourth instars, and the light color bands follow the same pattern as the earliest instars (Figs. 3e; 4a). Prolegs are pale brown close to white, distinguishing them from the yellowish-brown marks in the body color. Dorsal (D1, D2) and subdorsal (SD1, SD2) setae are found in tubercle projections in the thoracic and abdominal segments, except A10. Dorsal tubercles on T2-T3, and A8 are particularly conspicuous and reddish-brown in color. In the T2 and T3 segments the dorsal tubercles seem to be merged forming a horn-like protrusion in each segment. Furthermore, all body segments display lateral (L1, L2, L3) and subventral (SV1, SV2, SV3) setae, with the third lateral setae (L3) being generally shorter. The conical projection is more prominent than in the fourth instar, light brown with spinules in the cuticle, and two setae at its tip (Figs. 3a, f; 4a). Prepupal larvae become enlarged, with the head capsule noticeably narrower than the body, and the reddish-brown color in the tubercles becoming more evident.

Behavioral observations: Following the molt from the fourth instar, larvae consume their exoskeleton in > 80% of the cases, while the head capsule is often on the leaf surface or the floor of the plastic rearing container. Fifth-instar larvae (N = 2 individuals) were monitored using audio–video recordings for a total of one hour. They allocated their time primarily to resting (36.83% of the time; [~ 22.10 min h^{-1}]), feeding (34.04% of the time [~ 20.43 min h^{-1}]), and crawling (27.74% of the time [~ 16.65 min h^{-1}]). In the remaining 1.39% of the time (~ 0.83 min h^{-1}) they produced occasional point events, such as signaling (n = 10 signals; N = 2 individuals) and frass disposal (n = 2 frass events; N = 2 individuals). Larvae lay a few strands of silk near feeding locations on the leaf surface, replicating the behavior of fourth instars. Like the late third and fourth instars, the fifth also feeds by cutting the leaf and clipping out the non-eaten parts such as the veins and petiole. Once the entire leaf is consumed, or becomes dry, the larvae wander in search of a new fresh leaf. Usually, they rest in their consistent position with the head and terminal abdominal segments held up, as in the previous instars, and most of the time on the silk near the feeding site, but also on the twig, or branch (Fig. 4b). Prior to pupation, caterpillars cease feeding and search for a suitable pupation site. Upon finding an appropriate location at the leaf, the larva spins silk strands using thick brown silk to partially fold the leaves together. Next, it constructs a thin, silk yellowish cocoon around itself, often attaching it to the leaf's underside. The leaf curls partially over the cocoon, providing some concealment or protection. The caterpillar remains inside the cocoon for 1–2 days, which is considered the pre-pupal stage. The molting process occurred inside the cocoon (100%; N = 5 individuals). Typically, the head capsule of the fifth instar and residues of their exoskeleton remained attached to the posterior end of the pupa.

Development time: Of the 7 individuals that reached the fifth instar, 2 were preserved and no mortality was recorded, resulting in 5 individuals progressing to the pupa stage. Duration of the fifth instar is 6–9 days with an average developmental time of 8.00 ± 1.22 days (N = 5 individuals). The duration of the pre-pupa ranges from 1–2 days. If pre-pupal time is considered within the fifth instar the duration of the fifth instar ranges from 8–11 days with an average developmental time of 9.60 ± 1.14 days (N = 5 individuals) (Fig. 1b).

Pupa

Morphology: Length: 11.93–13.31 mm (12.78 ± 0.51, N = 5 individuals) (Fig. 1a). The pupa has a protective outer silk cocoon around it. Initially, the pupa is a cream color but turns dark brown after a day and becomes covered with a white bloom. Similarly, the cocoon transitions from yellowish to brown over a few days.

Development time: The pupal stage is about two weeks when at room temperature but can also overwinter.

Comparisons between sizes of Larval Instars

Head capsule width and body length were measured throughout larval development, from hatching to pupation. Five head capsules were collected from each of the 5 individuals that were followed through ontogeny. The distribution of head capsule widths revealed five distinct peaks (Fig. 2f), confirming that there are five larval instars. This was further confirmed by linear regression analysis, which showed a strong positive geometric growth relationship across instars ($F_{1,65} = 3747$; $P < 0.001$; $R^2 = 0.983$; Fig. 2h), and indicating that head capsule width is a reliable indicator of the instar stage. Body length measurements were also taken within 24 h of molting for each instar. While linear regression again indicated growth across instars ($F_{1,65} = 573.86$; $P < 0.001$; $R^2 = 0.896$; Fig. 2h), there was greater overlap in body lengths between instars compared to head capsule widths (Fig. 2g). This suggests that body length is less reliable for distinguishing between instars in *F. bilineata*.

Early Instar Grouping Formation

We investigated experimentally whether early instar (I and II) caterpillars exhibit social tendencies. Our hypothesis was that if early instar caterpillars form groups, they would preferentially locate and stay near other individuals on the same leaf. Nine experimental replicates were conducted, each beginning with 6 early instars (54 individuals in total). However, no group formation was observed at any of the observation points throughout the experiment. Within the first hour, all six larvae (per trial, each involving 3 leaves) typically dispersed across the original leaf where they were placed. Throughout the remaining observation period (7 h in total), the median number of caterpillars per leaf was two. It's important to note that even when two caterpillars were on a single leaf, they did not represent a group, as they occupied different regions of the same leaf. Collectively these results indicate that early instar *F. bilineata* do not exhibit grouping behavior.

Discussion

In this study, we document the ontogeny of the warty birch caterpillar, *F. bilineata*, a member of the Drepanidae family. This family is notable for its variation in larval social behavior, territoriality, and vibratory signaling, both within and between species (see Yack et al. 2001; Bowen et al. 2008; Scott et al. 2010a; Yadav et al. 2017, 2020), offering an excellent opportunity to test hypotheses regarding the mechanisms and evolution of insect sociality using comparative methods. To carry out such comparative studies, it is important to characterize the phenotypes (morphology and behavior) of each species. Therefore, the primary objectives of this study were to describe the immature stages of *F. bilineata,* distinguish larval instars, and document the morphological and behavioral changes that occur throughout its development.

Eggs

Falcaria bilineata adults laid their eggs in non-overlapping rows, aligned with their longest diameter and often touching each other. Eggs change color during development, and the neonates hatch asynchronously through exit holes oriented away from adjacent eggs. Interestingly, similar traits have been observed in other Drepanidae that share the same ecological niches, such as *D. arcuata* (Yadav and Yack 2018). The close proximity of eggs may reduce the exposed surface area, minimizing desiccation risk (Stamp 1980). The observed color changes might function as crypsis, aposematic, and/or photoprotection against ultraviolet radiation (Guerra-Grenier 2019). The orientation of the exit holes may help reduce injury to the neonates and damage to adjacent eggs, while asynchronous hatching may minimize intraspecific competition among newly emerged larvae (Takata et al. 2013; Szasz et al. 2024). At present, these hypotheses in Drepanidae remain untested but would be interesting to explore.

Larval morphology

Five larval instars of *F. bilineata* were confirmed by following the ontogeny of isolated individuals and counting their shed head capsules. This was confirmed using Dyar's rule (Dyar 1890), which predicts that larval head size remains relatively constant within each instar and follows a regular geometric progression across successive stages. Head capsule widths in *F. bilineata* showed a strong positive geometric growth pattern across instars and were closely aligned with previous reports (Dyar 1894a, b). Body length also increased significantly across development; however, it was less reliable for distinguishing instars due to overlaps in body length between instars. Thus, head capsule width in *F. bilineata* is a reliable method for identifying larval instars, as is done in various insects (e.g., Irigaray et al. 2006; Barrionuevo and San Blas 2016), including Drepanidae caterpillars (e.g., Yadav and Yack 2018).

Subtle morphological traits also help distinguish *F. bilineata* instars. As larvae grow, the shape of the head transitions from rounded in the first instar to bilobed, with a pronounced epicranial notch from the second instar onward. The head color changes from dark brown to yellowish, and the head banding pattern changes from a single white band above the frons to two bands, with the addition of a band crossing the frons and brown-black markings surrounding the stemmata. Similarly, the body color changes from rusty brown to yellowish brown, and the prolegs lighten from brown to pale brown close to white, with the most noticeable transition in the third instar. These changes in coloration and morphology align with earlier reports for *F. bilineata* (e.g., Dyar 1894a; Beutenmuller 1898). However, it is important to acknowledge that color assessments can be subjective and influenced by external factors, such as leaf color during larval rearing (LM Turchen, personal observation). Thus, caution is advised when using coloration to distinguish between instars. The transition from dark brown in early instars to yellowish brown in later instars may increase crypticity, while the lighter color of the prolegs may enhance countershading, allowing larvae to blend more effectively into their environment and avoid predation. Additionally, these color changes could potentially support thermoregulatory functions, as lighter tones might reflect solar radiation, helping larvae maintain optimal body temperatures. This could be especially relevant for *F. bilineata,* which reside on the leaf surface without shelter protection. Although these functions are speculative for *F. bilineata*, they are consistent with functions of color variation in other caterpillar species (e.g., Solensky and Larkin 2003; Rowland et al. 2008; Rowland 2009; Wagner and Hoyt 2022; Robinson et al. 2023), warranting further investigation.

Setae and tubercles also exhibit ontogenetic changes in their presence and prominence across larval instars. Dorsal setae were consistently present in all segments and located on tubercles in the thoracic and abdominal segments (except A10) across all instars. As the larvae grow, notable changes occur in the prominence of dorsal tubercles, particularly in the second, third, and fifth instars. Subdorsal setae exhibit changes in relation to their presence and association with tubercles across the thoracic and abdominal segments during the early instars (I-II). From the third instar onward, subdorsal setae were consistently present in all thoracic and abdominal segments and located on tubercles. Lateral and subventral setae also exhibit notable changes in presence across thoracic and abdominal segments in early instars (I-II), but from the third instar, they are present in all segments, although they were never associated with tubercles. Specific functions of the tubercles in *F. bilineata* are not known; however, in other lepidopteran species, tubercles can be equipped with spines, hairs, or glands that release irritating chemicals to deter predators (Battisti et al. 2011; Caro and Ruxton 2019). There is currently no evidence suggesting that the tubercles of *F. bilineata* produce urticant substances. However, their prominent appearance, reddish-brown coloration, and horn-like morphology may serve as visual signals, possibly functioning in aposematic mimicry to convey an impression of danger to potential predators, thus reducing the likelihood of attack. Tubercles and setae have also been investigated as potential sensory structures for detecting mechanical and acoustic stimuli in other caterpillar species (Tautz 1977; 1978; Taylor and Yack 2019). However, to date, there is no evidence supporting a sensory function for tubercles. In contrast, certain setae have been identified as vibroacoustic receptors in caterpillars (Tautz 1977; 1978; Taylor and Yack 2019). Although these possibilities have not been tested for *F. bilineata*, they present interesting avenues for further study.

Larval behavior

Behavioral patterns of established *F. bilineata* larvae were monitored across all instars, focusing particularly on resting, crawling, feeding, silk deposition, frass manipulation, and vibratory signaling. An experiment was also conducted to assess the tendency of early instars (I-II) to form social groups. Overall, time budgets revealed that established caterpillars spent most of their time resting, feeding, crawling, and laying silk, with frass manipulation and vibratory signaling occurring less frequently. Some behaviors were observed to change across instars. Detailed observations of each behavior are discussed below.

Resting was the most frequent larval activity, occupying 37–62% of the total time recorded in each instar (~22—37 min h^{-1}). It is important to note that larval activity was observed between 7:00 AM and 6:00 PM, which may have resulted in an overrepresentation of resting behavior, as many caterpillars exhibit increased activity at night (e.g., feeding activities; Suszczynska et al. 2017)—periods not covered in this study. Despite this limitation, our results showed that larvae from the first to fourth instars consistently spent more than 55% of their time resting (> 33 min. h^{-1}), while those in the fifth instar rested only 36% of the time (~22 min h^{-1}) and engaged in other activities (e.g., crawling). Early instars (I-II) typically rested at the extreme leaf tip, often on silk mats, whereas from the third instar onward, resting locations became more variable but remained closely associated with silk mats. Notably, all instars assumed a "dragon-like" posture during resting, with their anterior and terminal abdominal segments held at approximately a 45-degree angle—a posture also described as "dragonesque" (Wagner 2010). We suspect that this distinctive posture, combined with the selection of resting sites, may enhance camouflage in *F. bilineata* larvae. Camouflage in caterpillars is a well-documented anti-predator defense strategy, often involving background matching and specific body orientations (Stevens and Ruxton 2019). Examples include caterpillars that mimic bird droppings (Suzuki and Sakurai 2015) or adopt twig-like postures (Rowland et al. 2020) to avoid predation. Further research is needed to confirm whether the"dragon-like"posture in *F. bilineata* larvae serves a similar function.

Feeding was the second most frequent activity, occupying 18–43% of the total time recorded in each instar (~11–26 min·h^{-1}). Our findings showed a clear ontogenetic change in feeding style in *F. bilineata* from skeletonizing in early instars (I-II) to cutting in late instars (III-V). This change in feeding pattern aligns with documented changes in several Lepidoptera species (Hochuli 2001), including other Drepanidae species, such as *D. arcuata* (Yadav and Yack 2018). Such behavioral change likely reflects a combination of non-exclusive morphological and physiological factors. For instance, as caterpillars grow, small mandibles are replaced by larger, more robust ones that are better suited to cutting (Dockter 1993; Hochuli 2001). Additionally, changes in nutritional needs may influence this transition, with early instars targeting nutrient-rich soft tissues while later instars consume larger portions of the leaf for broader nutritional gains (Hochuli 2001). Early instars may also be more vulnerable to plant defenses and benefit from a more selective feeding strategy, targeting areas between veins and trichomes to avoid triggering plant defenses, while later instars may be more tolerant to defenses and consume whole leaves (e.g., Kariyat et al. 2018).

Crawling was also a common larval activity, particularly associated with short-distance movements between resting and feeding sites, occupying 1–27% of the total time recorded in each instar (~1–16 min h^{-1}). This behavior varied across developmental stages. Moderate crawling activity was observed in first instar larvae (7.18 min·h^{-1}), primarily due to extra activity in silk deposition along leaf edges, and in fifth instars (16.65 min·h^{-1}), likely due to the increase of energy demands that require additional foraging. In contrast, the second, third, and fourth instars were relatively stationary (< 3 min·h^{-1}), moving between resting and feeding locations. We suspect that the low mobility of *F. bilineata* larvae across instars may help avoid detection by predators, as excessive movement can increase visibility and the likelihood of encounters with natural enemies. Additionally, this behavior may also optimize silk mat investment, as frequent relocation would incur the energetic cost of repeatedly rebuilding these structures.

Silk deposition was recorded in first instars (~4.49 min h^{-1}) and second instars (~0.25 min h^{-1}) during video recordings; however, no direct silk deposition activity was observed during the recording periods of third to fifth instars, likely because these instars had already completed their silk deposition before the recordings began. Despite this, silk deposition was confirmed to occur across all instars as documented through observations and photographs. Our results showed that early instars (I-II instars) primarily laid silk mats at the extreme tip of the leaf, with additional strands laid at the tip edges. In contrast, later instars (III-V instars) concentrated their silk mats at the lateral edges of leaves near their feeding locations. Notably, no shelter construction was observed in any instar. Silk in Lepidoptera is known to serve multiple functions. For example, many caterpillar species construct silk mats on the leaf surface, which helps reduce the likelihood of dislodgement (Scoble 1992; Loeffler 1996). These silk mats may also serve to mitigate plant defenses, such as trichomes, which present significant challenges to soft-bodied insects (Zalucki et al. 2002; Kariyat et al. 2018). Evidence suggests that silk mats enable larvae to feed efficiently on leaves with high trichome density (Rathcke and Poole 1975; Despland 2019) and provide protection by allowing larvae to grip the silk when threatened by predators (Despland and Le Huu 2007; McClure and Despland 2011). Additionally, silk is also associated with escape behavior, such as lifeline hanging, where caterpillars jump off the leaf tip but remain attached by a silk thread (Brackenbury 1996; Sugiura and Yamazaki 2006). This behavior is common in free-living species (Sugiura and Yamazaki 2006) and may serve as an alternative to constructing protective silk shelters. The presence of silk mats in all instars warrants further investigation.

Frass disposal behavior was consistent across instars of *F. bilineata*, with larvae actively removing or depositing frass away from the leaf surface. Specific functions of manipulating frass by *F. bilineata* are not known; however, we suspect that by removing frass, the larvae may reduce the likelihood of detection by parasitoids or predators and/or minimize the risk of disease by pathogens, which in the last instance could contribute to a healthier microenvironment, enhancing survival (e.g., Weiss 2003; 2006).

Our study confirmed the presence of vibratory signaling across all larval instars of *F. bilineata*. All instars also possessed anal oar setae (PP1 setae) that have been reported to generate anal scraping signals in other Drepanidae species (Yack et al. 2001; Scott et al. 2010a). The PP1 setae that have been modified for signaling in Drepanidae vary in morphology between species (see Nakajima 1970, 1972; Scott 2012) and in future studies it would be interesting to examine the relationship between PP1 morphology and vibration characteristics and transmission across different host plants. However, this was beyond the scope of our study. Also, while assessing the function of signaling was not within the scope of this study, we propose possible functions. Research on other Drepanidae, such as *D. arcuata*, has suggested that vibratory signals in early instars function for recruitment and group formation (Yadav et al. 2017) and in later instars to mediate territorial encounters (Yack et al. 2001; Guedes et al. 2012). In *F. bilineata*, vibratory signaling has been reported to function in territorial displays during conspecific encounters in later instars (Bowen et al. 2008). Our experiment which involved placing early instar larvae on the same leaf revealed no evidence of group formation. Instead, all larvae dispersed within the first hour, suggesting a lack of social aggregation. Thus, the hypothesis that early instars use vibratory signals for recruitment is unlikely in *F. bilineata*. Our results show that isolated, early instar larvae produced vibratory signals sporadically. One hypothesis explaining this behavior is that larvae signal to advertise their presence to conspecifics (i.e. territorial signaling). Alternatively, vibrations could mimic predators, such as those produced by territorial displays of some spiders (Elias et al. 2003; 2008). We recently tested the first hypothesis, investigating whether larvae use vibratory signals to advertise their presence to conspecifics. Our findings provided strong evidence supporting territorial displays involving vibroacoustic signaling in neonates (Matheson et al. 2025). In contrast, the potential role of these vibrations in predator mimicry remains an intriguing but untested possibility, warranting further investigation.

Pupae

The pupae of *F. bilineata* are encased in a brown silk cocoon and covered by a powdery white bloom, a feature also noted by Dyar (1894a). These features distinguish *F. bilineata* from other Drepanidae species, such as *D. arcuata* (e.g., Yadav and Yack 2018). While the role of silk cocoons in various Lepidoptera species is well-documented, particularly regarding their functions in predator defense, thermal insulation, and camouflage (Lyons and Cartar 1996; Danks 2004; Chen et al. 2013), the white powdery bloom, has an unclear function. Proposed functions in different insect orders for cuticular 'bloom' include anti-fungal activity and desiccation resistance (Hadley 1986), but future research is needed to test these hypotheses in *F. bilineata* pupae.

Conclusion

Our study presents a comprehensive analysis of the morphological and behavioral changes across the ontogeny of *F. bilineata*. We described seven distinct immature stages, focusing primarily on five larval instars, and provided specific traits for their differentiation based on body length, head capsule width, and other morphological features. We documented behavioral patterns across larval instars, including resting, crawling, feeding, silk deposition, frass manipulation, and vibratory signaling. Furthermore, we experimentally confirmed that early instars do not form groups. These findings offer novel insights into the development and behavior of *F. bilineata*, enhancing our understanding of this species and laying a baseline for future research on Drepanidae as models for studying the evolution and mechanisms of vibratory communication and sociality.

Acknowledgements We thank Sarah Matheson for scanning electron microscopy of PP1 setae and Shannon J. Mahony for collecting the data from the early instar grouping formation experiment. We also acknowledge Queen's University Biological Station (QUBS) for allowing us to use their facilities for moth collection.

Author Contributions Conceptualization: L.M.T., J.E.Y.; Data curation: L.M.T.; Formal analysis: L.M.T., J.E.Y.; Funding acquisition: J.E.Y.; Investigation: L.M.T., J.E.Y.; Methodology: L.M.T., J.E.Y.; Project administration: L.M.T., J.E.Y.; Software: L.M.T.; Resources: L.M.T., J.E.Y.; Supervision: J.E.Y.; Validation: L.M.T., J.E.Y.; Visualization: L.M.T.; Writing – original draft: L.M.T., J.E.Y.; Writing – review & editing: L.M.T., J.E.Y.

Funding This research was funded by the Natural Sciences and Engineering Research Council of Canada (RGPIN 2014–05947 and RGPIN-2020–07056) to JEY, and the Canadian Foundation for Innovation New Opportunities Fund to JEY.

Data Availability The data that support the findings of this study are available within the article.

Declarations

Ethical Approval All applicable international, national, and institutional guidelines for the care and use of animals were considered in the present investigation.

Conflict of interest The authors declare that there are no conflicts of interest. J.E.Y. is a member of the editorial board of the Journal of Insect Behavior; however, J.E.Y. was not involved in the peer-review process or the decision to publish this manuscript.

References

Barrionuevo MJ, San Blas G (2016) Redescription of immature stages of the soybean looper (Lepidoptera: Noctuidae: Plusiinae). Can Entomol 148:247–259. https://doi.org/10.4039/tce.2015.62

Battisti A, Holm G, Fagrell B, Larsson S (2011) Urticating hairs in arthropods: their nature and medical significance. Annu Rev Entomol 56:203–220. https://doi.org/10.1146/annurev-ento-120709-144844

Beutenmuller W (1898) *Falcaria bilineata* Packard. Bull Am Mus Nat His 10:389–340

Bowen JL, Mahony SJ, Mason AC, Yack JE (2008) Vibration-mediated territoriality in the warty birch caterpillar *Drepana bilineata*. Physiol Entomol 33:238–250. https://doi.org/10.1111/j.1365-3032.2008.00627.x

Brackenbury J (1996) Novel locomotory mechanisms in caterpillars: life-line climbing in *Epinotia abbreviana* (Tortricidae) and *Yponomeuta padella* (Yponomeutidae). Physiol Entomol 21:7–14. https://doi.org/10.1111/j.1365-3032.1996.tb00829.x

Brehm G (2017) A new LED lamp for the collection of nocturnal Lepidoptera and a spectral comparison of light-trapping lamps. Nota Lepidopterol 40:87–108. https://doi.org/10.3897/nl.40.11887

Caro T, Ruxton G (2019) Aposematism: unpacking the defences. Trends Ecol Evol 34:595–604. https://doi.org/10.1016/j.tree.2019.02.015

Chen F, Hesselberg T, Porter D, Vollrath F (2013) The impact behaviour of silk cocoons. J Exp Biol 216:2648–2657. https://doi.org/10.1242/jeb.082545

Cocroft RB, Rodríguez RL (2005) The behavioral ecology of insect vibrational communication. Bioscience 55:323–334. https://doi.org/10.1641/0006-3568(2005)055[0323:TBEOIV]2.0.CO;2

Daviault L (1937) Contribution à l'étude des insectes du bouleau. Institut de zoologie, Université de Montréal, Montréal

Despland E (2019) Caterpillars cooperate to overcome plant glandular trichome defenses. Front Ecol Evol 7:232. https://doi.org/10.3389/fevo.2019.00232

Despland E, Le Huu A (2007) Pros and cons of group living in the forest tent caterpillar: separating the roles of silk and of grouping. Entomol Exp Appl 122:181–189. https://doi.org/10.1111/j.1570-7458.2006.00512.x

Dockter DE (1993) Developmental changes and wear of larval mandibles in *Heterocampa guttivitta* and *H. subrotata* (Notodontidae). J Lepid Soc 47:32–48

Bryner R (1999) Drepanidae-Drépanidés. In: Les papillons et leur biotopes: espèces; danger qui les menacent; protection. suisse et regions limitrophe, Pro Natura. pp 447–476

Dyar HG (1890) The number of molts of lepidopterous larvae. Psyche: J Entomol 5:420–422. https://doi.org/10.1155/1890/23871

Dyar HG (1894a) Life history of *Falcaria bilineata* Packard. J New York Entomol Soc 2:108–108

Dyar HG (1894b) Family Drepanidae. *Falcaria bilineata* Packard. Proc Boston Soc Nat Hist 26:402–403

Dyar HG (1895) Notes on Drepanid larvae. J New York Entomol Soc 3:66–69

Elias DO, Kasumovic MM, Punzalan D, Andrade MC, Mason AC (2008) Assessment during aggressive contests between male jumping spiders. Anim Behav 76:901–910. https://doi.org/10.1016/j.anbehav.2008.01.032

Elias DO, Mason AC, Maddison WP, Hoy RR (2003) Seismic signals in a courting male jumping spider (Araneae: Salticidae). J Exp Biol 206:4029–4039. https://doi.org/10.1242/jeb.00634

Friard O, Gamba M (2016) BORIS: a free, versatile open-source event-logging software for video/audio coding and live observations. Methods Ecol Evol 7:1325–1330. https://doi.org/10.1111/2041-210X.12584

Guedes RNC, Matheson SM, Frei B, Smith ML, Yack JE (2012) Vibration detection and discrimination in the masked birch caterpillar (*Drepana arcuata*). J Comp Physiol A: Neuroethol Sens Neural Behav Physiol 198:325–335. https://doi.org/10.1007/s00359-012-0711-8

Guerra-Grenier E (2019) Evolutionary ecology of insect egg coloration: a review. Evol Ecol 33:1–19. https://doi.org/10.1007/s10682-018-09967-8

Hadley NF (1986) The arthropod cuticle. Sci Am 255:104–113

Handfield L (1999) Le guide des papillons du Québec. Broquet, Boucherville, Québec.

Hochuli DF (2001) Insect herbivory and ontogeny: How do growth and development influence feeding behaviour, morphology and host use? Austral Ecol 26:563–570. https://doi.org/10.1046/j.1442-9993.2001.01135.x

Irigaray FJS-C, Moreno F, Pérez-moreno I, Marco V (2006) Larval growth and the effect of temperature on head capsule size in *Lobesia botrana* (Lepidoptera: Tortricidae). Environ Entomol 35:189–193. https://doi.org/10.1603/0046-225X-35.2.189

Kariyat RR, Hardison SB, Ryan AB, Stephenson AG, Moraes CM, Mescher MC (2018) Leaf trichomes affect caterpillar feeding in an instar-specific manner. Commun Integr Biol 11:1–6. https://doi.org/10.1080/19420889.2018.1486653

Loeffler CC (1996) Caterpillar leaf folding as a defense against predation and dislodgment: staged encounters using *Dichomeris* (Gelechiidae) larvae on goldenrods. J Lepid Soc 50:245–260

Lyons B, Cartar R (1996) Functional significance of the cocoon in two arctic *Gynaephora* moth species. Proc R Soc Lond B 263:1159–1163. https://doi.org/10.1098/rspb.1996.0169

Matheson SM, Turchen LM, Mauduit E, Yack JE (2025) Buzzing boundaries: tiny caterpillars vibrate to defend leaf tip territories. J Exp Biol 228:jeb249796. https://doi.org/10.1242/jeb.249796

McClure M, Despland E (2011) Defensive responses by a social caterpillar are tailored to different predators and change with larval instar and group size. Naturwissenschaften 98:425–434. https://doi.org/10.1007/s00114-011-0788-x

Mosher E (1917) *Falcaria bilineata* Packard. Maine Agric Exp Stn 259:75–84

Nakajima H (1970) A contribution to the knowledge of the immature stages of Drepanidae occurring in Japan (Lepidoptera). Tinea 8:167–184

Nakajima H (1972) Additions to the knowledge of the immature stages of Drepanidae occurring in Japan (Lepidoptera). Tinea 9:253–256

Powell JA, Opler PA (2009) Moths of western North America. University of California Press, Los Angeles, California

Rathcke BJ, Poole RW (1975) Coevolutionary Race continues: butterfly larval adaptation to plant trichomes. Science 187:175–176. https://doi.org/10.1126/science.187.4172.175

Tautz J (1978) Reception of medium vibration by thoracal hairs of caterpillars of *Barathra brassicae* L. (Lepidoptera, Noctuidae) II. Response characteristics of the sensory cell. J. Comp. Physiol. 125, 67–77 (1978). https://doi.org/10.1007/BF00656832

Robinson ML, Weber MG, Freedman MG, Jordan E, Ashlock SR, Yonenaga J, Strauss SY (2023) Macroevolution of protective coloration across caterpillars reflects relationships with host plants. Proc R Soc B 290:20222293. https://doi.org/10.1098/rspb.2022.2293

Rose AH, Lindquist OH (1997) Insects of eastern hardwood trees. Canadian Forestry Service. Forestry Technical Report 29.

Rowland HM (2009) From Abbott Thayer to the present day: what have we learned about the function of countershading? Phil Trans R Soc B 364:519–527. https://doi.org/10.1098/rstb.2008.0261

Rowland HM, Burriss RP, Skelhorn J (2020) The antipredator benefits of postural camouflage in peppered moth caterpillars. Sci Rep 10:21654. https://doi.org/10.1038/s41598-020-78686-4

Rowland HM, Cuthill IC, Harvey IF, Speed MP, Ruxton GD (2008) Can't tell the caterpillars from the trees: countershading enhances survival in a woodland. Proc R Soc B 275:2539–2545. https://doi.org/10.1098/rspb.2008.0812

Scoble M (1992) The Lepidoptera: form, function, and diversity. Oxford University Press, London, The Natural History Museum

Scott JL, Kawahara AY, Skevington JH, Yen SH, Sami A, Smith ML, Yack JE (2010a) The evolutionary origins of ritualized acoustic signals in caterpillars. Nat Commun 1:4. https://doi.org/10.1038/ncomms1002

Scott JL, Matheson SM, Yack JE (2010b) Variation on a theme: vibrational signaling in caterpillars of the rose hook-tip moth, *Oreta rosea* . J Insect Sci 10:1–15. https://doi.org/10.1673/031.010.5401

Solensky MJ, Larkin E (2003) Temperature-induced variation in larval coloration in *Danaus plexippus* (Lepidoptera: Nymphalidae). Ann Entomol Soc Am 96:211–216. https://doi.org/10.1603/0013-8746(2003)096[0211:TVILCI]2.0.CO;2

Stamp NE (1980) Egg deposition patterns in butterflies: Why do some species cluster their eggs rather than deposit them singly? Am Nat 115:367–380. https://doi.org/10.1086/283567

Stehr FW (1987) Order Lepidoptera. In: Stehr FW (ed) Immature insects, vol 1. Kendall/Hunt. Dubuque, Iowa, pp 288–596

Stevens M, Ruxton GD (2019) The key role of behaviour in animal camouflage. Biol Rev 94:116–134. https://doi.org/10.1111/brv.12438

Sugiura S, Yamazaki K (2006) The role of silk threads as lifelines for caterpillars: pattern and significance of lifeline-climbing behaviour. Ecol Entomol 31:52–57. https://doi.org/10.1111/j.0307-6946.2006.00755.x

Suszczynska A, Kaniewska MM, Bebas P, Giebultowicz JM, Kotwica-Rolinska J (2017) Circadian regulation of caterpillar feeding and growth. J Insect Physiol 101:113–122. https://doi.org/10.1016/j.jinsphys.2017.07.009

Suzuki TN, Sakurai R (2015) Bent posture improves the protective value of bird dropping masquerading by caterpillars. Anim Behav 105:79–84. https://doi.org/10.1016/j.anbehav.2015.04.009

Szasz M, Evans KG, Juliano SA (2024) Hatching asynchrony affects intraspecific competition among larval mosquitoes. Ecol Entomol 49:905–915. https://doi.org/10.1111/een.13363

Takata M, Koyama S, Satoh T, Fugo H (2013) Asynchronous hatching and brood reduction by filial cannibalism in the burying beetle *Nicrophorus quadripunctatus*. J Ethol 31:249–254. https://doi.org/10.1007/s10164-013-0373-1

Tautz J (1977) Reception of medium vibration by thoracal hairs of caterpillars of *Barathra brassicae* L. (Lepidoptera, Noctuidae) I. Mechanical properties of the receptor hairs. J Comp Physiol 118:13–31. https://doi.org/10.1007/BF00612334

Taylor CJ, Yack JE (2019) Hearing in caterpillars of the monarch butterfly (*Danaus plexippus*). J Exp Biol 222:211862. https://doi.org/10.1242/jeb.211862

Minet J, Scoble MJ (1999) The Drepanoid/Geometroid assemblage. In: Kükenthal W (ed) Evolution, Systematics, and Biogeography. De Gruyter, Berlin, pp. 301–320. https://doi.org/10.1515/9783110804744.301

Turchen LM, Cosme L, Yack JE, Guedes RNC (2022) Bug talk trends & biases: literature survey and meta-analyses of vibratory sensing and communication in insects. Entomol Gen 42:335–348. https://doi.org/10.1127/entomologia/2022/1380

Virant-Doberlet M, Stritih-Peljhan N, Žunic-Kosi A, Polajnar J (2023) Functional diversity of vibrational signaling systems in insects. Annu Rev Entomol 68:191–210. https://doi.org/10.1146/annurev-ento-120220-095459

Wagner DL (2010) Caterpillars of Eastern North America: a guide to identification and natural history. Princeton University Press, Princeton, New Jersey. https://doi.org/10.1515/9781400834143

Wagner DL, Hoyt AC (2022) On being a caterpillar: structure, function, ecology, and behavior. In: Marquis RJ, Koptur S (eds) Caterpillars in the middle: tritrophic interactions in a changing world. Springer International Publishing, Cham, pp 11–62. https://doi.org/10.1007/978-3-030-86688-4_2

Weiss MR (2003) Good housekeeping: why do shelter-dwelling caterpillars fling their frass? Ecol Lett 6:361–370. https://doi.org/10.1046/j.1461-0248.2003.00442.x

Weiss MR (2006) Defecation behavior and ecology of insects. Ann Rev Entomol 51:635–661. https://doi.org/10.1146/annurev.ento.49.061802.123212

Yack J, Smith M, Weatherhead P (2001) Caterpillar talk: acoustically mediated territoriality in larval Lepidoptera. P N A S 98:11371–11375. https://doi.org/10.1073/pnas.191378898

Yack J (2016) Vibrational signaling. In: Pollack G, Mason A, Popper A, Fay R (eds) Insect Hearing. Springer Handbook of Auditory Research, vol 55. Springer, Cham. pp. 99–123 https://doi.org/10.1007/978-3-319-28890-1_5

Yadav C, Guedes RNC, Matheson SM, Timbers TA, Yack JE (2017) Invitation by vibration: recruitment to feeding shelters in social caterpillars. Behav Ecol Sociobiol 71:51–64. https://doi.org/10.1007/s00265-017-2280-x

Yadav C, Smith ML, Yack JE (2020) Transcriptome analysis of a social caterpillar, *Drepana arcuata*: De novo assembly, functional annotation and developmental analysis. PLOS One 15:e0234903. https://doi.org/10.1371/journal.pone.0234903

Yadav C, Yack JE (2018) Immature stages of the masked birch caterpillar, *Drepana arcuata* (Lepidoptera: Drepanidae) with comments on feeding and shelter building. J Insect Sci 18:1–9. https://doi.org/10.1093/jisesa/iey006

Yadav C, Yack JE, Smith ML (2022) Octopamine receptor gene influences social grouping in the masked birch caterpillar. BMC Res Notes 15:211–217. https://doi.org/10.1186/s13104-022-06102-3

Zalucki MP, Clarke AR, Malcolm SB (2002) Ecology and behavior of first instar larval Lepidoptera. Annu Rev Entomol 47:361–393. https://doi.org/10.1146/annurev.ento.47.091201.145220

Audacity Team (2024) Audacity®: free audio editor and recorder. Version 3.5.1 https://www.audacityteam.org/. Accessed 20 June 2024

Danks HV (2004) The roles of insect cocoons in cold conditions. Eur J Entomol 101:433–437. https://doi.org/10.14411/eje.2004.062

Kristensen NP, Scoble MJ, Karsholt O (2007) Lepidoptera phylogeny and systematics: the state of inventorying moth and butterfly diversity. Zootaxa 1668:699–747. https://doi.org/10.11646/zootaxa.1668.1.30

R Core Team (2024) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/. Accessed 20 June 2024

RStudio Team (2024) RStudio: integrated development for R. RStudio Inc. http://www.rstudio.com/. Accessed 20 June 2024

Scott JL (2012) The evolutionary origins of vibratory signals in Drepanidae caterpillars: a comparative study on morphology, phylogenetics and behaviour. PhD Thesis, Carleton University

Scott JL, Yack JE (2012) Vibratory territorial signals in caterpillars of the poplar lutestring, *Tethea or* (Lepidoptera: Drepanidae). Eur J Entomol 109:411–417. https://doi.org/10.14411/eje.2012.053

van Nieukerken EJ, Kaila L, Kitching IJ, Kristensen NP, Lees DC. 127. et al. 2011. Order Lepidoptera Linnaeus, 1758. Zootaxa 3148:212–221 https://doi.org/10.11646/zootaxa.3148.1.41