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Tiny Amphibious Insects Use Tripod Gait for Traversal on Land, Water, and Duckweed

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Synopsis Insects exhibit remarkable adaptability in their locomotive strategies in diverse environments, a crucial trait for foraging, survival, and predator avoidance. *Microvelia americana*, tiny 2–3 mm insects that adeptly walk on water surfaces, exemplify this adaptability by using the alternating tripod gait in both aquatic and terrestrial terrains. These insects commonly inhabit low-flow ponds and streams cluttered with natural debris like leaves, twigs, and duckweed. Using high-speed imaging and pose-estimation software, we analyze *M. americana* movement on water, sandpaper (simulating land), and varying duckweed densities (10%, 25%, and 50% coverage). Our results reveal *M. americana* maintain consistent joint angles and strides of their upper and hind legs across all duckweed coverages, mirroring those seen on sandpaper. *Microvelia americana* adjust the stride length of their middle legs based on the amount of duckweed present, decreasing with increased duckweed coverage and at 50% duckweed coverage, their middle legs' strides closely mimic their strides on sandpaper. Notably, *M. americana* achieve speeds up to 56 body lengths per second on the deformable surface of water, nearly double those observed on sandpaper and duckweed, which are rough, heterogeneous surfaces. This study highlights *M. americana*'s ecological adaptability, setting the stage for advancements in amphibious robotics that emulate their unique tripod gait for navigating complex terrains.

Introduction

In nature, water surfaces are seldom clear; a pond's surface is often littered with debris like fallen leaves, twigs from overhead trees, and small floating plants such as duckweed (family Lemnaceae), which can cover an entire pond's surface (Hillman 1961). This obstacle ridden surface is where neustonic insects such as water striders mainly traverse, contending with predators (Mounts 1989; Sih et al. 1990), competitors (Toubiana and Khila 2019; Watanabe et al. 2023), and the challenge of walking on water (Crumière et al. 2016; Krupa and Sih 1998). The water striders (in the order Hemiptera) are a group with much variety, consisting of species with varying preferences for water or land and of leg spans ranging from <3mm to 30mm (Tseng and Rowe 1999). Studies have extensively explored the water strider's locomotion mechanisms, which exploit surface tension using legs with dense hair coverage (Bush et al. 2007; Gao and Jiang 2004; Hurchalla and Drelich 2019; Perez Goodwyn et al. 2008). However, previous research mainly investigates water striders on clear water (Crumière et al. 2016; Hu and Bush 2010). To really understand the characteristics of locomotion in a complex environment, we will investigate a water strider that is able to traverse water, land, and other obstacles it may encounter. We will look at *Microvelia americana*, a water strider that can navigate both water and land using a single gait: the alternating tripod gait (Fig. 1C) (Bush and Hu 2006; Hu and Bush 2010). The tripod gait is well studied for insects on land, especially in ants and cockroaches (Chun et al. 2021; Humeau et al. 2019; Kram et al. 1997; Reinhardt and Blickhan 2014; Wahl et al. 2015; Weihmann et al. 2017). Most other water striders do not use the al-

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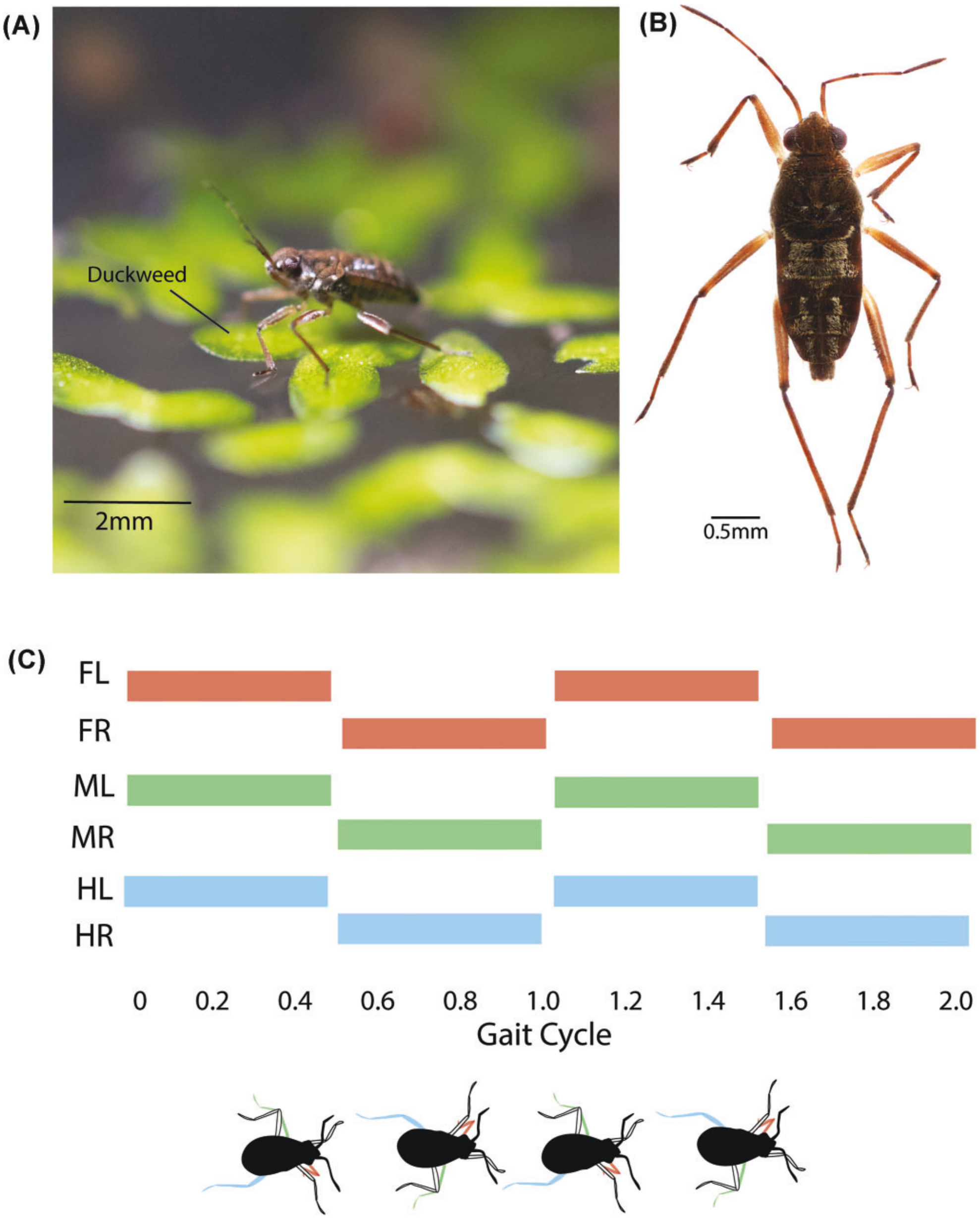

Fig. 1 *Microvelia americana* and its alternating tripod gait. (A) An *M. americana* standing on duckweed fronds. Image courtesy of Dr. Pankaj Rohilla. (B) High-resolution image of an *M. americana*. (C) Gait plot indicating the power stroke (filled rectangles) and recovery phase (blank rectangles) of the alternating tripod gait. The illustration below corresponds to the *M. americana*'s gait cycle.

ternating tripod gait—striders like Gerridae find land traversal challenging (if not impossible) due to their dependence on water contact for all legs using a specialized rowing gait (Crumière et al. 2016). Such studies on water striders have looked at land and water preference, along with stride lengths and speeds, but details of how *M. americana* adjusts its gait for different surfaces is sparse.

While *Microvelia* are not the only amphibious critters (Andersen 1976; Crumière et al. 2016; Hu and Bush 2010), it is one of the few to use just the alternating tripod gait on every surface it traverses, and one of the fastest for its body size (Crumière et al. 2016). Other arthropods, like the fishing spiders (Dolomedes), can move on both land and water but must switch between two gaits (Suter 2013; Suter et al. 1997). Some terrestrial, tropical ants adopt the alternating tripod gait for emergency water escapes but only for brief periods and with mixed success (Bohn et al. 2012). *Camponotus schmitzi* ants, in symbiosis with pitcher plants, swim in digestive fluids but only for short durations and in limited capacity (Yanoviak and Frederick 2014). While each specimen is able to sufficiently traverse their specific environments, few species can navigate across water, land,

and other surfaces as the *Microvelia* can (Crumière et al. 2016).

How the miniscule *M. americana* can accomplish this multi traversal feat using just the alternating tripod gait has intriguing implications for how we design machines which traverse in complex environments. Robotics research has applied the alternating tripod gait on complex surfaces, primarily focusing on terrestrial environments (Li et al. 2009). Uncovering how *M. americana* manages various substrates in its daily pond life, including rough surfaces such as rocks, debris, sand, water, and duckweed (Fig. 1A), is key for understanding the versatility of the alternating tripod gait that distinguishes it from other water striders and from other terrestrial arthropods. The *M. americana*'s consistent gait on different terrains opens potential for microrobots designed for robust travel across diverse landscapes in the field (Chen et al. 2018; Dallmann et al. 2023; Li et al. 2009; Song et al. 2024; Woodward and Sitti 2018).

This paper explores the multifaceted terrains *M. americana* frequently navigates, offering new avenues for alternating tripod gait research. We examine *M. americana*'s characteristics of locomotion on three different substrates: water, duckweed-covered water, and dry sandpaper to replicate locomotion on land. Duckweed on water and rocky surfaces are common amongst *M. americana*'s environment (Herring 1950; McPherson and Taylor 2006; Taylor and McPherson 2003). Utilizing high-speed video and pose estimation software, we will analyze the kinematics—body speed, stroke amplitude, and frequency—of *M. americana* as they navigate using the alternating tripod gait on each surface.

Materials and methods

Setup

We obtained *M. americana* from ponds and creeks from Kennesaw, Georgia. The specimens were kept in a 17.5 × 14.0 × 6.5-inch^3 plastic container. The container held water kept at a constant temperature of 20°C and duckweed from the insects' native bodies of water. The insects were provided with circadian lighting 12 h out of the day, from 8 A.M. to 8 P.M. Additionally, the specimens were fed once each day with fruit flies procured from Carolina Biological Monday through Friday. We examined the locomotion of the specimens on three different surfaces: water, 1000-grit aluminum oxide sandpaper from Uxcell, and water covered with duckweed. Live duckweed, *Lemna minor*, was obtained from Carolina Biological. These substrates were chosen for their presence in the *M. americana*'s natural environment and for a comparison of how general roughness and heterogeneity on a surface influences locomotion. To understand specific mechanics on different substrates, the substrates were tested as separated conditions rather than all in one environment. We estimated the duckweed percent coverage via image processing of a picture of the duckweed-water surface with ImageJ (Schindelin et al. 2012). We first used color threshold to detect green within the image. The image was then converted to black and white, where the white pixels were the original green pixels and all other pixels were converted to black pixels. The image was then binarized and we calculated the percent of white pixels within the image, using the analyze particle function within ImageJ to measure the duckweed coverage. If the calculated value was within 2% of the target percent coverage, we proceeded with experiments. Examples of binarized images and their calculated percent coverage are in Supplementary Figure S2. In total, the locomotion of three specimens for each type of surface was examined.

Recording

A Photron FASTCAM MINI AX 2000 set at a resolution of 1024 by 1024 pixels with a frame rate of 2,000 frames per second was used to record the locomotion of the *M. americana* on the different surfaces. A Nikon 70–200 mm f/2.8G ED VR II AF-S NIKKOR Zoom Lens was mounted onto the camera. The camera and lens were attached to a vertically placed Thorlabs Optical Rail and pointed at the specimens' dorsal sides. We placed the insects into 10.0 by 10.0 by 1.5-cm Thermo Scientific Petri dishes, each dish being either filled halfway with water, covered with 1000-grit sandpaper, or filled halfway with water and covered with varying amounts of duckweed (Fig. 2). These Petri dishes were raised slightly above a table, put against a white background, and placed directly under the camera's lens. An LED light was also lit about 3 inches underneath the Petri dishes for enhanced recording quality. Each insect was recorded individually and gently poked with a small stick for movement to evoke an escape response on every substrate, following existing water-strider research methods (Crumière et al. 2016; Hu and Bush 2010). A "trial" is a recording of an insect where it moved at least three body lengths. On sandpaper and on water, we tested three specimens ($N = 3$) and total trials per substrate was $n = 21$ (seven trials of each individual). For duckweed, $N = 3$ and total trials per different coverage was $n = 15$ (five trials of each individual). Individuals were assigned random substrate orders for testing.

Tracking, data acquisition, and analysis

After recording, the following points on the specimen were tracked for each recording: the coxae, tibiofemoral

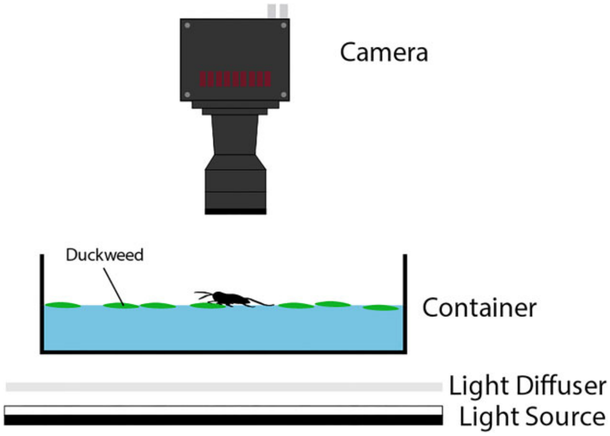

Fig. 2 Experimental Setup Schematic of experimental setup. A high-speed camera is mounted above a container of water with duckweed on top. The container rests on a diffuser. A light source is set at a short distance below the diffuser to provide more even lighting. *Microvelia americana* are recorded individually running on the water partially covered with duckweed.

joints, tibiotarsal joints, tarsi tips, the abdomen tip, and the head. DeepLabCut (DLC) pose estimation machine learning software was utilized entirely for the sandpaper and water surfaces when it came to tracking of all the aforementioned points (Nath et al. 2019) (see Supplementary Movies S1 and S2). However, on the water-duckweed surface, DLC was used only to track the tip of the abdomen, the head, the coxae, and the tibiofemoral joints. We tracked the rest of the points manually using PFV4 since DLC was unable to track these particular points with sufficient accuracy (see Supplementary Movie S3). Ultimately, we used the data gathered from the videos and the tracking (position and time of each point) to calculate the displacement, velocity, joint angles, and step amplitude for each recorded specimen. We did not calculate body speed on 10% or 25% coverage because the organism would mainly traverse the duckweed or the water part and not both consistently throughout a trial, creating a bias of speed per trial. The kinematics of the left and right leg for each pair were averaged together.

Statistical analysis

For statistical analysis, we used a linear mixed effects model (Bates et al. 2015) to find if the set of treatment effects yielded differences amongst the means of each group with post-hoc Tukey's difference criterion to find which pairs of treatment effects were statistically different (all pairwise comparisons in Supplemental Information). We used linear mixed effects model, since number of trials vary per surface and to account for any possible random effects from individual specimen. We compared different models with and without trial number as a treatment effect and body length as a random effect and found that the model with only substrate type as a treatment effect was either a better fit or statistically similar ($P > 0.05$) to other models. Therefore, we used the model with only substrate type as a treatment effect. In all models, specimen number is treated as a random effect. A custom R (R version 4.4.0) script (Bates et al. 2015; Genz and Bretz 2009; Hothorn et al. 2008; R Core Team 2024; RStudio Team 2020) was used for statistical analysis. We defined statistical significance as $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.

Results

Body and leg velocity

We found that *M. americana* is significantly faster on water, achieving a maximum speed of 56 body lengths per second (bl/s) (Fig. 3A, $P < 0.001$, $N = 3$, $n = 21$ with seven trials per individual). Our values for body speed are similar to what has been measured in prior research (Crumière et al. 2016; Hu and Bush 2010). In contrast, its maximum body speeds on sandpaper and with 50% duckweed coverage, at 26.5 bl/s and 28.7 bl/s, respectively ($P > 0.05$), are about half that on water (for each sandpaper and water, $N = 3$, $n = 21$). Across substrates, *M. americana*'s upper legs move at similar maximum speeds (Fig. 3B). Yet, on water, *M. americana*'s middle and hind legs moved faster than on sandpaper and duckweed at 51 bl/s ($P < 0.001$) and 46 bl/s ($P < 0.001$), respectively. This trend mirrors the body speed observations, which might explain the lower body speeds on sandpaper and 50% duckweed, where the middle and hind legs did not exceed speeds of 40 cm/s.

Joint angles

We measured the tibiotarsal joints and tibiofemoral joints, along with step amplitudes for all legs across the three different substrates (Fig. 4B). The ampltiude of the tibiotarsal joints ($\theta_{AJ,max}$) showed an increasing trend from water to sandpaper to duckweed for all legs ($P < 0.05$, Fig. 4C). On both duckweed and sandpaper, the amplitudes of tibiofemoral joints ($\theta_{EJ,max}$) for the upper and hind legs were higher than those on water ($P < 0.001$). The middle leg presented an exception, as its $\theta_{EJ,max}$ was lowest on sandpaper ($P < 0.001$). In terms of tibiofemoral joints, both upper leg and hind legs exhibited higher amplitudes on sandpaper and duckweed compared to water ($P < 0.001$).

Step amplitudes and stride lengths

For water, the step amplitudes and stride lengths are lowest in the upper legs and highest in the middle and

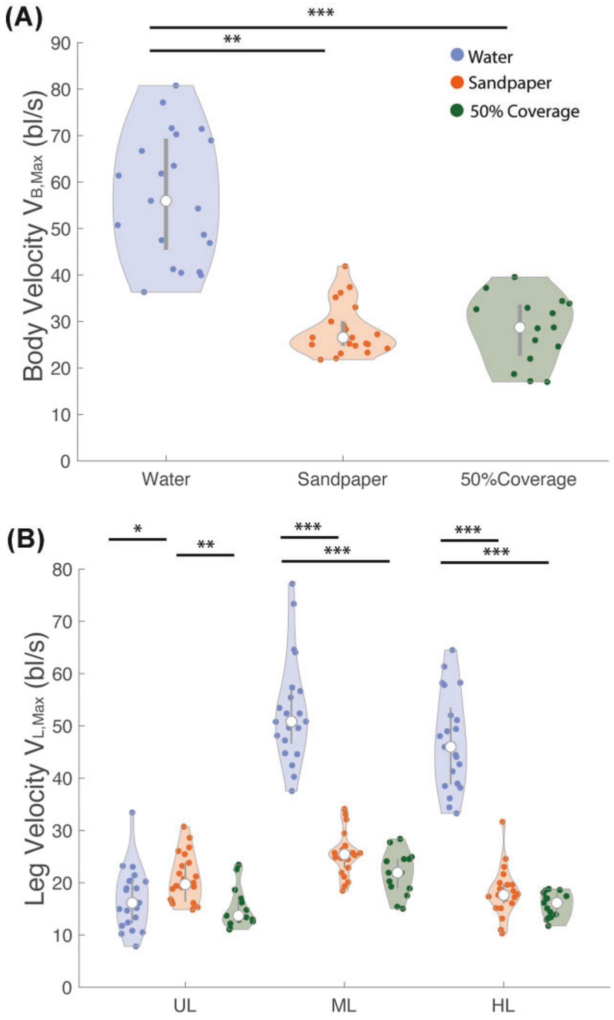

Fig. 3 Maximum velocities of body and of legs across substrates. Maximum velocities V_{max} of *M. americana* on sandpaper and duckweed are comparable, whereas movement on purely water is distinct. Each dot represents a trial (recording of insect moving). (A) Body velocity comparison on water, sandpaper, and 50% duckweed coverage. (B) Leg velocity comparison on water, sandpaper, and 50% duckweed coverage for each leg location (upper legs, middle legs, and hind legs). White circles represent the median. Bar represents second and third quartiles. We defined statistical significance as $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.

hind legs (Fig. 4 E and F, $P < 0.001$, see Supplementary Movie S4). Fig. 4(A) illustrates the increase in stride length for water compared to sandpaper. Interestingly, the step amplitudes and stride lengths for both middle leg and hind legs decrease in the presence of solid surfaces (duckweed and sandpaper, see Supplementary Movies S7 and S8), correlating with their reduced maximum velocities on these heterogenous surfaces (Fig. 3). On water, while the hind leg's tibiofemoral amplitude remains low, its stride lengths are higher, whereas on duckweed, despite shorter stride lengths than on water, the tibiofemoral amplitude increases. For the upper legs, an increase in stride length accompanies rising tibiofemoral amplitude.

The effect of duckweed coverage on stride length

We compare the average stride lengths of the upper, middle, and hind legs across water, sandpaper, and three levels of duckweed coverage (10%, 25%, and 50%, Fig. 5A–C). We found no statistical difference in stride lengths among all duckweed coverages and sandpaper for upper and hind legs ($P > 0.05$), indicating that *M. americana* exhibits similar stepping behavior on duckweed and sandpaper, regardless of surface coverage by obstacles. The stride lengths of both upper and hind legs show that *M. americana* approaches all levels of solid substrates with a uniform stepping pattern. When observing *M. americana* walking on 10% or 25% duckweed coverage, we notice that they maintain their stride length and tibiofemoral joint angles for most of their traversal whether they are on water or duckweed (see Supplementary Movie S5). The stride length of the upper legs is found to be the shortest on water (0.19 bl, $N = 3$, $n = 21$) compared to other substrates (Fig. 5A, $P < 0.001$). In contrast, the stride lengths of the middle and hind legs are higher on water than on duckweed and sandpaper (Fig. 5B and C, $P < 0.001$), showcasing an inverse trend. Specifically, the stride lengths of the middle legs decrease as the friction or heterogeneity of substrates (% duckweed) increases (Fig. 5B, $P < 0.001$). With *M. americana* moving slower on solid substrates (Fig. 3A and B), their upper legs became more active, displaying higher joint angles (Fig. 4).

Discussion

Navigating complex environments necessitates that organisms adapt or modify their gait for survival. For the tiny *Microvelia*, navigating a pond's complex and obstacle-laden environment requires adaptability over all surfaces. Our findings reveal that *M. americana* not only locomotes on water, duckweed, and sandpaper but also adapts its gait to the variation of these surfaces. Across all substrate types—unimpeded water and heterogeneous substrates of sandpaper and duckweed—*M. americana* exhibited the alternating tripod gait (Fig. 1C).

Previous research investigates modified tripod gaits on terrestrial surfaces. Blaberid cockroaches switch from the alternating tripod gait to a metachronal gait, reducing vertical amplitudes and enhancing lateral amplitudes to speed up on land (Weihmann et al. 2017). Similarly, wood ants (Reinhardt and Blickhan 2014) and fruit flies (Wosnitza et al. 2013) increase their stride frequencies to hasten land movement. North African

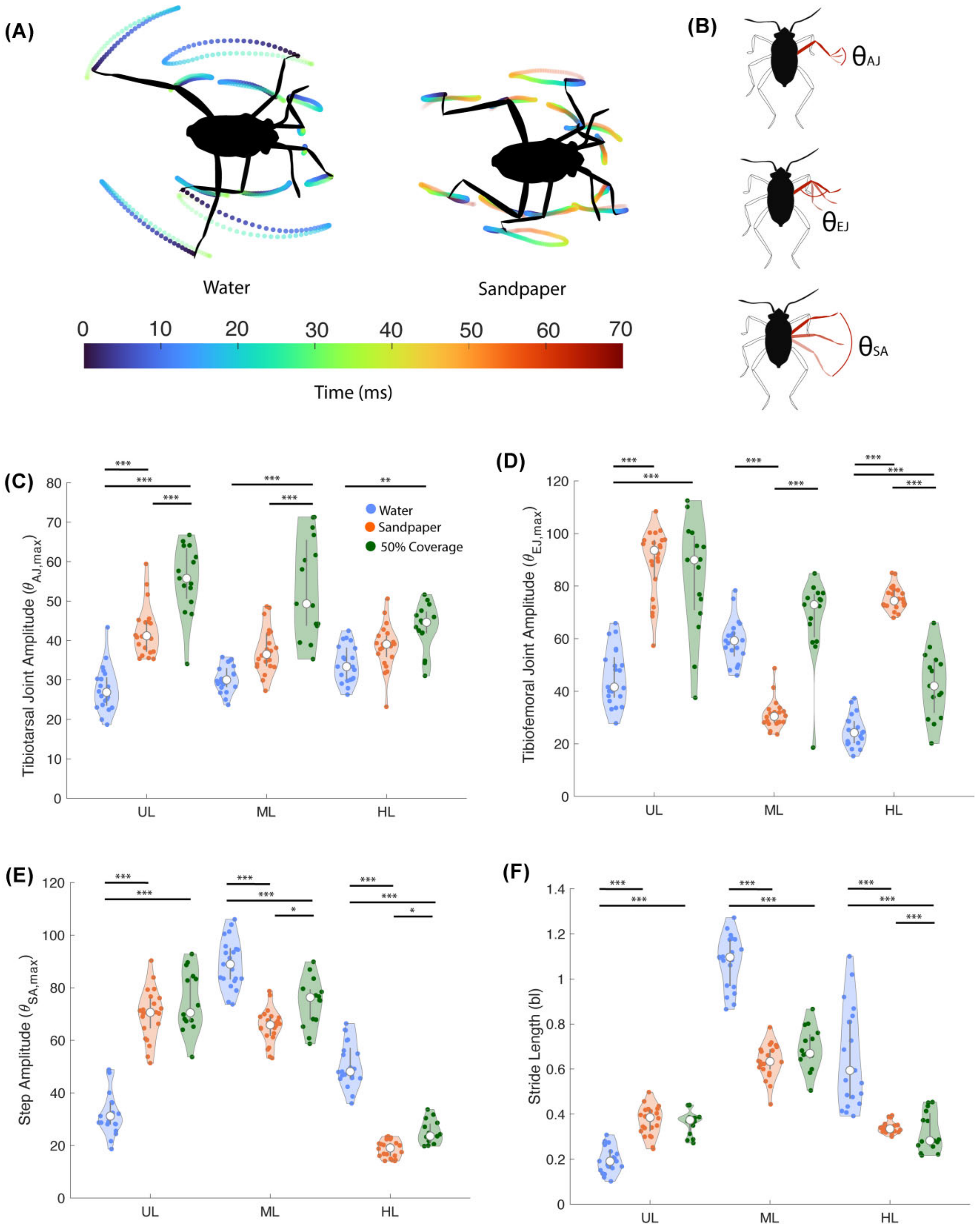

Fig. 4 Kinematics of *M. americana* locomotion on different substrates. (A) *Microvelia americana* tarsi and tibiofemoral joint trajectories on sandpaper compared to on water. (B) Schematic demonstrating how each angle is calculated. From top to bottom: tibiotarsal joint (AJ), tibiofemoral joint (EJ), and stroke amplitude (SA). (C–E) Amplitudes of each leg, upper leg (UL), middle leg (ML), and hind leg (HL), according to the joint angles illustrated in (B), across substrates. (F) Stride length comparison across substrates. White circles represent the median. Bar represents second and third quartiles. We defined statistical significance as $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.

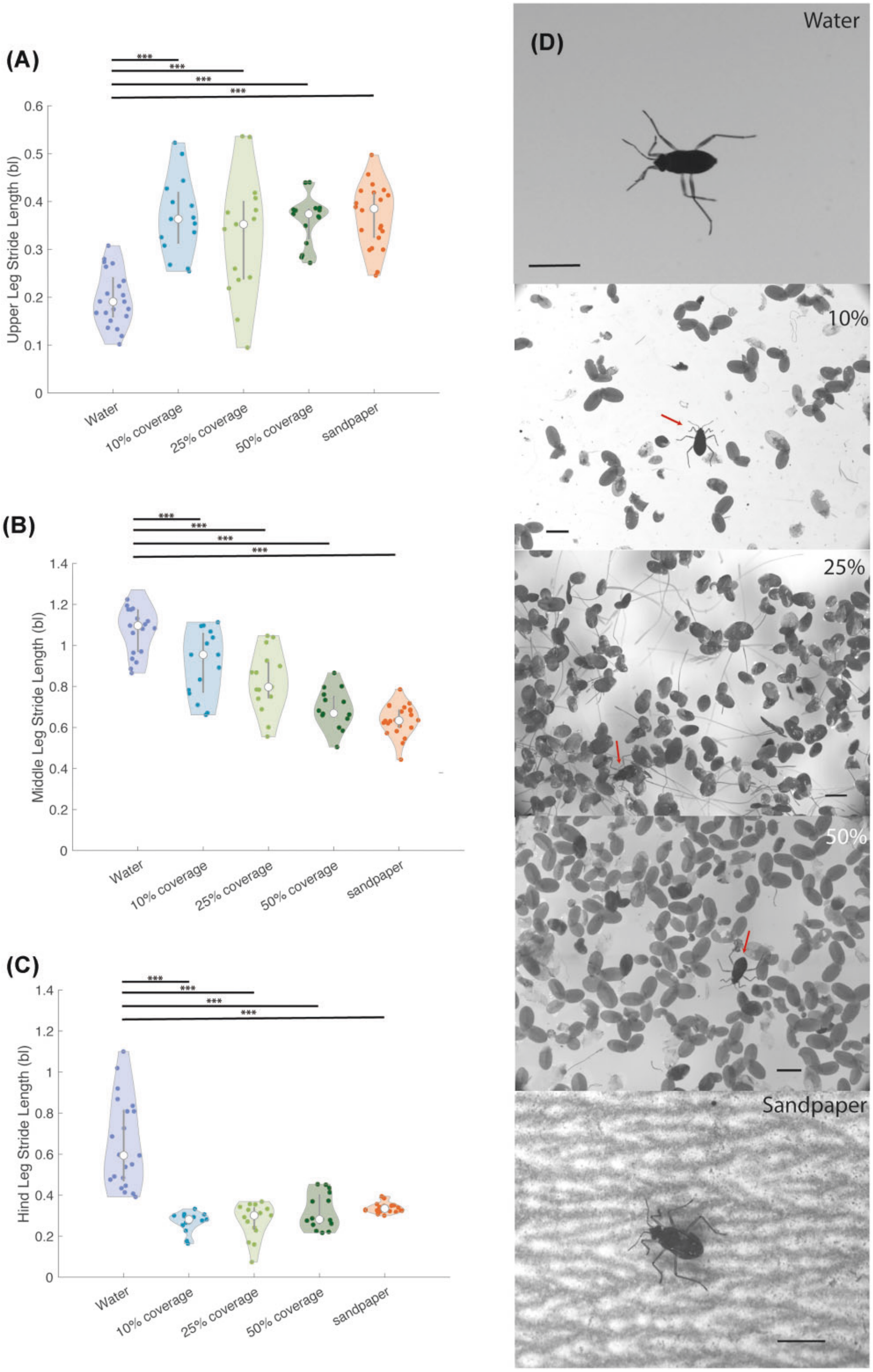

Fig. 5 Stride length comparison across substrates and varied duckweed coverages (10%, 25%, 50%). (A) Average stride length of upper legs on each substrate shows that *M. americana* increases their upper leg's stride at the presence of a solid substrate. (B) Average stride length of middle legs across substrates show that increase in duckweed coverage leads to a decrease in stride length. At 50% coverage the stride length of the middle leg is similar to the stride length on sandpaper. (C) Average stride length of hind legs reveals that *M. americana* decrease the stride of their hind lengths at the presence of a solid substrate. (D) Photos of each substrate with an individual *M. americana*. Each scale bar represents 2 mm. Arrows show where *M. americana* is located. From top to bottom, the substrates are clear water, water with 10% duckweed coverage, 25% duckweed coverage, 50% duckweed coverage, then sandpaper. Duckweed is sometimes found with submerged routes underneath the frond as seen in 25% coverage image. White circles represent the median. Bar represents second and third quartiles. We defined statistical significance as * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

desert ants shorten their stance phase to boost their body speed (Wahl et al. 2015). The changes in gait found in terrestrial hexapods are distinct from those used by water-walking insects due to different constraints. Either insects must learn to swim to shore or the water walking insect must maintain a careful balance of surface tension on the deformable surface of water in order to not drown.

There exists some exploration of tripod gaits in water—tree canopy ants, *Pachycondyla* spp. and *O. bauri*, use their contralateral front legs and middle legs to row on water surfaces in a modified alternating tripod gait (Yanoviak and Frederick 2014), using their hind legs for roll stability to prevent from flipping over. Other tree canopy ants, such as *C. americanus*, use their middle legs as rudders rather than for rowing (Yanoviak and Frederick 2014). However, these excursions into fluids are temporary—with *C. schmitzi* ants, for instance, which live symbiotically with the pitcher plant, staying in fluid for <45 s, unlike the *M. americana* which spends most of its time on water (Bohn et al. 2012; Crumière et al. 2016). Other neustonic organisms, if they want to traverse on land, either have to switch to a new gait the fisher spider switches from a rowing gait on water to an alternating tetrapod gait on land) or simply cannot traverse on land at all (water striders such as Gerridae, Rhagovelia, and Velia cannot use their rowing gait on land) (Andersen 1976; Crumière et al. 2016; Hu and Bush 2010; Santos et al. 2017; Suter 2013; Suter and Wildman 1999). Thus, *M. americana* serves as a prime supplement to what is missing thus far as we ask how this organism is able to locomote on every substrate with just one gait.

Microvelia (Fig. 1B) are known to traverse both land and water, though prior research primarily focused on smooth substrates, neglecting plant-surface substrates (e.g. duckweed Fig. 1A), and lacked within-species comparisons (Crumière et al. 2016). To address this, we tested *M. americana* on high friction sandpaper to mimic the rough terrain (rocks) surrounding their aquatic habitats and on duckweed-covered water surfaces to assess locomotion on natural, heterogeneous surfaces within their environment. Along with noticeable visual differences in the gait (Fig. 4A), we identified distinctive gait properties for *M. americana* across the three different substrates, described in the next sections.

Upper legs and hind legs contribute more on land vs. water

Microvelia americana achieve significantly higher speeds on water than on land or duckweed-covered areas (Fig. 3), as demonstrated by their increased step amplitudes and speeds on water (Fig. 4E). The middle legs display longer stride lengths and larger step amplitudes than the other legs when on water (see Supplementary Movie S4), consistent with previous studies that assign propulsion to the middle legs (Andersen 1976; Crumière et al. 2016). Acting as oars, the water strider legs push against the water (Gao and Feng 2011; Steinmann et al. 2021), with the middle legs stroking at a higher amplitude to provide the most propulsion. This action suggests that decreasing the tibiofemoral joint amplitude in the hind legs could lead to less power use, more energy conservation while pushing against the frictionless smooth surface of water (Labbé et al. 2019). On sandpaper and with 50% duckweed coverage, however, *M. americana* increase their hind legs' joint angles while reducing their stride lengths and step amplitudes (see Supplementary Movies S7 and S8). They also heighten the joint angles in their upper legs along with increasing stride lengths and step amplitudes (Fig. 4C–F). This adjustment occurs because *M. americana* bend their legs more, possibly lifting them higher to navigate the topology of frictional rough surfaces. On such surfaces, *M. americana* face difficulty sweeping and extending their legs as easily as on water, due to obstacles obstructing their tarsi, leading to shorter stride lengths and greater leg bending in the upper and hind legs. Foot trajectory comparisons on water versus more frictional land surfaces further illustrate these differences (Fig. 4A). Terrestrial insects using the alternating tripod gait, like cockroaches, also show higher hind tibiofemoral joint amplitudes on frictional surfaces (Kram et al. 1997; Ritzmann et al. 2004).

Microvelia americana treat duckweed as a land-like surface

Across all substrates, *M. americana*'s middle legs demonstrate the least variance in stride lengths, especially when comparing water, various duckweed coverages, and sandpaper (Fig. 5A–C). These legs also maintain tibiofemoral joint angle values relatively consistent (Fig. 4D). This consistency suggests that the middle legs, known for being the main propulsers on water (Andersen 1976; O'Neil et al. 2024), maintain a similar function across different terrains. Our findings indicate that *M. americana* navigate duckweed coverages similarly to how they would navigate sandpaper, treating both as "land" conditions. While adapting their gait to accommodate substrates floating on water, *M. americana* distribute more work to the other legs on land-like surfaces as the middle leg stride lengths decrease on duckweed and sandpaper.

Limitations

While aiming to reproduce the complex and varied system of a pond during our *M. americana* recordings, the range limitations of our high-speed camera and ability to reliably use DLC to track joints in this tiny insect constrained the area available for *M. americana* locomotion. Our study also encompassed a small sample size and examined a preliminary selection of substrates (limited range of duckweed coverages and one sandpaper type). Despite these constraints, our experimental setup yielded consistent results across tests. Future studies could expand the number of specimens, possibly including different juvenile instars for developmental comparisons and explore additional substrates or duckweed coverage densities. Future studies may also aim to capture the transition of leg kinematics between differing substrates, as an *M. americana* would navigate its natural environment. We observed a z-component in the amplitude of leg and joint movements, which our study did not capture. Accurately tracking leg movements in the z-direction would offer a more complete understanding of leg behavior on heterogeneous, rough surfaces.

We also noted that duckweed fronds move when *M. americana* traverse them. Future studies could quantify the movement of these fronds during *M. americana* tarsi interactions. Examining locomotion on wet versus dry surfaces could provide additional insights, given that *Microvelia* inhabit environments where they may encounter both as the land surfaces are likely to be wet being nearby water. *M. americana*'s primary movements—to pursue prey or escape predators—mean that their cross-substrate locomotion is not always continuous. For instance, on surfaces with sparsely scattered duckweed (10% and 25% duckweed), *M. americana* often move across larger water areas and halt upon reaching duckweed. This behavior likely serves as an underwater-predator evasion strategy, yet it limited our observations of smooth transitions between aquatic and terrestrial locomotion.

Conclusions

In our study, we determined how *M. americana* modifies the alternating tripod gait to traverse on different surfaces through high-speed imaging and pose-estimation deep-learning software. Through our results, we discover that *M. americana* move their upper legs at a higher stride length on land than water, suggesting that the upper legs provide more propulsion on land and may be needed to facilitate walking on rougher terrain. Furthermore, we discover that the stride lengths of the upper legs and hind legs are statistically similar across all duckweed coverages and sandpaper within this study. This suggests that once *M. americana* know that solid debris is present on water, that they will adjust their upper and hind legs to move similarly to their movement on land. *M. americana* were also found to decrease their step amplitude with increasing duckweed coverage. Since the middle legs are used as the main means of propulsion, our data suggest that the *M. americana* are adjusting the stride of their middle legs to move more quickly on more variable terrain.

This unique application of a common terrestrial gait, the alternating tripod gait, for aquatic running showcases the potential for bioinspired designs in cross-terrain and amphibious micro-robots. Inspired by *M. americana*, future robotic designs might only require a single adaptable gait for multifaceted environmental navigation, offering insights into mechanosensory affordances for multi-environmental adaptability (Dallmann et al. 2023; Li et al. 2009; Spagna et al. 2007; Suter and Wildman 1999). For example, a bio-inspired robot based on *M. americana*, can utilize a six-legged alternating tripod gait that senses changes in the environment based on topological height difference, slip, and friction, and adjust its gait behavior to accommodate different environments. The adjustments in joint and step amplitudes will allow the robot to mimic *M. americana* and maneuver between land and water. These findings highlight opportunities for further research in gaits adjustments across substrates, the biological actuation behind traversal in diverse environments, and the implications for semi-aquatic robotics and bio-inspired design in navigating complex media, such as sand, or transitional environments (Li et al. 2009; Snell-Rood 2016). Ultimately, these results can influence the design of future amphibious microbots that can better traverse rough and uncertain terrain that may include random debris.

Author contributions

J.O. and H.W. designed the experiments. J.O., K.Y., G.D., and H.W. carried out the experiments and acquired the data. J.O. analyzed the data and interpreted the results. M.S.B. reviewed the design and execution of experiments, the data analysis, the interpretations, and the manuscript. J.O., K.Y., G.D., and M.S.B. contributed to writing the manuscript.

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Supplementary data

Supplementary data available at *ICB* online.

Conflict of interest

No conflicts.

Data availability

The data underlying this article are available in the article and in its online supplementary material.

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