COMPLIMENTARY/POSTER SESSION PAPER

Muscle Fatigue in the Latch-Mediated Spring Actuated Mandibles of Trap-Jaw Ants

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Synopsis   Muscle fatigue can reduce performance potentially affecting an organism's fitness. However, some aspects of fatigue could be overcome by employing a latch-mediated spring actuated (LaMSA) system where muscle activity is decoupled from movement. We estimated the effects of muscle fatigue on different aspects of mandible performance in six species of ants, two whose mandibles are directly actuated by muscles and four that have LaMSA "trap-jaw" mandibles. We found evidence that the LaMSA system of trap-jaw ants may prevent some aspects of performance from declining with repeated use, including duration, acceleration, and peak velocity. However, inter-strike interval increased with repeated strikes suggesting that muscle fatigue still comes into play during the spring loading phase. In contrast, one species with directly actuated mandibles showed a decline in bite force over time. These results have implications for design principles aimed at minimizing the effects of fatigue on performance in spring and motor actuated systems.

Introduction

Muscles that experience prolonged or repeated contractions eventually experience physiological fatigue, with reduced capacity to exert force and do work. Muscle fatigue can decrease whole-animal performance, which can have important effects on their ecology. Many studies have examined the cascading neuromuscular and metabolic mechanisms that cause muscle fatigue (Cairns et al. 2005; Allen et al. 2008; Enoka and Duchateau 2008) and its consequences on muscle power output and performance. Most studies focus on movements directly actuated by muscle contraction, where prolonged or repeated use of a muscle correlates with a decrease in movement performance (James et al. 1995; Maluf et al. 2005; Williams et al. 2013). In some organisms, however, muscles instead load a biological spring (tendon or ligament) held in place by a latch to be released at a later time. These latch-mediated spring actuated (LaMSA) systems (Longo et al. 2019) are prevalent in biology and represent an interesting comparison with directly actuated systems. In the present study, we sought to understand the effect of muscle fatigue on the performance of a LaMSA system.

In animals that employ LaMSA mechanisms, muscle activity is temporally decoupled from movement and can be separated into phases (Gronenberg 1996a; Ilton et al. 2018; Longo et al. 2019). Starting with a latching phase, the accelerating mass (projectile, appendage, or whole body) is held in place by a latch. This enables the loading phase, where contraction of a muscle stores po-

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Table 1 Collection and voucher deposition information for ants used in this study

Species	Mandible mode	Colonies	Collection location	Voucher location	Voucher number
A. paripungens	Spring Actuated	1	Darwin, Australia	NMNH	USNMENT01124355
C. chromaiodes	Directly Actuated	2	Urbana, Illinois, USA	UIUC	USNMENT01128951
D. armigerum	Spring Actuated	2	Nouragures, French Guiana Cayenne, French Guiana	UIUC	USNMENT01128952
O. brunneus	Spring Actuated	3	Venus, Florida, USA	NMNH	USNMENT01128953
O. relictus	Spring Actuated	2	Venus, Florida, USA	NMNH	USNMENT01128954
P. californicus	Directly Actuated	1	Hurricane, Utah, USA (AntsAlive)	UIUC	USNMENT01124405

tential energy in a spring. During spring actuation, the latch releases the accelerating mass (often controlled by a fast trigger mechanism) and transfers potential energy from the spring into kinetic energy of the accelerating body (Divi et al. 2020). The accelerating body transitions to ballistic motion after the spring stops exerting a force on it. Although muscle activity is limited to the beginning of this process, muscle fatigue could theoretically impact the performance of a LaMSA movement by reducing the amount of energy that is stored in the spring during the loading phase.

LaMSA mechanisms can enable greater velocity, acceleration, and power density than would be possible from direct muscle actuation. For example, salamander tongues that use LaMSA mechanisms reach velocities five times greater and power densities over 100 times greater than tongues directly actuated by muscle (Deban et al. 2020). Similarly, Sutton et al. (2019) showed that below a certain body size, insects can jump significantly higher and with greater energy density using LaMSA mechanisms. In ants, the focus of the current study, the mandibles of species that have evolved LaMSA have velocities, accelerations, and power densities orders of magnitude greater than closely related species whose mandibles are directly actuated by muscle (Booher et al. 2021). Despite the great deal of attention given to the morphology, energetics, and evolution of LaMSA movements, relatively little is known about if or how muscle fatigue plays a role in these systems.

Although most ants possess directly actuated mandibles, at least four independent clades, including more than 600 species, have convergently evolved "trap-jaws"—fast-closing mandibles powered by LaMSA mechanisms (Larabee and Suarez 2014). Each lineage uses different morphological structures to act as a latch, spring, and trigger, but they all power rapid mandible strikes that are used for predation of fast or dangerous prey (Moffett 1986; Gronenberg 1996b; De la Mora et al. 2008). In at least one genus, *Odontomachus*, mandibles are also used to power antipredator escape jumps (Patek et al. 2006; Spagna et al.

2009; Larabee and Suarez 2015). As all trap-jaw ants use a large adductor muscle to load the spring, and sustain these contractions for many seconds (Gronenberg 1995a; Larabee et al. 2017), trap-jaw ants are a good case study for examining the influence of muscle fatigue on LaMSA systems.

Mandibles are critical multi-purpose tools for ants, and in arthropods generally. Therefore, factors that reduce ant performance, such as muscle fatigue, are expected to be important for shaping their evolution. We tested if performance of the LaMSA mandibles of trap-jaw ants showed signs of being influenced by muscle fatigue. Because muscles must maintain their contraction during the loading phase of a strike, we tested the hypothesis that muscle performance occurring between strikes (reflecting the time it takes to load a spring or muscle recovery) would decrease with repeated use. To accomplish this, we recorded multiple consecutive mandible strikes with high-speed videography and derived their kinematic performance for four species of trap-jaw ants. For comparison, we examined the role of muscle fatigue in two species of ants whose mandibles are actuated directly by muscles by measuring any decrease in performance during bite force experiments. Our hypothesis for directly actuated mandibles was that repeated or prolonged bites would result in decreased bite performance.

Materials and methods

Study organisms

Colonies of four species of trap-jaw ants were collected live for study: *Anochetus paripungens*, *Daceton armigerum*, *Odontomachus brunneus*, and *O. relictus* (Table 1). To study directly actuated mandibles, colonies of the carpenter ant, *Camponotus chromaiodes*, were collected live and individuals of the harvester ant, *Pogonomyrmex californicus*, were purchased from a commercial vendor (AntsAlive, Hurricane, UT, USA). Laboratory nests were constructed out of plaster-filled petri dishes placed inside plastic boxes. Ants were pro-

vided with water and sugar water *ab libitum* and fed crickets, flies, or termites one to three times a week. Nests were kept at 25°C, 50% relative humidity, and a 12-h light-dark cycle.

Trap-jaw ant performance

Trap-jaw ant mandible performance fatigue was measured using high-speed videography of repeated mandible strikes using methods similar to previous studies (Larabee et al. 2017; Gibson et al. 2018). Briefly, ants were restrained by the posterior dorsum of the head to an insect pin with utility wax (Kerr Laboratory Products, Orange, CA, USA). This pin was attached to a thin rod fitted onto a micromanipulator and ants were positioned with their mandibles perpendicular to the optical axis of a Zeiss SteREO Discovery V20 Microscope (Carl Zeiss Inc., Oberkochen, Germany). Strikes were backlit with an LED light and recorded with a Phantom V9.1 high-speed camera (Vision Research Co. Wayne, NJ, USA) at framerates of 50,000–100,000 frames per second. To account for adaptation of the mechanoreceptors that trigger the strike reflex, strikes were elicited under two different experimental protocols: (1) for *Anochetus* and *Odontomachus*, five strikes within 15 s were provoked by gentle blowing from the experimenter, (2) for *Daceton*, as many strikes as possible were elicited over the course of 2 min with air from a nasal bulb at a rate of one puff per second. Following recordings, mandible length was measured with a Leica DFC 425 digital microscope camera connected to a Leica M205 C stereo microscope (467 nm resolution) (Lecia Microsystems, Wetzlar, Germany). Strikes were recorded from three workers of *A. paripungens*, 40 workers of *D. armigerum*, six workers of *O. brunneus*, and five workers of *O. relictus*. When the same individual was recorded twice, at least 5 min separated the recorded strike sequence.

Strike kinematics were estimated from digitized mandible strikes using the same method as Larabee et al. (2018). Using the x–y coordinates of the mandible tip and the mandible length, the angular displacement of mandible during the strike was calculated trigonometrically. Raw displacement data were smoothed in R (ver. 4.1.2) using the *pspline* package (ver. 1.0–18), and this smoothed data was used to estimate strike kinematic traits (Fig. 1). Five traits were estimated for each strike: (1) *Strike duration*—the time from mandible rest to when they stop making forward progress. (2) *Peak angular velocity* and (3) *peak angular acceleration* were calculated from the first and second derivatives of the displacement data, respectively. (4) Mandible *acceleration duration* is an estimate of the how long the spring applies a force to the mandible and was calculated from
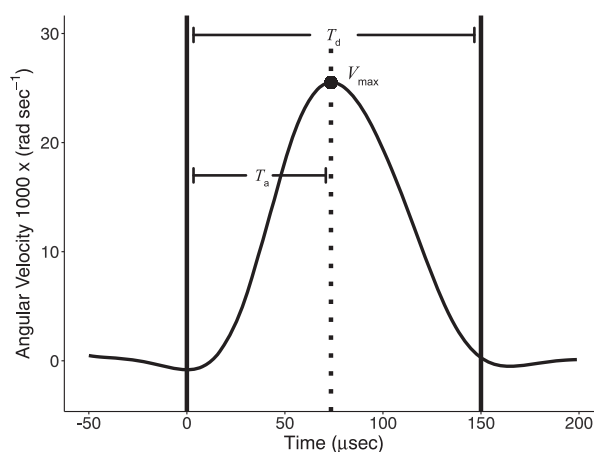

Fig. 1 Stereotypical trap-jaw ant performance. Angular velocity as a function of time. Velocity was derived from the x–y coordinates of mandible tips from high-speed videography. Performance traits: strike duration (T_d), peak angular velocity (V_{max}), peak angular acceleration (not shown—calculated from second derivative of change in displacement), acceleration duration (T_a), and strike interval (not shown) were derived from these data.

the time of rest to peak velocity. (5) *Strike interval* was defined as the time elapsed between start times between strikes. Traits for the left and right mandible were averaged together for each strike, and estimated only for the first five recorded strikes.

Performance fatigue was assessed for the four species of trap-jaw ant by measuring how performance traits changed over the course of five repeated strikes. To account for differences in body size (correlated with peak performance of LaMSA systems [Spagna et al. 2008]), % change in performance since the first strike was compared for these analyses. Data were analyzed separately for each species with the exception of *O. brunneus* and *O. relictus*, where these species were pooled to simplify interpretation of the results. This is justified because these taxa are closely related sister species (Larabee et al. 2016; Matos-Maraví et al. 2018; Fernandes et al. 2021), use the same trap-jaw mechanism, and are very similar in overall body size. Also, there was no difference in strike performance between the two species (two-sided t-test, mean strike duration: $t = -0.368$, $P = 0.72$; mean peak velocity: $t = -1.73$, $P = 0.112$; mean acceleration duration: $t = -0.589$, $P = 0.566$). Differences in strike performance were tested with a one-way repeated measures ANOVA using the *Rstatix* package (ver 0.7.0) in R. Samples that violated the assumption of sphericity had a Greenhouse-Geisser correction applied. Post-hoc pairwise t-tests with a Bonferroni correction for multiple comparisons were used to test for differences between strike 1 and strikes 2–5. In the case of strike interval, comparisons were made between interval 1 and intervals 2–4. Raw data and scripts of all analyses, in-

Table 2 Performance summary of trap-jaw (LaMSA) ant mandibles

Species	N	Min. duration (μs)	Peak angular velocity (rad/s)	Peak angular acceleration (rad/s²)	Min. acceleration duration (μs)	Strike interval (s)
A. paripungens[a]	4	80.2 ± 11.6	$43.4 \times 10^3 \pm 2.3 \times 10^3$	$1.33 \times 10^9 \pm 0.151 \times 10^9$	51.4 ± 10.6	1.10 ± 0.22
D. armigerum	20	78.9 ± 23.3	$24.4 \times 10^3 \pm 8.5 \times 10^3$	$1.09 \times 10^9 \pm 0.994 \times 10^9$	64.3 ± 13.2	5.98 ± 3.34
O. brunneus	11	108 ± 19.5	$43.5 \times 10^3 \pm 3.3 \times 10^3$	$1.24 \times 10^9 \pm 0.152 \times 10^9$	64.6 ± 12.1	1.78 ± 0.411
O. relictus	7	112 ± 21.8	$46.7 \times 10^3 \pm 4.1 \times 10^3$	$1.34 \times 10^9 \pm 0.163 \times 10^9$	68.0 ± 12.3	1.76 ± 0.988

[a]Data originally published in Gibson et. al (2018). All values are means ± standard deviation. Species means for strike duration, and acceleration duration are calculated from individuals minimums. Species means for peak velocity and acceleration. Species means for strike interval are based on individual means.

cluding filming conditions and kinematics calculations are available in supplementary material.

Directly actuated mandible performance

Repeated biting performance of non-LaMSA ant mandibles was studied with bite force experiments. A total of 20 workers of *Camponotus* and 21 workers of *Pogonomyrmex* were used in bite force experiments. A Flexiforce ELF-system piezoelectric force sensor (Tekscan, South Boston, MA, USA) was trimmed, covered in parafilm, and calibrated according to the manufacturer's instructions. The sensor was sampled at 150 Hz and a gain setting ("sensitivity setting") of 18 . Ant workers were restrained by the body using soft forceps and allowed to voluntarily bite the sensor. Recording was triggered when the force sensor surpassed a 0.1 N threshold and lasted for 120 or 180 s. The resulting bite force traces were cleaned with a 10 Hz low-pass Butterfield filter using the R package signal (ver. 0.7 -7).

Two methods were used to detect performance fatigue from bite force experiments. First, bite traces were divided into discrete bites by identifying local maxima using a threshold criterium. Bites were defined to start and stop whenever bite force increased above or decreased below, respectively, 10% of the experiment maximal bite force. Three performance measures were estimated from these filtered bite data: (1) *Bite duration* was calculated as the difference between bite start and stop times. (2) *Maximum bite force* was the local maximal force of each bite. (3) *Bite interval* was calculated from the time between the start of each bite. Differences in % change in bite performance for the first five bites were tested with a one-way repeated measures ANOVA similar to the analysis of trap jaw performance. The second method involved identifying individual bites from ants that were at least 30 s long. Trends in force during long bites were then tested with linear regression and a *t*-test was used to test if the mean slope was significantly different from zero.

Results

Trap-jaw ant performance

At least five strikes were elicited within the experimental trial period for 4 sequences of *Anochetus*, 20 sequences of *Daceton*, and 18 sequences of *Odontomachus*. After five successive strikes, it often became difficult to coerce ants to strike, but some individuals continued to display a willingness to use their jaws. One individual of *Daceton*, for example, released 13 strikes within 20 s. Summaries of trap-jaw strike performance are provided in Table 2.

Most trap jaw performance traits did not vary consistently across strikes (Fig. 2, Supplementary Table S1). Strike duration, peak angular velocity, peak angular acceleration, and acceleration time all displayed variation within species and individuals (coefficient of variation ranged from 9 to 85%), but the % change in performance did not vary consistently across repeated strikes (all one-way repeated measures ANOVA tests had $P > 0.23$, see Supplementary Table 1 for full results). The one performance trait that did show a change in all species was strike interval (one-way repeated measures ANOVA: *Anochetus* $F_{(3, 9)} = 19.2, P < 0.001$; *Daceton* $F_{(3, 57)} = 3.72, P = 0.016$; *Odontomachus* $= F_{(3, 51)} = 10.4, P < 0.001$). In *Odontomachus*, strike interval changed most quickly, increasing by $35.7 \pm 48.8\%$ during interval 2 (post-hoc paired *t*-test: $t = -3.10$, adjusted $P = 0.006$). By interval 4 (between strikes 4 and 5), the strike interval increased for all species, ranging from mean % change of $69.1 \pm 73.5\%$ in *Odontomachus* to $86.2 \pm 32.9\%$ in *Anochetus* (post-hoc paired *t*-test for all species had adjusted $P < 0.02$).

Directly actuated mandible performance

General bite behavior differed between the two non-LaMSA species (Fig. 3, Table 3), with *Camponotus* having more short bites (bite number: 7.1 ± 4.7 bites, bite duration: 6.3 ± 4.2 s) and *Pogonomyrmex* fewer, longer bites (bite number: 3.6 ± 2.4 bites; bite du-
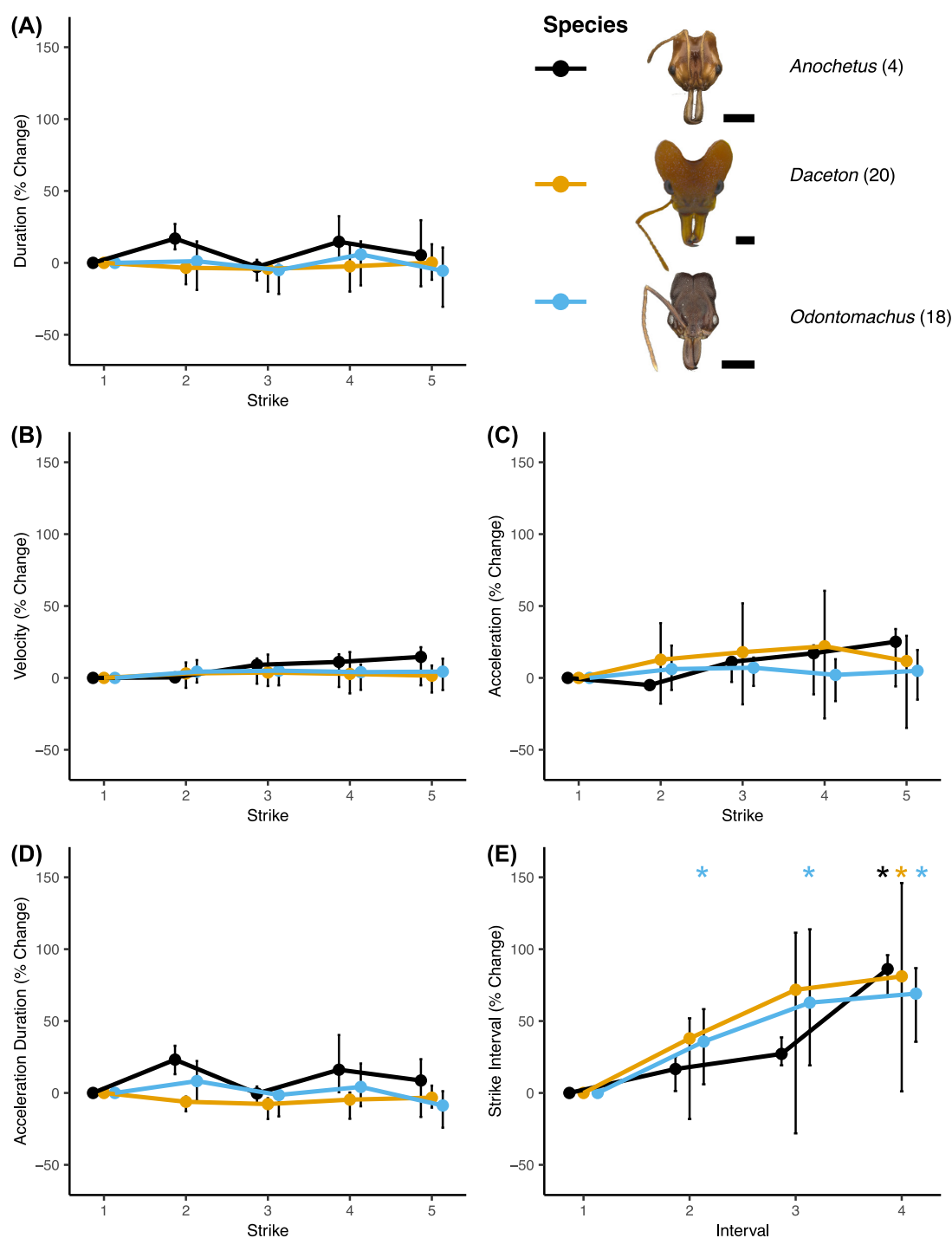
Fig. 2 Most performance traits of trap-jaw strikes do not change over repeated strikes. Percent change in performance since strike 1 is shown for each performance trait **(A)** strike duration, **(B)** peak angular velocity, **(C)** peak angular acceleration, **(D)** acceleration duration, and **(E)** strike interval. All values are means ± interquartile range. Strikes that have a significant change in performance since the first strike are labeled with asterisks. Species denoted by line color: *Anochetus* (black), *Daceton* (yellow), and *Odontomachus* (blue). Images for *A. paripungens* (CASENT0217510) and *O. brunneus* (CASENT0270601) were downloaded and edited from AntWeb. Version 8.73. California Academy of Science, online at http://antweb.org. Accessed March 14, 2022. Image of *D. armigerum* (JCG2017065) is from J.C. Gibson. Scale bars = 1 mm.

ration: 30 ± 27 s). Sixteen *Camponotus* workers and seven workers of *Pogonomyrmex* had at least five bites for a repeated measures test. Bite force decreased significantly over the course of five bites only in *Cam-ponotus* (one-way repeated measures ANOVA: *Camponotus* $F_{(2.75, 41.3)} = 3.56$, $P < 0.025$; *Pogonomyrmex* $F_{(1.8, 10.8)} = 2.5$, $P = 0.131$). By bites four and five, peak bite force decreased by 29.5 and 25.9%, re-
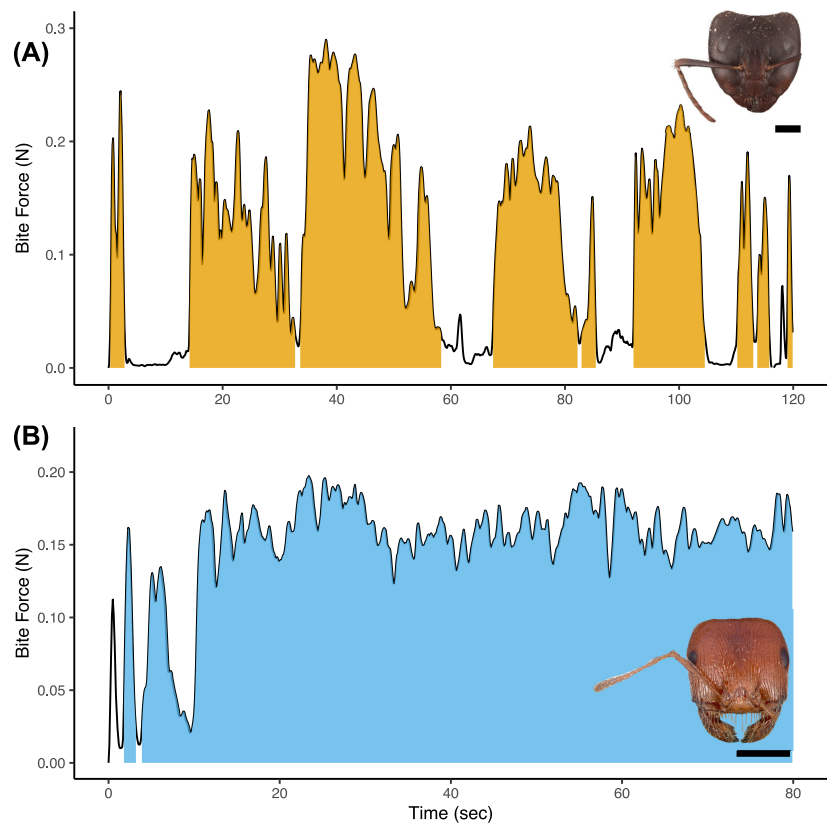
Fig. 3 Stereotypical bite force plot from a directly actuated mandible. Bite force (N) plotted over time (s) after lowpass Butterfield filter was applied to data. Bites were defined (shaded regions) after force increased over 10% of maximal bite force and lasted longer than 0.1 s. Data shown for **(A)** *Camponotus* and **(B)** *Pogonomyrmex*. Images for *C. chromaoides* (FMNHINS0000045774) and *P. californicus* (CASENT0005710) were downloaded and edited from AntWeb. Version 8.73. California Academy of Science, online at http://antweb.org. Accessed March 14, 2022. Scale bars = 1 mm.

Table 3 Performance summary of directly actuated (non-LaMSA) ant mandibles

Species	N	Number of bites	Peak bite force (N)	Bite with peak force	Mean bite duration (s)	Mean bite interval (s)
C. chromaiodes	20	7.13 ± 4.73	0.335 ± 0.298	2.67 ± 2.63	6.32 ± 4.20	9.34 ± 3.52
P. californicus	18	3.61 ± 2.45	0.233 ± 0.061	2.8 ± 2.59	30.0 ± 27.2	13.6 ± 12.2

All values are means ± standard deviation. Species means for bite duration and interval are calculated from individual means.

spectively (Fig. 4). Bite duration and interval did not change over repeated bites for either species (Table 4).

A total of 4 individuals of *Camponotus* and 13 individuals of *Pogonomyrmex* had individual bites longer than 30 s that were used to analyze the change in bite performance in a single bite (Supplementary Figure 1). The three bites that started with the highest force, all declined by over 50% over the course of their duration. However on average, the slope of force during these long bites was not significantly different from zero (one-sample *t*-test: $t = -1.3711$, df = 16, $P = 0.189$). A total of 11 bites out of 17 bites had a negative slope, with force decreasing over time, but this was not significantly dif-

ferent from a random distribution (Chi-square analysis of independence: $X^2 = 2.25$, df = 1, $P = 0.134$).

Discussion

We sought to determine how muscle fatigue affects LaMSA systems by measuring the change in performance of repeated mandible strikes in trap-jaw ants. We found evidence for fatigue in only one performance trait, strike interval, which increased over repeated mandible strikes by 69–86%. Because strike interval is when the adductor muscle is loading the spring and recovering from any fatigue, these results are consistent with our hypothesis that muscle fatigue would most af-

Table 4 One-way repeated measures ANOVA of % change in bite performance

Species	N	Trait	df	F	P	Bite 2	Bite 3	Bite 4	Bite 5
Camponotus	18	Duration*	2.11, 31.6	2.14	0.132		-	-	-
		Peak bite force*	2.75, 41.3	3.56	0.025	0.904	0.976	**0.003**	**0.006**
		Bite interval	2, 30	1.48	0.244	-	-	-	-
Pogonomyrmex	20	Duration*	1.78, 10.7	1.30	0.307	-	-	-	-
		Peak bite force*	1.8, 10.8	2.50	0.131	-	-	-	-
		Bite interval*	1.06, 6.36	0.808	0.409	-	-	-	-

Samples that violated the assumption of sphericity had a Greenhouse-Geisser correction applied to reduce the degrees of freedom (asterisks). For peak bite force, the only trait with significant differences between bite, post-hoc pairwise *t*-tests between bite 1 and the four following bites were performed. *P*-values had a Bonferroni correction applied for multiple comparisons, and tests with a *P*-value < 0.05 are in **bold**.
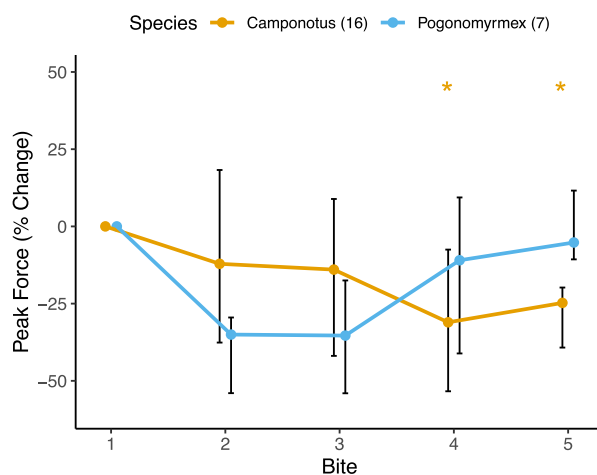

Fig. 4 Directly actuated mandibles display some decrease in performance over repeated use. Percent change in bite force over repeated bites. All values are means $\pm$ interquartile range. Bites that have a significant change in performance since the first bite are labeled with asterisks. Species are denoted by line color: *Camponotus* (yellow), *Pogonomyrmex* (blue).

fect performance traits related to the loading phase of the LaMSA mechanism. During a loading phase sometimes lasting seconds, the adductor muscle is likely experiencing tetany and an increased interval reflects the muscle's need to replenish ATP or Ca^{2+} or a longer time required to load the spring.

Performance traits associated with other phases of the LaMSA mechanism displayed no signs of fatigue in our experiments. Acceleration time, reflecting the spring actuation phase (Longo et al. 2019), did not change significantly across all strikes and species. This suggests that ants compressed their springs to a consistent strain over repeated strikes. Similarly, peak velocity and acceleration did not decrease over repeated strikes. Strike velocity and acceleration occur during the ballistic phase of the strike and are the result of the integrated dynamics of all the components of the LaMSA system (latch, motor, spring, and unlatching motor) and how

they interact during the preceding phases of the LaMSA mechanism (Ilton et al. 2018).

The repeatability of trap-jaw mandible strike performance is consistent with the idea that once a LaMSA system has loaded their spring to some threshold compression, performance is fixed with little room for modulation. Once a given spring, whose stiffness and mechanical performance is fixed, is loaded, performance would be determined by trigger dynamics (Divi et al. 2020). In other LaMSA-powered systems, such as mantis shrimp, performance can be modulated, with strike speed and energy depending on behavioral context or medium (water vs. air) (Kagaya and Patek 2016; Green et al. 2019; Feller et al. 2020). Behavioral motivation in a given context likely plays an important role in determining how much the spring is compressed, but little is known about the sensory or control mechanisms that influence spring compression.

Increased strike intervals were similar across all three genera of trap-jaw ants under study, despite them using different LaMSA mechanisms. Ants in the genus *Daceton* use their labrum as a latch to block their mandibles from closing like a doorstop, and a fast trigger muscle pulls the labrum out of the way to release a strike (Gronenberg 1996b). In contrast, *Anochetus* and *Odontomachus* use a modification of the mandible joint to latch their jaws open (Gronenberg et al. 1993; Gronenberg 1995a, 1995b; Gronenberg and Ehmer 1996). A fast trigger muscle derived from the mandible adductor muscle unlatches the jaws and allows them to snap shut. The trend of increased strike interval was consistent even across slight differences in experimental protocol, with *Daceton* allowed to strike repeatedly over a much longer period (2 min) than *Anochetus and Odontomachus* (15 s). However, the longer trial time for *Daceton* may explain the increased variability in their strike interval (Fig. 2) whether due to more variation in behavioral motivation or more time for the adductor muscle to recover from fatigue. In either case, these results are consistent with our hypothesis that fatigue in a

LaMSA system is related to muscle activity during the loading phase.

An alternative interpretation of the increased strike interval of LaMSA mandibles is that mechanosensory neurons adapted to the stimulus that elicited mandible strikes. We attempted to mitigate this possibility by using two different stimuli (continual puffs of air from the experimenter compared with timed puffs of air from a nasal bulb), which resulted in similar strike interval increases. Our experimental protocol cannot identify the exact mechanism responsible, but both sensory adaptation and muscle fatigue are steps in a cascade of mechanisms that regulate physiological fatigue (Cairns et al. 2005; Enoka and Duchateau 2008; Tuthill and Wilson 2016). Future studies will require intracellular or electrophysiological recordings of the adductor muscle and relevant mechanoreceptors involved to disentangle these two potential factors.

While we found some evidence of performance fatigue in non-LaMSA ant mandibles, our protocol likely limited the conclusions we can draw from those experiments. We found a decrease in peak bite force in *Camponotus* over repeated bites, but measuring bite force under voluntary conditions can be influenced by many factors that were not controlled for in our experiments. Animal motivation, for example, can increase the interindividual measurement variation for traits like bite force and locomotion in vertebrates (Losos et al. 2002; Anderson et al. 2008). Controlling for this behavioral by measuring muscle activity directly (*in vivo* or in isolated muscle) (Syme and Tonks 2004; Wilson and James 2004), or even monitoring the metabolic signs of fatigue (such as intracellular concentrations of lactate or ATP) (Williams et al. 2013; Kirkton and Yazdani 2021) would provide clearer understanding of the role fatigue plays in ant muscle. Combined with data on muscle geometry and fiber composition (Paul and Gronenberg 1999; Paul 2001; Püffel et al. 2021), this kind of fatigue data would provide a robust model of the determinates of bite force and performance in both trap-jaw ants and biting ants.

Contrasting results from our fatigue experiments for LaMSA versus non-LaMSA mandibles reinforce the conclusion that LaMSA mechanisms overcome some of the performance consequences of muscle fatigue. Assuming mandible adductor muscle fatigue is responsible for the reduced bite force performance of directly actuated mandibles and increased strike interval of LaMSA mandibles (see above for caveats), only directly actuated muscles experience a decrease in mandible performance from repeated use. Trap-jaw mandible performance that is relevant to their ecology (duration, velocity, acceleration, and likely mandible impact force) was unaffected. We suggest this is because unlinking muscle activity from appendage motion also unlinks the reduced performance of a fatigued muscle from mandible performance.

Muscle fatigue could, therefore, have very different effects on the ecology of trap-jaw mandibles compared to directly actuated mandibles. Most trap-jaw ants are solitary foragers, individually capturing prey items and bringing them back to the nest (Fowler 1980; Ehmer and Holldobler 1995). Consequently, a factor like fatigue, that reduces the performance of their mandibles (strike speed or force) could theoretically reduce a colony's ability to acquire nutrition. In contrast, nontrap-jaw ants often recruit nestmates for foraging and cooperatively capture and transport prey or other resources. The cooperative chains formed by *Leptogenys* foragers to capture large millipede prey offer a dramatic example of this behavior (Peeters and Greef 2015). Cooperative behavior minimizes the impact of performance reduction on any single individual and allows for functional redundancy. For example, older workers of leaf-cutting ants whose mandibles are worn down and less efficient at cutting leaves switch to other tasks like leaf-carrying (Schofield et al. 2010). Future work could attempt to quantify the burden muscle fatigue might have on foraging efficiency in trap-jaw ants.

By decoupling muscle activity and motion, LaMSA systems overcome many of the constraints inherent to muscle performance. Most commonly noted, tradeoffs between muscle velocity and force can generally limit how quickly a motion can occur, especially in small animals (Sutton et al. 2019). LaMSA systems are also more robust to the effects of temperature than direct muscle action, as has been demonstrated in vertebrates with ballistic tongue projection, such as frogs, chameleons, and salamanders (Anderson and Deban 2010; Deban and Lappin 2011; Anderson et al. 2014). Our data suggest the performance of LaMSA mechanisms are also more resistant to muscle fatigue. This feature of LaMSA movements could serve as another source for bio-inspired design that seek to optimize limiting fatigue in motor driven movements.

Supplementary data

Supplementary Data available at *ICB* online.

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