

Kinematics of horizontal and vertical caterpillar crawling

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SUMMARY

Unlike horizontal crawling, vertical crawling involves two counteracting forces: torque rotating the body around its center of mass and gravity resisting forward movement. The influence of these forces on kinematics has been examined in the soft-bodied larval stage of *Manduca sexta*. We found that crawling and climbing are accomplished using the same movements, with both segment timing and proleg lift indistinguishable in horizontal and vertical locomotion. Minor differences were detected in stride length and in the delay between crawls, which led to a lower crawling speed in the vertical orientation. Although these differences were statistically significant, they were much smaller than the variation in kinematic parameters between animals. The ability of *Manduca* to crawl and climb using the same movements is best explained by *Manduca's* relatively small size, slow speed and strong, controlled, passive grip made possible by its proleg/crochets.

Key words: kinematics, *Manduca sexta*, soft-bodied, caterpillar, vertical locomotion, climbing, crawling.

INTRODUCTION

Animals as diverse as geckos, lemurs, frogs, woodpeckers, leopards and insects use climbing to find food or shelter or to avoid predation. Some are mainly ground dwellers whereas others are arboreal and spend their life in trees or plants. In both cases climbing is an important mode of locomotion. Climbing can range from locomotion on a slope to vertical ascending or descending. To distinguish climbing from horizontal locomotion, Hunt and colleagues suggested defining climbing as a descent or ascent at an angle of ≥ 45 deg. (Hunt et al., 1996). Many strategies for climbing are used; most involve attachment to substrates by an interlocking grip using claws, by strong bonds between the animal and its substrate using adhesives or suction or by using a friction grip, which combines the previously mentioned strategies (Cartmill, 1985). Vertical climbing brings an extra challenge in the form of pitch back or torque, pulling the upper body away from the substrate and pushing the lower part against the substrate. The magnitudes of these forces depend on the distance of the center of mass (COM) from the substrate and the mass itself (see Fig. 1A). To minimize torque a climbing animal should keep its COM close to the substrate.

Some animals adapt their locomotion when ascending; the squirrel and other larger animals wrap their front legs around the substrate for easier pulling while folding their hind legs underneath their body for easier pushing (Cartmill, 1985). Other animals such as snakes keep their COM close to a vertical substrate by circling and folding their body around it (Astley and Jayne, 2007).

In addition to the system used for gripping, size is an important factor in climbing. Larger animals must find strong structures to support themselves. The heaviest arboreal animals known are orangutans, which can weigh up to 90 kg (Kay, 1974). Climbing also brings the risk of falling, which is more damaging for larger animals whose bones are relatively weaker with a smaller safety factor (Biewener, 1982). Another aspect is the cost of climbing, which scales differently than that of horizontal locomotion. Smaller animals moving horizontally use more energy per unit mass than larger animals, yet the additional costs of climbing are constant per unit mass. The additional costs of climbing are therefore

proportionally smaller in smaller animals (Taylor et al., 1972). From an energetics standpoint the locomotion of small animals should be less affected by orientation. This has been seen in desert ants (*Cataglyphis fortis*) and wood ants (*Formica pratensis*) whose walking kinematics were, after correcting for speed, only little affected by slope (Seidl and Wehner, 2008). The same is true for gecko species, *Gekko gecko* and *Eublepharis macularius* (both weighing about 50 g), although the climbing performance of the ground-dwelling species (*E. macularius*) was limited in speed (Zaaf et al., 2001). However, the stride length and swing duration of the hindlimb in heavier iguana lizards (*Dipsosaurus dorsalis* about 60 g) were found to change when moving from horizontal to a 30 deg. slope (Jayne and Irschick, 1999).

All of these examples involve animals with stiff, articulated skeletons with easily quantifiable kinematics. Soft-bodied animals such as caterpillars are also excellent climbers whose motions during climbing closely resemble those in other orientations. *Manduca sexta* (Linnaeus 1763) initiates crawling with the tip of its abdomen, the terminal prolegs (TP) and continues with an anterior grade wave of steps with its remaining four pairs of prolegs (see Fig. 1B–D). The three pairs of thoracic legs also undergo an anterior grade wave of steps; however, these movements are less consistent than those of the prolegs (Johnston and Levine, 1996). In addition, the prolegs have a strong gripping system and they contribute more to locomotion than the thoracic legs. During crawling, muscles are activated to ‘unhook’ the prolegs. The prolegs are then lifted (not shortened) and carried forward by waves of segmental contractions (Belanger and Trimmer, 2000; Trimmer and Issberner, 2007). This might have implications for locomotion in the vertical orientation as the COM is moved further away from the substrate. Furthermore, the location of the COM is likely to vary during locomotion even when measured in relation to the body. There are no septa dividing body segments, so body fluids and tissues can be displaced along the anterior–posterior axis. For instance, the gut does not move in tandem with the moving body segments during locomotion (M. A. Simon and B.A.T., unpublished data). During locomotion some body segments stretch while others do not and, as body segments do not

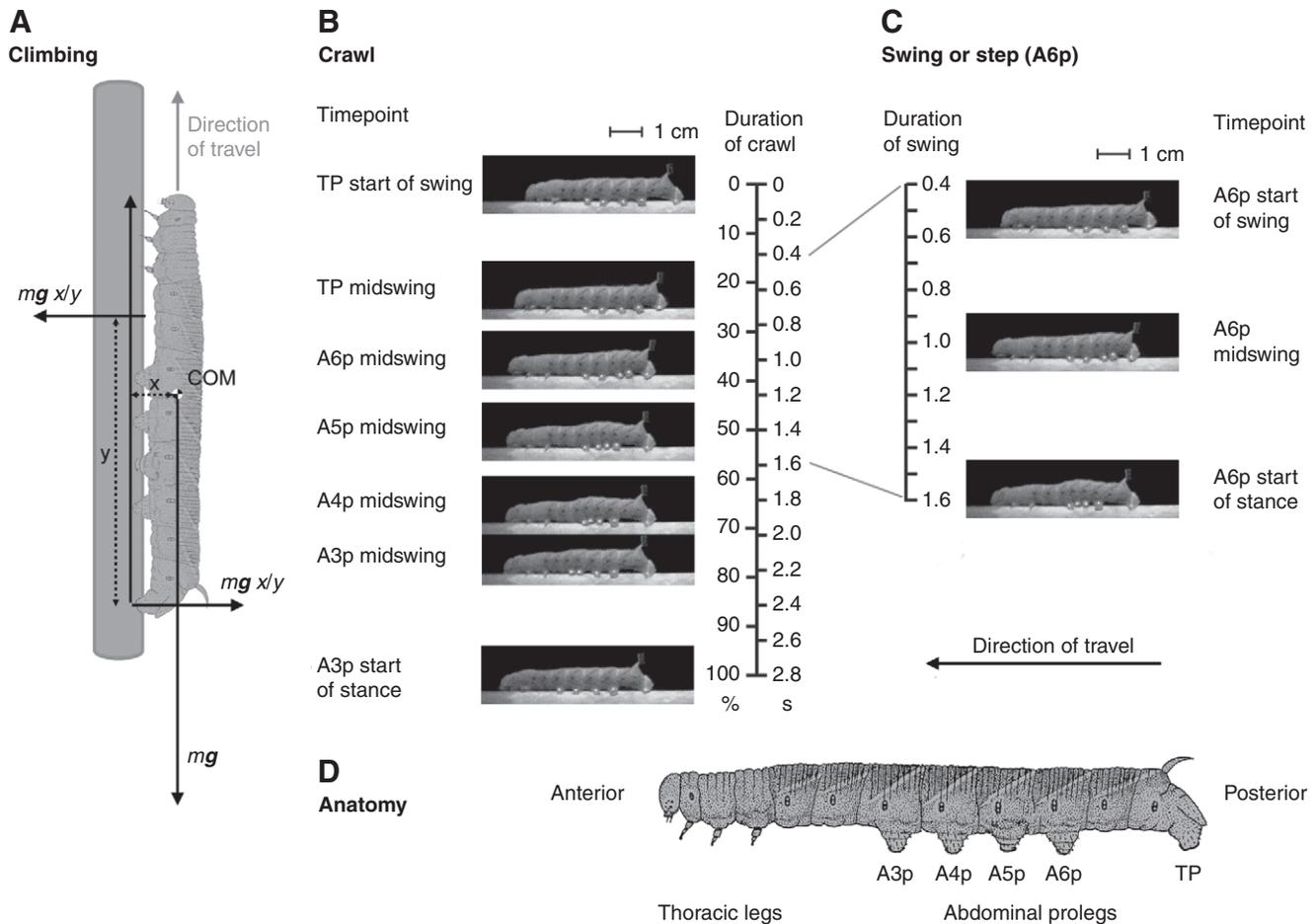


Fig. 1. (A) Forces acting on *Manduca sexta* in a vertical position. The caterpillar has to counteract its weight ($=mg$: mass times gravitational acceleration) by moving upward as well as avoid turning (torque) by pulling its body towards the substrate with the legs above the centre of mass (COM) and pushing it away with those legs below the COM. The COM is estimated to be located between segment A3 (abdominal segment three) and A4 (abdominal segment four). As the thoracic legs are believed to contribute little to gripping the substrate, the $mg \ x/y$ vector on the upper body is expected to be located posterior to the thoracic legs. (B) A full crawl starts with the TP (terminal prolegs) lifting away from the substrate. Each proleg is lifted and placed forward. The proleg on abdominal segment three (A3p) touching down is defined as the end of the crawl. In this individual, a crawl lasted 2.8 s. Midswing indicates the time point at which the velocity of a proleg along the substrate is maximal. The swing phase of a proleg overlaps with the swing phase of its predecessor and its successor. (C) The swing phase for the proleg on abdominal segment A6 (A6p), starting with the start of the swing phase and ending with the start of the stance phase. (D) Anatomy of a second day fifth-instar of the *Manduca sexta* larva.

have a fixed volume as seen in earthworms, this drastically complicates locating the COM along the body axis. Although the COM might change relative to the body when the caterpillar is moving, during rest in a horizontal orientation the COM is located between A3 (abdominal segment three) and A4 (abdominal segment four).

Given these marked differences from other climbing animals, it is interesting to compare the kinematics of upright horizontal and upward vertical crawling. Our hypothesis is that *M. sexta* larvae should minimize torque during vertical climbing by adjusting their kinematics. Minimizing torque can prevent bending of the body and also minimizes the forces with which a caterpillar needs to pull its upper body towards the substrate and push its lower body away from the substrate.

MATERIALS AND METHODS

Animals

Both male and female larval *Manduca sexta* (2.048 ± 0.139 g for 20 out of 25 animals) were used in this experiment. The animals were

reared on an artificial diet in an incubator with a 17h:7h L:D cycle at 27°C (Bell and Joachim, 1978). Animals used for experiments were in their second day of the fifth-instar. Data were collected from 25 animals.

Preparation

Before data collection, the *M. sexta* were anesthetized on ice for 30–60 min and five 168 μ m diameter fluorescent beads (Duke Scientific, Palo Alto, CA, USA) were attached to the cuticle using Elmer's rubber cement (Elmer's Products, Columbus, OH, USA). Beads were attached to the tip of each left proleg, above the planta, as well as on the left TP (see Fig. 1B–D). The animals were placed on wooden dowels (0.78 cm in diameter) and given time to fully recover from the anaesthesia. The same substrate was used in the experiments.

Experiment

The crawling substrate was illuminated by an ultraviolet lamp (Model B, 100 W, long wavelength; Blak-Ray, Upland, CA, USA).

Crawling behaviour was captured on a digital camera (Canon ZR10, Canon USA, Lake Success, NY, USA) with a Hoya green (X1) filter (Edmund Optics, Barrington, NJ, USA) at $29.97 \text{ frames s}^{-1}$, transferred to a computer using an IEEE 1394 multimedia connection and recorded through Video Wave Professional 7 software (Roxio, Santa Clara, CA, USA). All the crawling used in this analysis was in a straight line without lateral movements hence a second camera was not essential. Both the crawling substrate and the camera were mounted to a frame that could be rotated to study both horizontal and vertical upward crawling while keeping the camera position (and aspect ratio) fixed relative to the caterpillar.

Data analysis

Using APAS software (Ariel Performance Analysis System, v. 1.0, Ariel Dynamics, San Diego, CA, USA) video recordings were cropped, making sure that for each animal at least five crawls (five waves of steps, each proleg lifted five times in total) were visible both in vertical and horizontal orientations. The positions of the beads were tracked relative to a fixed point in space and the workspace was calibrated using a custom-built marker frame. The

position, displacement and velocity of the beads were calculated off-line using a 2-D direct linear transform. Further analysis was done in Sigmaplot 2000 v. 6.10 (SPSS Inc., Chicago, IL, USA). The following parameters were calculated.

Proleg lift

To isolate and combine data for proleg lift during individual steps, the times of successive peaks in forward velocity were determined from smoothed data (Loess, sampling proportion, 0.1; polynomial degree, 1) using Data View (v. 4.7, W. J. Heitler, University of St Andrews, Scotland). The displacement of A3p (proleg on abdominal segment three), A4p (proleg on abdominal segment four), A5p (proleg on abdominal segment five), A6p (proleg on abdominal segment six) and TP in the y -plane (perpendicular to the wooden dowels in both orientations) was determined after subtracting systematic changes in baseline position [lowess-filtered baseline with smoothing parameter of 0.8 (Porges and Bohrer, 1990)]. The timing of the peaks in forward velocity was used to align displacement data for successive steps in each segment and to calculate mean displacement perpendicular to the substrate (proleg lift) for each

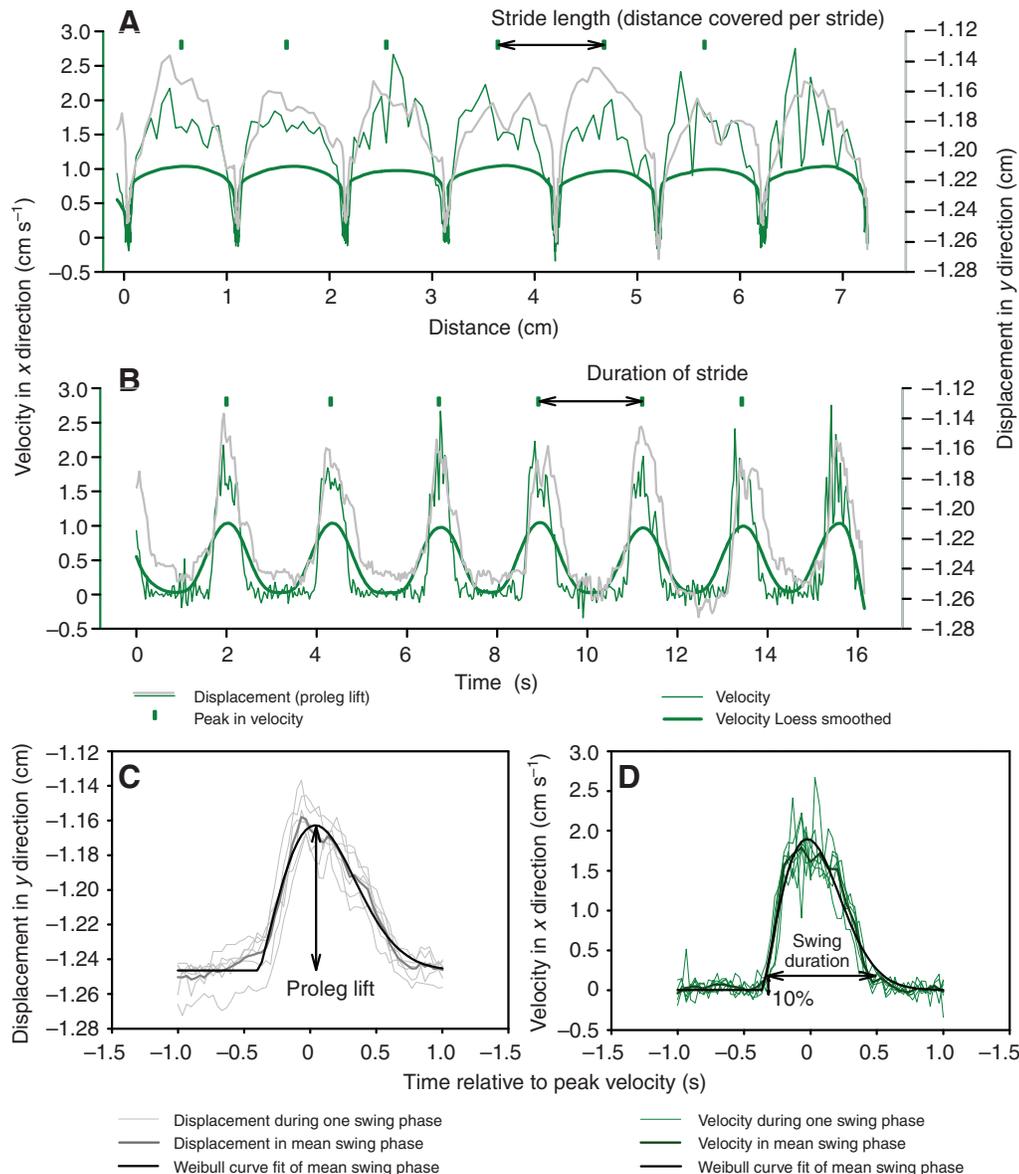


Fig. 2. Data for A3p (prolegs on abdominal segment three) in one individual caterpillar in the horizontal orientation. (A) Velocity of A3p in the x -plane and displacement of A3p in the y -plane versus distance moved along the substrate. The timing of the maximum in velocity was used to calculate stride length. (B) Velocity of A3p in the x -plane and displacement of A3p in the y -plane versus time. The timing of the maximum velocity was used to calculate the duration of stride. (C) The displacement of A3p away from the substrate over several steps (thin gray lines) was averaged (thick dark gray line) and smoothed (black line). Proleg lift is the maximum lift of the smoothed curve. (D) The velocity of A3p along the substrate over several steps (thin green lines) was averaged (thick dark green line) and smoothed (black line). The duration of the swing was calculated at 10% of the maximum velocity.

segment over several steps. The maximum proleg lift in the mean step was determined from a Weibull curve fit (see Fig. 2C). Differences in caterpillar sizes are corrected for by paired statistical analysis.

Swing duration

When a leg is in stance phase, its point of contact with the substrate should have a velocity of zero. Velocity along the substrate was used to calculate the duration of swing. For each animal, the mean velocity over a step was calculated using three to seven steps per animal per orientation as described above. This mean step was then used to determine the timing of the start and the stop of the swing phase from a Weibull curve fit. Start and stop were measured at 10% of the maximum velocity for each proleg and used to calculate swing duration (see Fig. 1C; Fig. 2D).

Stride length

A stride for each proleg consists of swing and stance phases. Hence, the distance moved during the swing phase is the stride length. Stride

length was calculated by determining the time between successive peaks in proleg velocity and measuring how much the leg had moved along the substrate during that time. This was calculated for A3p as the stride length should be similar for all prolegs. The mean stride length per animal was calculated for each orientation (see Fig. 2A).

Crawling speed and stride frequency

Crawling speeds were calculated for each stride and averaged for each animal in each orientation. Stride frequency was calculated from the timing of proleg velocity peaks (see Fig. 2B).

Timing within crawl, duration of crawl and total duration of swing

A crawl is a wave of steps, beginning with the onset of swing in TP and ending with the onset of stance in A3p. Crawls can overlap in time. Therefore, a stride or cycle (e.g. start of swing of TP to next start of swing of TP) is not appropriate for comparing the timing of prolegs relative to each other; a new stride can start before a crawl is finished and there is considerable variation among animals

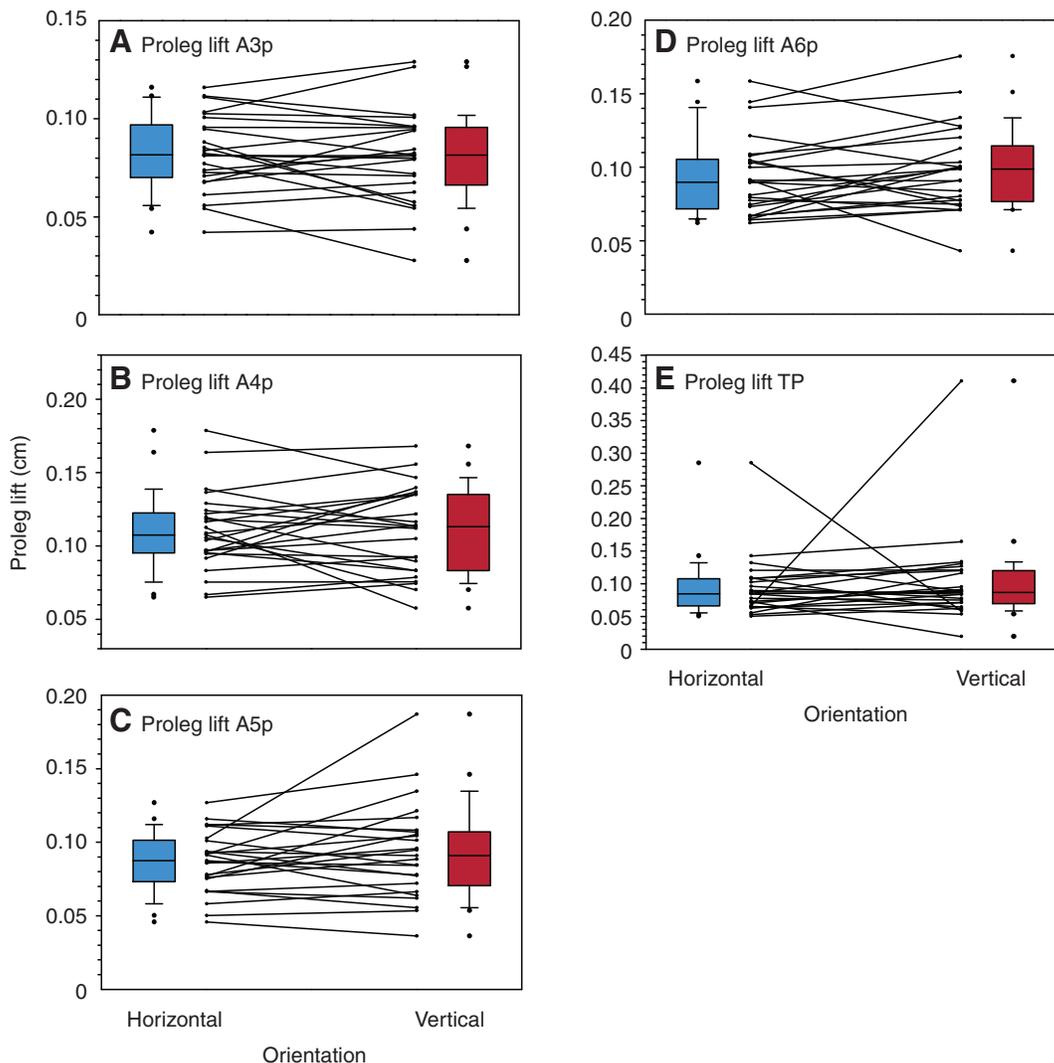


Fig. 3. A comparison of proleg lift for each proleg during horizontal and vertical crawling. The maximum proleg lift distance away from the substrate is shown for prolegs A3p (prolegs on abdominal segment three) to TP (terminal prolegs) (A–E) for caterpillars crawling horizontally (points at the left) or vertically (points on the right). Each line joins data points for an individual caterpillar. The boxplots show quartile 1 (Q1), median and quartile 3 (Q3) for all caterpillars ($N=25$). Outliers, indicated by dots, are 1.5 interquartile range or more removed from Q1 or Q3.

in the timing between crawls. The coordination of proleg movements relative to each other was calculated from the times between peaks in the velocities of successive prolegs in a crawl. In combination with data about the duration of the swing phases, the total duration of crawl was calculated (see Fig. 1B). To find differences in the timing of the prolegs (and to compensate for speed differences), the time points were expressed as a percentage of the entire crawl; the onset of swing phase in TP was defined as 0% and the onset of stance phase of A3 at the end of the wave of steps was defined as 100%. The beginning of swing, the timing of the peak of velocity and the beginning of stance phase were then compared for the horizontal and vertical orientations. These data were also used to calculate the duration of swing for each proleg relative to the duration of the crawl. Durations of swing (as a percentage of the duration of the crawl) for all five prolegs were summed and compared for horizontal and vertical crawling.

Timing between crawls

To standardize for different crawling speeds, swing and stance timing for each proleg were normalized to the time taken to complete a stride (see Fig. 2B). The peak of velocity in TP was considered 0% and the peak of velocity in TP in the next crawl was considered 100%. The timing of peak velocity of A3p was then expressed as a percentage of the stride duration.

Statistical analysis

Stride length, crawling speed, timing between crawls, stride frequency, duration of crawl and total duration of swing in the horizontal and vertical orientation were compared using paired *t*-tests in SPSS (v. 16.0.1, SPSS Inc.). Proleg lift, swing duration and timing within crawls were tested using repeated-measures MANOVAs in JMP (v. 5.0.1.2, SAS Institute, Cary, NC, USA). Descriptive statistics are indicated in means \pm standard error (\pm s.e.m.).

RESULTS

There were no significant differences in the distance the caterpillars lifted their prolegs away from the substrate when comparing

horizontal upright and vertical upward locomotion ($F=0.2354$ d.f.=4, 192 $P=0.918$). Fig. 3 shows the pairwise comparison for each proleg. Proleg lift can be higher or lower in individual caterpillars crawling vertically; however, there is no trend. There were also no significant differences in the durations of the swing phase for each proleg ($F=0.5117$ d.f.=4, 192, $P=0.727$), the duration of a crawl ($t=1.460$, d.f.=24, $P=0.1572$) and the timing of the beginning of swing, midswing and beginning of stance within a crawl ($F=1.0194$ d.f.=12, 576, $P=0.429$). In other words, the crawling pattern looks very similar as is illustrated in Fig. 4. There was no difference in total duration of swing during a crawl ($t=0.859$, d.f.=24, $P=0.3987$).

The caterpillars covered significantly more distance per stride in the horizontal orientation (0.852 ± 0.022 cm) than in the vertical orientation (0.801 ± 0.020 cm) ($t=2.918$, d.f.=24, $P=0.0075$). As Fig. 5 shows this difference is small compared with the variation between animals. The caterpillars' stride frequencies were higher in the horizontal orientation (21.34 ± 1.50 strides min^{-1}) than in the vertical orientation (17.31 ± 1.36 strides min^{-1}) ($t=3.064$, d.f.=24, $P=0.0053$). The timing of A3p swing in one stride (midswing TP to midswing TP) was significantly later in the horizontal ($54.66\pm 1.49\%$) than in the vertical orientation ($49.44\pm 1.47\%$) ($t=3.486$, d.f.=24, $P=0.0019$) (Fig. 6). This increase in time between crawls and, together with the larger distance covered per crawl, contributed to slower crawling in the vertical orientation (0.241 ± 0.021 cm s^{-1}) compared with the horizontal orientation (0.310 ± 0.024 cm s^{-1}) ($t=3.165$, d.f.=24, $P=0.0042$). An overview of the parameters tested, descriptive statistics and the results of comparing horizontal and vertical locomotion can be found in Table 1.

DISCUSSION

The environment and workspace of *Manduca*

Manduca caterpillars are extremely effective climbers. They hatch directly on their food source and move around to reach new leaves as they devour those that are closest. They do not need to move quickly [except during the strike reflex (Walters et al., 2001)] and mostly remain hidden by holding onto the stem or reaching to eat from the underside edges of leaves. They can crawl from branch to

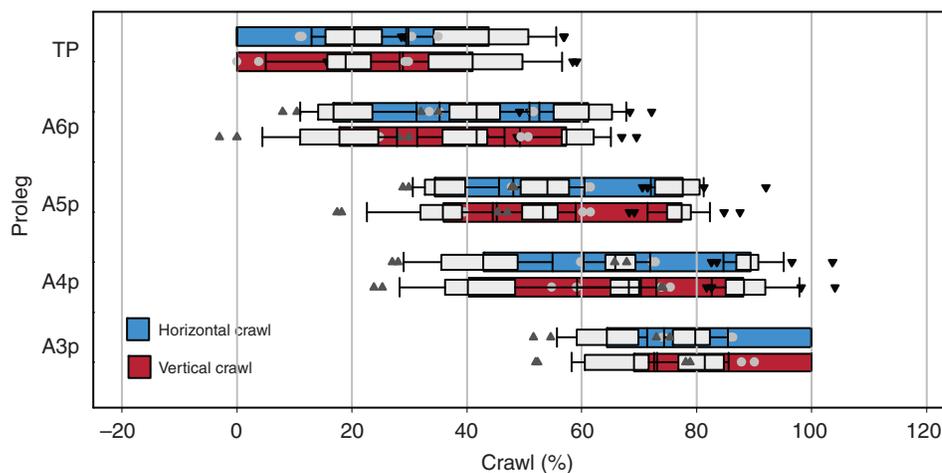


Fig. 4. Timing of swing, midswing and stance of each proleg within a crawl. Onset of swing in TP (terminal prolegs) is considered to be 0% and onset of stance in A3p (prolegs on abdominal segment three) is considered to be 100%. For each proleg, the relative timing of (from left to right) the beginning of swing, midswing and beginning of stance were calculated and displayed as boxplots [quartile 1 (Q1), median and quartile 3 (Q3) for all caterpillars ($N=25$, 3–7 crawls averaged for each animal)]. Outliers indicated by dark gray triangles for the start of swing, light gray dots for midswing and black triangles for the start of stance, are 1.5 interquartile range or more removed from Q1 or Q3. Swing duration is indicated by the blue (horizontal crawl) and red (vertical crawl) boxes. A4p, prolegs on abdominal segment four; A5p, prolegs on abdominal segment five; A6p, prolegs on abdominal segment six.

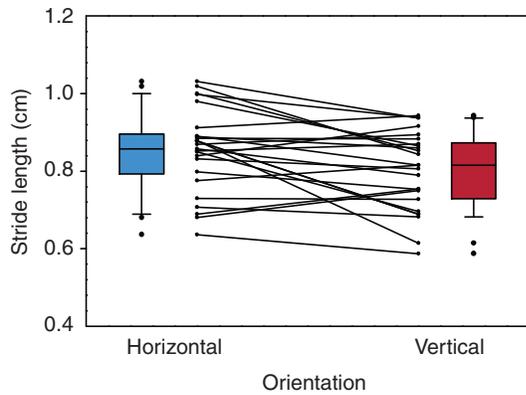


Fig. 5. Stride length (distance covered per crawl = distance covered per swing phase) as measured for A3p (prolegs on abdominal segment three) in caterpillars crawling horizontally (points at the left) or vertically (points on the right). Each line joins data points for an individual caterpillar. The boxplots show quartile 1 (Q1), median and quartile 3 (Q3) for all caterpillars ($N=25$). Outliers, indicated by closed dots, are 1.5 interquartile range or more removed from Q1 or Q3.

branch by lifting their anterior portions away from the substrate and casting back and forth until they contact a new surface and then by bridging gaps that can be almost an entire body length. During the later part of the fifth-instar, the larvae begin 'wandering' behavior in which they climb down the plant to the ground and move considerable distance away before burrowing to begin pupation.

For most animals living in three-dimensional complex environments, gravity would be expected to be an important force and indeed most animals have special gravity-sensing organs. In larger mammals, these are the semi-circular canals that can sense orientation and head acceleration. For *M. sexta* no special gravity sensors have been reported but behavioral evidence suggests that they do sense gravity, perhaps through proprioception. First, on the substrate we used for testing, the caterpillars tended to circle around the dowel in the vertical orientation but would stay on top of the dowel in the horizontal orientation. Circling a vertical substrate keeps the COM close to the substrate, which can be advantageous when climbing on bending twigs. Similar behavior is seen in climbing snakes (Astley and Jayne, 2007). Second, if the planta hairs are touched during locomotion the caterpillar withdraws that proleg; reattachment to the substrate occurs faster in animals hanging upside down than in animals on top of a dowel (Belanger et al., 2000).

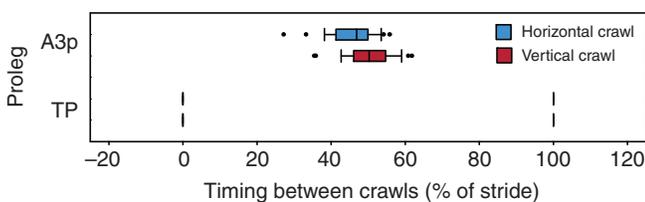


Fig. 6. Timing of midswing of A3p (prolegs on abdominal segment three) within one stride of TP (terminal prolegs). Midswing TP was considered to be 0% and the next midswing of TP was considered to be 100%. The timing the midswing of A3p is displayed as a boxplot [quartile 1 (Q1), median and quartile 3 (Q3) for all caterpillars ($N=25$, 2–7 crawls averaged for each animal)]. Outliers, indicated by closed dots, are 1.5 interquartile range or more removed from Q1 or Q3. Horizontal crawl is indicated in blue, vertical crawl is indicated in red.

Crawling is robust for climbing

When an animal climbs upward there are two effects of gravity that are not present in horizontal crawling: (1) a backwards force that resists forward motion and (2) torque pulling and pushing different body parts in the plane of the substrate. Both forces are strongly influenced by the weight of the animal (see Fig. 1). Although *M. sexta* caterpillars appear to sense gravity, their kinematics differ very little between crawling upright horizontally and ascending vertically. Even for kinematic parameters that were significantly different in each orientation the differences were extremely small compared with those between different animals. No combination of measurements can be used to determine if an animal is crawling horizontally or climbing.

Similarly small differences in kinematics have been found in other animals within *Manduca*'s weight range. For example, when the effect of speed was removed, running cockroaches (<1 g) had the same stride frequency and contact time running horizontally or on inclined surfaces (Full and Tullis, 1990; Goldman et al., 2006). Geckos (~2 g) also have similar kinematics when running horizontally and vertically (Autumn et al., 2006).

Torque considerations generally demand that the body is kept close to the substrate during climbing but proleg lift was unaffected in *Manduca*. Similarly, proleg swing duration and the relative timing of the proleg movements within a crawl were independent of orientation. These results however cannot exclude the possibility of the timing and sequence of muscle activation within a crawl in horizontal locomotion being different from those of a climbing caterpillar. Also, the level of activation might be altered in order to keep the kinematics the same [as observed in climbing cockroaches (Watson et al., 2002)] but this remains to be confirmed by simultaneous electromyography. In addition, ground reaction forces (GRF) can be different while kinematics are the same (Jindrich and Full, 1999). GRF measurements could give more insight in the location and magnitude of forces acting on a climbing caterpillar. There is a wide variation in each kinematic parameter, yet these parameters change little between the horizontal and vertical orientation. This indicates that the caterpillars' mode of locomotion is relatively insensitive to the orientation the animal is in.

Effects of climbing on movements

Clearly, the need to lift the body during climbing has some consequences for locomotion even in smaller animals. Locusts (*Schistocerca gregaria* ~2 g) have similar leg swing duration in horizontal, vertical and upside down locomotion. However, the time between one flexor burst and the next is much longer when climbing, resulting in slower locomotion (Duch and Pflüger, 1995). *Manduca* also climbed more slowly (on average) than they crawled horizontally. This was the consequence of two changes: (1) each crawl covered less distance (94% of that covered in the horizontal orientation) and (2) the delay between crawls was longer (~10%). These differences are extremely small and are unlikely to have a major effect on normal survival or growth. *Manduca sexta* caterpillars are cryptic and do not rely on speed for survival from predators nor do they compete with other species for most of their native food sources (Kingsolver and Woods, 1997; Ojeda-Avila et al., 2003). Furthermore, vertical climbing costs for small animals are only a small component of the total costs of locomotion (Taylor et al., 1972) and therefore are not a major factor in the kinematics. For soft-bodied animals, the cost of transport is already relatively high and for caterpillars it is 4.5 times higher than that predicted for animals with similar masses with stiff skeletons (Casey, 1991). Presumably energy is spent on tissue movements along and within

Table 1. (A) Descriptive statistics and outcome of repeated-measures MANOVA (both between and within groups) of proleg lift, duration swing and timing within crawl. (B) Descriptive statistics and outcome of two-sided paired *t*-tests of stride length, crawling speed, timing between crawls (proportional to a stride), stride frequency, duration of crawl and total duration of swing (sum of duration of swing phase proportional to a stride for each proleg)

(A) Repeated-measures MANOVA						
Parameter	Horizontal means \pm s.e.m. (per proleg)	Vertical means \pm s.e.m. (per proleg)	Between/ within	<i>F</i> -values	d.f.	<i>P</i> -values
Proleg lift (cm)	TP: 0.094 \pm 0.009	TP: 0.102 \pm 0.014				
	A6p: 0.093 \pm 0.005	A6p: 0.098 \pm 0.006	B	0.3602	1, 48	0.5512
	A5p: 0.087 \pm 0.004	A5p: 0.094 \pm 0.007	W	0.2354	4, 192	0.9181
	A4p: 0.110 \pm 0.005	A4p: 0.110 \pm 0.006				
	A3p: 0.083 \pm 0.004	A3p: 0.081 \pm 0.005				
Duration swing (s)	TP: 1.202 \pm 0.106	TP: 1.296 \pm 0.129				
	A6p: 1.132 \pm 0.041	A6p: 1.230 \pm 0.063	B	1.0092	1, 48	0.3201
	A5p: 1.109 \pm 0.044	A5p: 1.261 \pm 0.052	W	0.5117	4, 192	0.7272
	A4p: 1.244 \pm 0.067	A4p: 1.309 \pm 0.097				
	A3p: 0.974 \pm 0.050	A3p: 0.983 \pm 0.042				
Timing within crawl (%)	TP Start: 0 \pm 0	TP Start: 0 \pm 0				
	Mid: 20.70 \pm 1.31	Mid: 18.75 \pm 1.54				
	Stop: 42.57 \pm 1.90	Stop: 41.09 \pm 2.21				
	A6p	A6p				
	Start: 18.80 \pm 1.47	Start: 17.40 \pm 1.77				
	Mid: 41.93 \pm 1.16	Mid: 39.75 \pm 1.35				
	Stop: 60.50 \pm 1.23	Stop: 58.30 \pm 1.17				
	A5p	A5p				
	Start: 36.64 \pm 1.12	Start: 34.76 \pm 1.53	B	0.1757	1, 48	0.6769
	Mid: 53.94 \pm 0.93	Mid: 52.32 \pm 1.124	W	1.0194	12, 576	0.4290
	Stop: 77.33 \pm 0.94	Stop: 77.24 \pm 0.88				
	A4p	A4p				
	Start: 43.49 \pm 2.10	Start: 44.79 \pm 2.98				
	Mid: 66.49 \pm 0.78	Mid: 67.26 \pm 1.01				
	Stop: 89.35 \pm 0.88	Stop: 89.09 \pm 1.11				
A3p	A3p					
Start: 64.28 \pm 1.28	Start: 66.67 \pm 1.45					
Mid: 79.59 \pm 0.79	Mid: 80.35 \pm 1.11					
Stop: 100 \pm 0	Stop: 100 \pm 0					
(B) Two-sided paired <i>t</i> -tests						
Parameter	Horizontal means \pm s.e.m.	Vertical means \pm s.e.m.	<i>t</i> -value	d.f.	<i>P</i> -values	
Stride length (cm)	0.852 \pm 0.022	0.800 \pm 0.020	2.918	24	0.0075	
Crawling speed (cm s ⁻¹)	0.310 \pm 0.024	0.241 \pm 0.021	3.165	24	0.0042	
Timing between crawls (%)	54.66 \pm 1.49	49.44 \pm 1.47	3.486	24	0.0019	
Stride frequency (strides min ⁻¹)	21.34 \pm 1.50	17.31 \pm 1.36	3.064	24	0.0053	
Duration of crawl (s)	2.778 \pm 0.148	3.056 \pm 0.172	1.460	24	0.1572	
Total duration of swing (%)	206.53 \pm 4.36	202.11 \pm 5.62	0.859	24	0.3987	

TP, terminal prolegs; A3p, prolegs on abdominal segment three; A4p, prolegs on abdominal segment four; A5p, prolegs on abdominal segment five; A6p, prolegs on abdominal segment six; d.f., degrees of freedom; s.e.m., standard error of the mean.

the body that do not directly result in locomotion (Casey, 1991). Because caterpillars crawl slowly (0.241–0.310 cm s⁻¹ on average in the present study), and forward momentum is small and episodic, there is no sustained acceleration and the effect of gravity is therefore minimized during vertical climbing. Another reason for the high costs of caterpillar locomotion is that muscles and body wall have low resilience, dissipating a large proportion of mechanical work (40–60%) during cycles of strain (Dorfmann et al., 2008; Dorfmann et al., 2007; Lin et al., 2008; Woods et al., 2008). This inability to store elastic energy makes caterpillar locomotion less efficient.

Adaptations of *Manduca* for movement in complex branched structures

Although the small size and slow speed of *Manduca* are important factors in its climbing ability, perhaps the most important contribution is made by the strong gripping system (the prolegs). Each proleg consists of an extendable pouch on the ventral hemisegment with a fleshy lobe (the planta) at its tip (Barbier, 1985; Snodgrass, 1961). There is a row of hooks (crochets) on the planta that can be deployed to engage with irregularities on the substrate at the start of stance phase. The gripping force may far

exceed other forces acting on the body during crawling and attempts to pull a caterpillar off a rough substrate can result in the crochets tearing out of the planta rather than release from the surface. The gripping system is used regardless of the orientation the caterpillar is in.

The passive state of the prolegs is in the adducted position with the crochets extended by tonic body pressure and body wall elasticity (Mezoff et al., 2004). Release is achieved by unhooking the crochets and retracting the prolegs by active muscle contraction (Belanger and Trimmer, 2000; Belanger et al., 2000; Mezoff et al., 2004). As the gripping system is equally strong in horizontal and vertical orientation, the hooking and unhooking activities do not lead to differences in the kinematics. It has been argued that a strong gripping system in climbing can be seen as the animal creating a non-vertical surface for itself, like climbing a ladder (Cartmill, 1985). The interlocking grip on the substrate prevents an ascending or descending caterpillar from rotating backwards or sideways due to torque. The strong gripping system in caterpillars also serves another function: lateral stability. Unlike sprawled legs, caterpillar prolegs on each segment are located relatively close to each other. A strong gripping system is necessary to avoid rolling sideways when crawling horizontally. As Brackenbury argues, this phenomenon also leads to a high duty factor, which limits the crawling speed (Brackenbury, 1999).

LIST OF ABBREVIATIONS

A3	abdominal segment 3
A4	abdominal segment 4
A3p	prolegs on abdominal segment 3
A4p	prolegs on abdominal segment 4
A5p	prolegs on abdominal segment 5
A6p	prolegs on abdominal segment 6
COM	center of mass
deg.	degree(s)
<i>g</i>	gravitational acceleration
GRF	ground reaction forces
<i>m</i>	mass
s.e.m.	standard error or the mean
TP	terminal prolegs

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