

When cats need to see to step accurately?

Maxim Volgushev¹ , Celina T. Nguyen^{2,3}, Gautam S. Iyer² and Irina N. Beloozerova^{2,4} 

¹Department of Psychological Sciences, University of Connecticut, Storrs, CT, USA

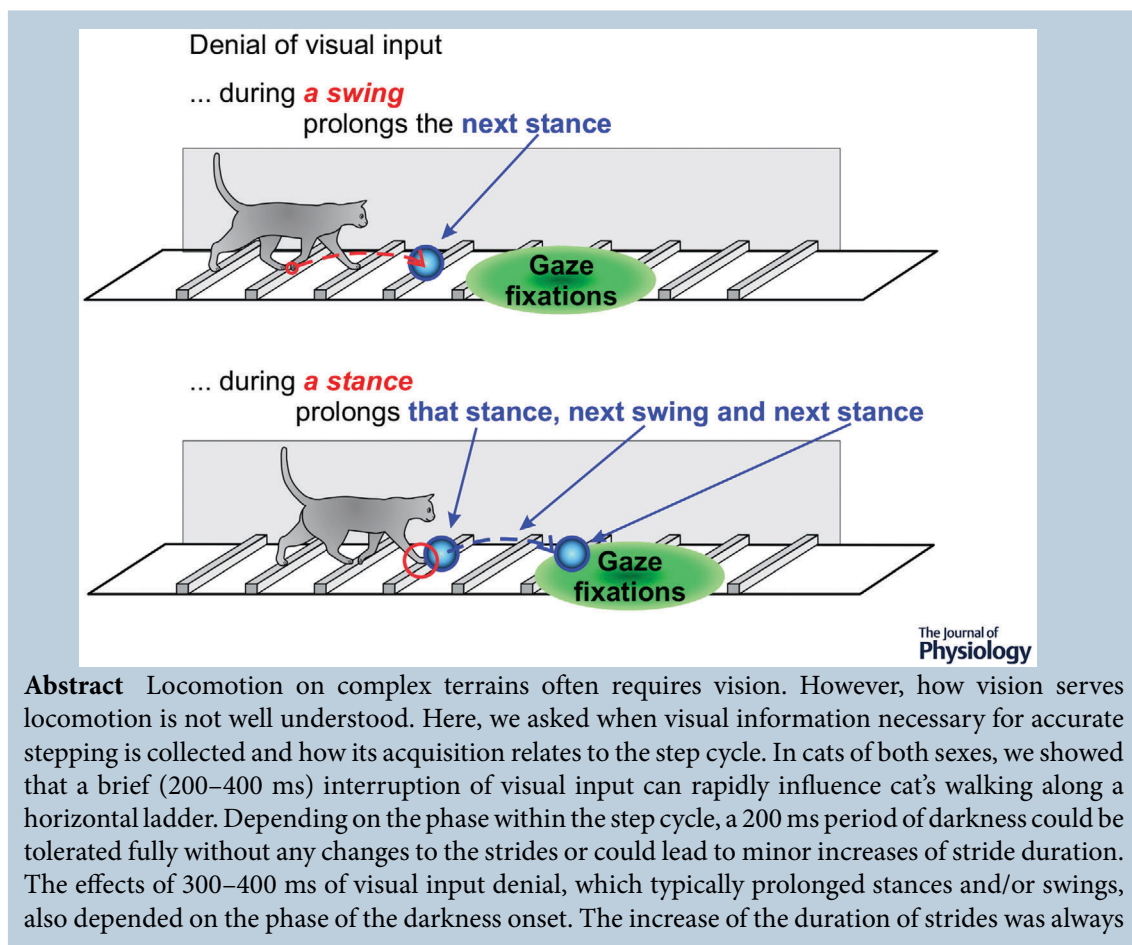
²Barrow Neurological Institute, St Joseph's Hospital and Medical Center, Phoenix, AZ, USA

³Neurosciences Graduate Program, University of California San Diego, La Jolla, CA, USA

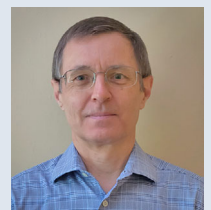
⁴School of Biological Sciences, Georgia Institute of Technology, Atlanta, GA, USA

Edited by: Richard Carson and William Taylor

The peer review history is available in the Supporting Information section of this article (<https://doi.org/10.1113/JP282255#support-information-section>).



Maxim Volgushev studied physiology and higher brain functions at Lomonosov Moscow State University. After a PhD in visual neurophysiology (Moscow) and postdoctoral studies on mechanisms of response selectivity in visual cortex (Göttingen) and synaptic transmission and plasticity (Frankfurt), he continued research into these topics in his laboratory at Ruhr-University Bochum (1996–2007; habilitation in Physiology, 2000) and, since 2007, in the University of Connecticut. Other research interests include fast neuronal encoding and action potential generation, origin of slow oscillations, and regulation of synaptic transmission, plasticity and learning by adenosine. The present work on vision for locomotion is an expansion into the exciting field of sensorimotor integration.



shorter than the duration of darkness. We conclude that visual information for planning a swing is collected starting from the middle of the preceding stance until the beginning of the current swing. For a stance (and/or a swing of the other paw), visual information is collected starting from the end of the previous stance and until the middle of the current stance. Acquisition of visual information during these windows is not uniform but depends on the phase of the step cycle. Notably, both the extension of these windows and their non-homogeneity are closely related to the pattern of gaze behaviour in cats, described previously. This new knowledge will help to guide research and understanding of neuronal mechanisms of visuomotor integration and modulation of visual function by strides during locomotion.

(Received 10 August 2021; accepted after revision 29 October 2021; first published online 11 November 2021)

Corresponding author Maxim Volgushev, Department of Psychological Sciences, University of Connecticut, Storrs, CT 06269, USA. Email: maxim.volgushev@uconn.edu

Abstract figure legend Accurate stepping on crosspieces of a horizontal ladder requires vision. We show that collection of visual information for locomotion is not uniform throughout the stride cycle. In cats, brief denial of visual input during a swing prolongs the next stance of that forelimb. Denial of visual input during a stance prolongs this stance, and the next swing and stance. This step-cycle dependence of the acquisition of visual information for locomotion and the pattern of affected swings and/or stances are closely related to the pattern of gaze behaviour in cats, described previously.

Key points

- Cats, like humans, rely on vision to navigate in complex environments.
- In cats walking along a horizontally placed ladder, we show that visual information required for accurate stepping is collected in a non-uniform manner throughout the stride cycle.
- Brief denial of visual input during a swing prolongs the next stance of that forelimb. Denial of visual input during a stance prolongs this stance, as well as the next swing and stance. Denial during the first half of a stance has a greater effect than during the second half.
- The phase dependence of the use of vision for accurate stepping and the pattern of affected swings and stances are closely related to the previously described pattern of gaze behaviour in cats.
- This new knowledge opens new perspectives for research into neuronal mechanisms of visuomotor coordination and visual function during walking and for understanding related disorders.

Introduction

Cats, like humans, rely on vision for locomotion on a complex terrain. However, it is not well understood how vision is used during locomotion that requires accurate stepping. Given that locomotor movements are periodic in nature, the recurring step cycle sets an intrinsic clock for other related processes. Within each cycle, visual information about future targets for foot landing is collected and processed to be used by motor systems for adjusting locomotor programs for accurate stepping. These motor programs for moving a foot and landing it on a target are then executed. The cyclic nature of planning and execution of motor programs during locomotion predicts that demands on vision will change during the step cycle. Indeed, both humans and cats shift their gaze along the walking path in synchrony with strides, looking at targets for the next one to

four foot landings (Hollands *et al.* 1995; Hollands & Marple-Horvat, 1996; Rivers *et al.* 2014; Matthis *et al.* 2017, 2018; Zubair *et al.* 2019). In humans, a brief denial of visual input for 400–500 ms at the end of stance increases its duration, but denial during a swing does not prolong the swing (Hollands & Marple-Horvat, 1996), showing that acquisition, processing and use of visual information depend on the phase of the step cycle. For cats, which provide a valuable model for studies of neuronal mechanisms of the use of vision during locomotion and, in a broader context, of visuomotor integration, knowledge about step-cycle dependence of the demand on visual input is missing.

Previous research in cats walking on a horizontal ladder revealed a characteristic relationship of the pattern of gaze behaviour to the step cycle (Rivers *et al.* 2014; Zubair *et al.* 2019). Gaze fixations occur around the middle of a forelimb swing, which is the middle of stance of the

other forelimb. Episodes of constant gaze, i.e. looking at a constant distance ahead while walking, is another gaze behaviour during which visual information can be collected. Constant gaze episodes occur at the beginning of stance of one forelimb and the end of stance of the other forelimb, during the time when both forelimbs are on the crosspieces of the ladder. Notably, qualitatively similar periodic gaze behaviour synchronized with the step cycle was observed in cats walking not only in complex environments that require vision for accurate stepping, but also on a flat surface, which does not require vision, and even when cats were walking on a flat surface in complete darkness, without any visual input (Zubair *et al.* 2019). Thus, control of gaze during locomotion is not just vision-driven, but is an intrinsic component of the ongoing locomotor behaviour. This implies that a gaze fixation or an episode of a constant gaze directed at the pathway does not necessarily reflect the need for collecting visual information for accurate stepping. Rather, some fixations and constant gaze episodes could reflect a locomotion-set pattern of gaze movements but are redundant for the task of accurate stepping. Such redundancy predicts that, after a brief period of visual input denial, a compensatory increase of the duration of strides necessary for acquiring essential visual information will be brief, shorter than the input denial.

To test these predictions and to study the dynamics of demand on vision during locomotion, we used brief periods of denial of visual input (light off for 200, 300 or 400 ms) starting at different phases of the step cycle in cats walking along a horizontal ladder. A brief account of a part of this study was published in abstract form (Nguyen *et al.* 2019).

Methods

Ethical approval

All procedures and experiments were conducted in accordance with US National Institutes of Health guidelines and with the approval of the Barrow Neurological Institute Animal Care and Use Committee (protocol #236, assurance #A3519-01). The authors understand the ethical principles under which *The Journal of Physiology* operates and confirm that this study complies with the animal ethics checklist (Grundy, 2015).

Animals

Three cats purchased from a certified commercial class B dealer were used for experiments: FM (male, 3.7 kg), TI (male, 3.8 kg) and SF (female, 2.9 kg). Cats TI and SF were intact throughout the study. Cat FM was intact during the first two recording sessions; he was then implanted with electrodes for recording neuronal activity in the motor

cortex, thalamus and cerebellum for a different project (data not reported here) and, after recovery, used for an additional seven recording sessions for the present study.

Locomotion on a horizontal ladder

Positive reinforcement (food) was used to adapt cats to the experimental situation and to engage them in locomotion (Skinner, 1938; Pryor, 1975). Cats walked in an experimental chamber that had a test corridor, where cats walked by stepping on crosspieces of a horizontal ladder (Fig. 1A; 2.5 m $\times$ 0.3 m), and a return corridor with a flat floor. The crosspieces of the ladder were 5 cm wide to securely accommodate the cat's paw (which is $\sim$ 3 cm in diameter; e.g. de Carvalho *et al.* 2015). Crosspieces were elevated 6 cm above the floor, and their centres were spaced by one-half of a cat's average stride length during locomotion in the chamber with a flat floor (25 cm; Beloozerova & Sirota, 1993; Beloozerova *et al.* 2010). Cats walked at a self-selected speed. After each round through both the test and return corridors, food was dispensed into a feeding dish in a corner of the chamber, regardless of the cat's performance on the ladder. During a recording session, a cat walked in the experimental chamber for between 35 min and 2.5 h, completing 80–218 passages through the chamber. On an experimental day, only one recording session was conducted.

All three cats were veteran animals in the laboratory. Before experiments for the present study were started, the cats were trained to walk in an experimental chamber for a food reward and were used to obtain data for other studies (e.g. Chu *et al.* 2017). They completed many hundreds of passages along the ladder used in these experiments and similar ladders for other studies and were also familiar with the possibility that lights in the room might go off while they walk in the chamber.

Recording strides

The strides were recorded using a low-voltage (2–5 mV) circuit signalling contact of sensors on the paws with the supporting surface. The tops of the crosspieces of the ladder and the floor of the chamber were covered with an electrically conductive rubberized material, which was connected to the common ground. The cats were accustomed to wearing electromechanical sensors on their paws (Beloozerova & Sirota, 1993; Rivers *et al.* 2014; Zubair *et al.* 2019). The sensors (a metal plate $\sim$ 0.1 mm thin, $\sim$ 10 mm in diameter) were attached to the soles of the forepaws with electro-isolative adhesive tape and connected with wires to a standard cat collar and from there to the recording equipment. Contact of the paw with the grounded surface (beginning of a stance) resulted in a reduction of voltage in the sensor circuit; interruption of the contact (end of stance, beginning

of a swing) resulted in restoration of the voltage. The signals were recorded, along with gate and light-level signals (Fig. 1), using the computerized data acquisition and analysis package Power-1401/Spike-2 (Cambridge Electronic Design, Cambridge, UK). The recorded signals

allowed detection of the timing of swings and stances using a simple threshold (Fig. 1*B–D*). We will refer to the full movement cycle of one limb from the beginning of a swing to the beginning of the next swing of the same limb as a step cycle or stride, and to one-half of such a cycle that

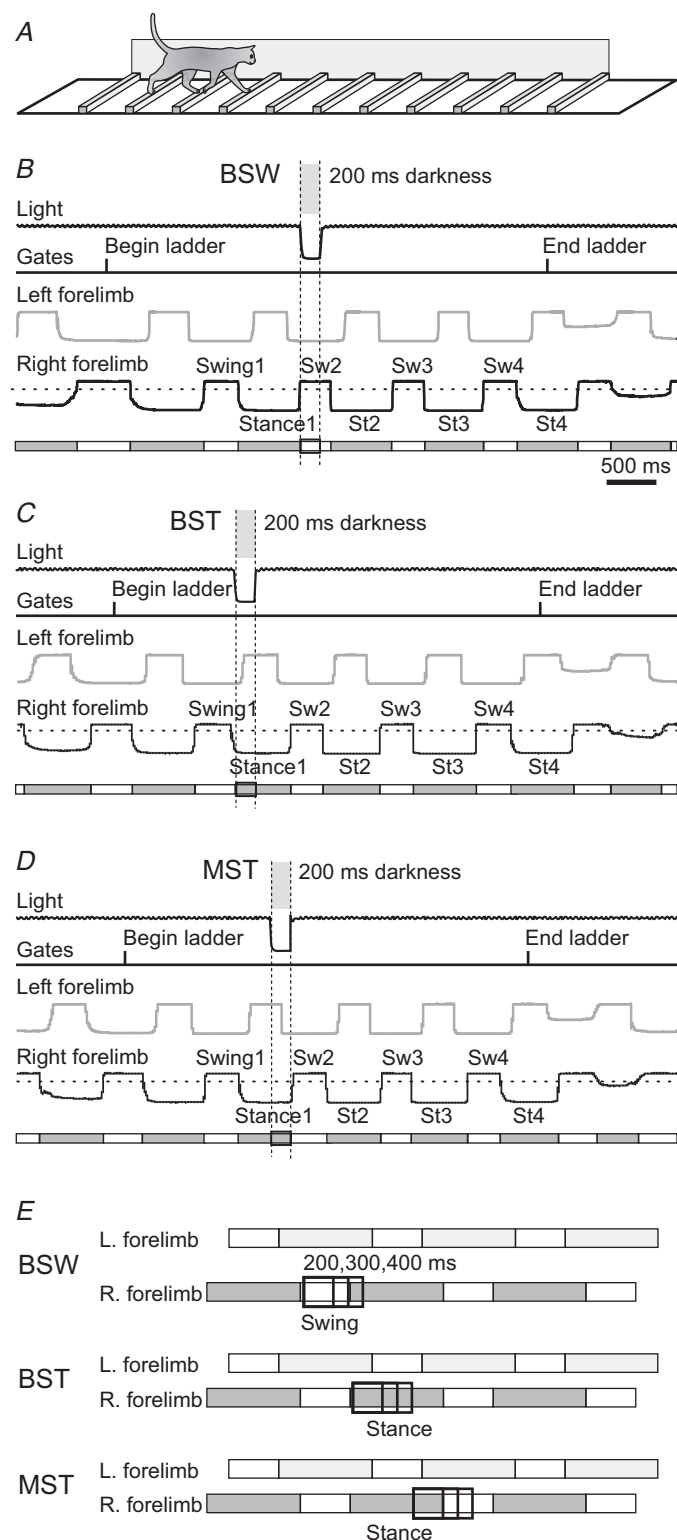


Figure 1. Experimental protocol

A, a cat walking on a horizontal ladder. *B–D*, examples of recording and measurement of the duration of strides in three passages along the ladder. *B–D*, visual input was interrupted by a short (200 ms) period of darkness starting at different phases of the stride: at the beginning of a swing (*B*; BSW); at the beginning of a stance (*C*; BST); and in the middle of a stance (*D*; MST). In *B–D*, from top to bottom: light in the room, with the period of darkness (downward deflection) highlighted by a grey vertical bar and vertical dashed lines; gates trace shows the time when the cat entered (begin ladder) and left (end ladder) the ladder swing corridor; left forelimb stances (deflections down) and swings (deflections up); right forelimb stances (St) and swings (Sw), numbered, with a measuring threshold indicated by a horizontal dashed line; and the sequence of stances (grey bars) and swings (open bars) for the right forelimb. The swings and stances of the right forelimb were used for timing reference throughout the paper. Recordings in *B–D* are from one experiment, cat FM. *E*, schematic presentation of all experimental conditions. Black frames of different length show schematically the duration of darkness in different experiments (200, 300 and 400 ms). See main text for details.

includes a swing as a 'step'. Signals from the right forepaw were used as a reference for all timings in this study.

Photo sensors paired with infrared light-emitting diodes detected when the cat started and ended a passage along a corridor ('gates' signal in Fig. 1B–D, see also Fig. 2A in the Results below). The emission wavelength of the diodes was 850–900 nm, which is outside the visible spectral range of the cat (Guenther & Zrenner, 1993).

Brief interruption of visual input at different phases of the stride cycle

The experimental room was completely isolated from external light sources. Internal light sources from equipment were covered. Computer screens were located in a separate room. The only light source in the experimental room was a computer-controlled set of overhead fluorescent bulbs providing standard laboratory illumination of ~500 lux. Interruption of visual input was achieved by turning these lights off. When the lights were turned off, within 17 ms the level of illumination in the room fell to <0.01 lux, as measured by a T-10 illuminance meter (Konica Minolta, Ramsey, NJ, USA). The timing of the periods of darkness was recorded using a photocell ('light' trace in Fig. 1B–D). Note that although fully dark-adapted cats need only $\sim 3 \times 10^{-7}$ lux to see (Gunter, 1951), full dark adaptation takes ≥ 30 min, and first behavioural signs of dark adaptation in cats appear after ≥ 5 min in the dark (Lamotte & Brown, 1970; Lankheet *et al.* 1996). The periods of darkness used in our experiments (200–400 ms) are too short to produce any significant increase of sensitivity to compensate for a >10,000-fold decrease of the illumination level. Thus, for a walking cat, the room could be considered completely dark, and visual input interrupted, for the duration of the light-off period.

To study the effects of brief interruption of visual input on strides on the ladder, the lights in the experimental room were turned off for 200, 300 or 400 ms starting at different phases of the step cycle: at the beginning of a swing (BSW), at the beginning of a stance (BST) or in the middle of a stance (MST). The BSW darkness was triggered by the swing-on signal, and the lights went off within <20 ms after the swing onset (Fig. 1B). With average swing duration in cats between 200 and 400 ms in our experimental conditions (e.g. Beloozerova *et al.* 2010; see also Table 1), the BSW darkness period would either be contained within the swing or could cover all of the swing and extend into the following stance, depending on the duration of the darkness (200, 300 or 400 ms) and the strides of the cat (Fig. 1E). Note that the schemes in Fig. 1E and descriptions in this paragraph relate the periods of darkness to unperturbed control strides, without taking into consideration a possible change of the duration

Table 1. Parameters of unperturbed (control) strides

Parameters of strides	Cat FM	Cat TI	Cat SF
Duration of swing (ms)	328 ± 41	280 ± 27	423 ± 53
Duration of stance (ms)	488 ± 67	341 ± 51	441 ± 73
Duration of step cycle (ms)	816 ± 91	621 ± 70	868 ± 109
Duty factor (%)**	59.7 ± 4.1	54.8 ± 2.9	50.5 ± 4.4
Number of strides	1101	553	625

Values are gross averages across all recording sessions, expressed as the mean ± SD.

**Duty factor shows relative duration of the stance phase within the step cycle.

of swings or stances by the periods of darkness. The BST darkness was triggered by the stance-on signal, and the lights went off within <20 ms after the stance onset (Fig. 1C). The average stance duration in our experimental conditions was 350–500 ms (e.g. Beloozerova *et al.* 2010), meaning that the period of darkness was typically contained within a stance, covering its first part or most of the stance (Fig. 1E). The MST darkness was also triggered by the stance-on signal, but the lights went off with a delay, starting at about the middle of the stance (Fig. 1D). With short (200 ms) MST darkness, the lights were back on at the end of stance; longer darkness periods (300 and 400 ms) could extend into the next swing (Fig. 1E).

During one recording session, all periods of darkness had the same duration (200, 300 or 400 ms), but started at different phases of the step cycle. Periods of darkness occurred on 70–90% of passages through the recording chamber; during a passage, only one darkness period could occur while the cat was in any part of the chamber: on the ladder, in a turn or in the return corridor. On 30–70% of the passages, the light-off period occurred when the cat was on the ladder. To minimize effects of learning within a session, we: (i) interleaved, in a pseudo-random order, passages in which the light was turned off with unperturbed passages; (ii) randomized the stride during which the light was turned off such that it could be in any part of the chamber; and (iii) varied the timing of the light-off period within the step cycle. For the analysis, we used passages in which the BSW, BST or MST darkness, as illustrated in Fig. 1, started when the cat was completely on the ladder and compared them with unperturbed passages; the remaining passages were excluded from analyses, although they were necessary to minimize possible confounds attributable to learning.

Data processing and statistical analysis

The duration of swings and stances was measured using signals from the paw sensors (Fig. 1). The beginning of

a passage along the ladder was defined as time of the 'begin ladder' signal from a photo sensor paired with infrared light-emitting diode at the beginning of the ladder corridor (Fig 1 see also Fig 2A below). The end of a passage along the ladder was defined as time of the 'end ladder' signal from a photo sensor paired with a similar diode at the end of the ladder (Fig 1 see also Fig 2A below). Signals from the right forepaw were used as the reference for all timings in the study.

The following criteria were used for including data from a passage in the analysis. (i) Good signal recorded from the right paw sensor, with clearly detected swings and stances. (ii) Smooth walking along the ladder. Although measures were taken to minimize possible distractions, cats could occasionally be distracted and the regularity of the walking pattern disrupted. A passage was considered disrupted and excluded from analysis if the duration of any swing or stance was more than two times longer than the average for the recording session, or in which cats stopped on the ladder (stance > 2 s long) for reasons not related to the light-off period. (iii) For unperturbed control passages inclusion criterion was also no lights off while the cat was on the ladder. (iv) For perturbed passages inclusion criterion was that lights-off period was while the cat was completely on the ladder, and not turning to or from the ladder. (v) Absence of significant trends in walking speed. Food-motivated cats typically start off a session with fast running, then slow down somewhat and continue for a good while at a relatively steady pace, and then slow down further as they become full toward the end of the session. Recording periods during which durations of unperturbed strides plotted against time showed little trend were selected for the analysis (see Fig. 2B).

From the passages that fulfilled the above criteria, we used data from four strides made while the cat was on the ladder: durations of four swings and four stances (see Fig. 2A below). To reveal whether a period of darkness affected the strides, we first compared the duration of each swing and stance during the passages with one condition of the perturbation (light-off period with the same onset timing, BSW, BST or MST, and same duration, occurring during the same stride on the ladder: first, second, third or fourth) with the respective swings and stances during unperturbed control passages (see Fig. 3C below). To combine data for the same light-off onset timing and duration (e.g. BSW 300 ms in Fig. 3D below) but for different strides along the ladder and different recording sessions, we calculated: (i) a change of each swing or stance as a percentage, for which we normalized the duration of each swing or stance by the respective mean duration during unperturbed passages from that recording session; and (ii) a change of each swing or stance in milliseconds, as a difference between the duration of each swing and stance and the respective

mean duration during unperturbed passages from that recording session. Next, the dark-perturbed passages were aligned by the swing or stance, during which the lights were turned off (see Fig. 3D below). Average changes as percentages and in milliseconds were calculated for the aligned swings and stances and compared with control conditions.

For calculation of significance, Student's *t*-test (two-tailed, unequal variance) and Fisher's *F*-test (two-tailed) were used. Differences were considered significant at $P < 0.05$. Throughout the text, *P*-values are given in full for $P > 0.001$ and as $P < 0.001$ for $P < 0.001$; mean values are given with the SD.

Results

We studied the dynamics of demands on vision during locomotion on the ladder, using brief periods of darkness (light off for 200, 300 or 400 ms) that denied visual input during different phases of the step cycle. These short periods of denial of visual input never caused cats to miss a crosspiece on the ladder. However, depending on the duration and the timing of the darkness within the step cycle, they could prolong certain phases of the stride.

Database

Data were obtained from three cats in 17 recording sessions: nine with cat FM, four with cat SF and four with cat TI (Table 2). With cats SF and TI, recording sessions were made over compact periods of 1 month. With cat FM, two recording sessions on two consecutive days were conducted before the cat was implanted with brain electrodes for a different project, and the remaining seven sessions were conducted over a period of 9 days, 1 year later, > 6 months after recovery from the implantation surgery. One recording session was conducted per day. During a recording session, a cat walked in the experimental chamber for between 35 min and 2.5 h, with most sessions lasting 1–1.5 h. In each session, cats completed between 80 and 218 passages through the chamber. During 30–70% of passages (randomized), the lights were turned off for 200, 300 or 400 ms at various phases of the step cycle while the cat was at different locations along the ladder. Figure 1 illustrates an experiment with the lights off for 200 ms. For the analysis, we used passages during which the lights were turned off while the cat was walking on the ladder (19–64 per session; 'perturbed' passages) and compared them with passages that were not perturbed by the darkness. Table 2 shows the number of recording sessions and passages included in the analysis for each condition.

Strides during unperturbed walking on the ladder

Figure 2A shows a typical passage of a cat along the ladder (cat TI). The cat started the passage stepping with its right forepaw on the first crosspiece, and made four strides (four swings and stances) along the ladder before exiting it. The cat walked with a fairly regular pace during the first 37 min of this recording session, making 113 passages. Thirty-four of these passages were completed under uninterrupted illumination and were used for the quantification of unperturbed strides for this session. Figure 2B shows durations of swings (magenta) and stances (blue) of uninterrupted strides. The durations of each swing and stance from one passage are represented by symbols of the respective colour, vertically stacked at

the time of the beginning of the passage. As the session progressed, the cat walked slightly slower, as evidenced by a tendency of the duration of swings and stances to increase. The average duration of swings and stances from unperturbed passages of this session are shown in Fig. 2C as green circles with error bars (SD), connected by a green dashed line. The durations of the swings were shorter than the durations of stances, meaning that there was an overlap between the stances of the right and left forepaws, and both forepaws were on the crosspieces of the ladder before the next swing. This is typical for cat locomotion at the speeds observed in this study.

The average durations of swings and stances during unperturbed passages from other recording sessions with cat TI are shown in Fig. 2C with symbols of different

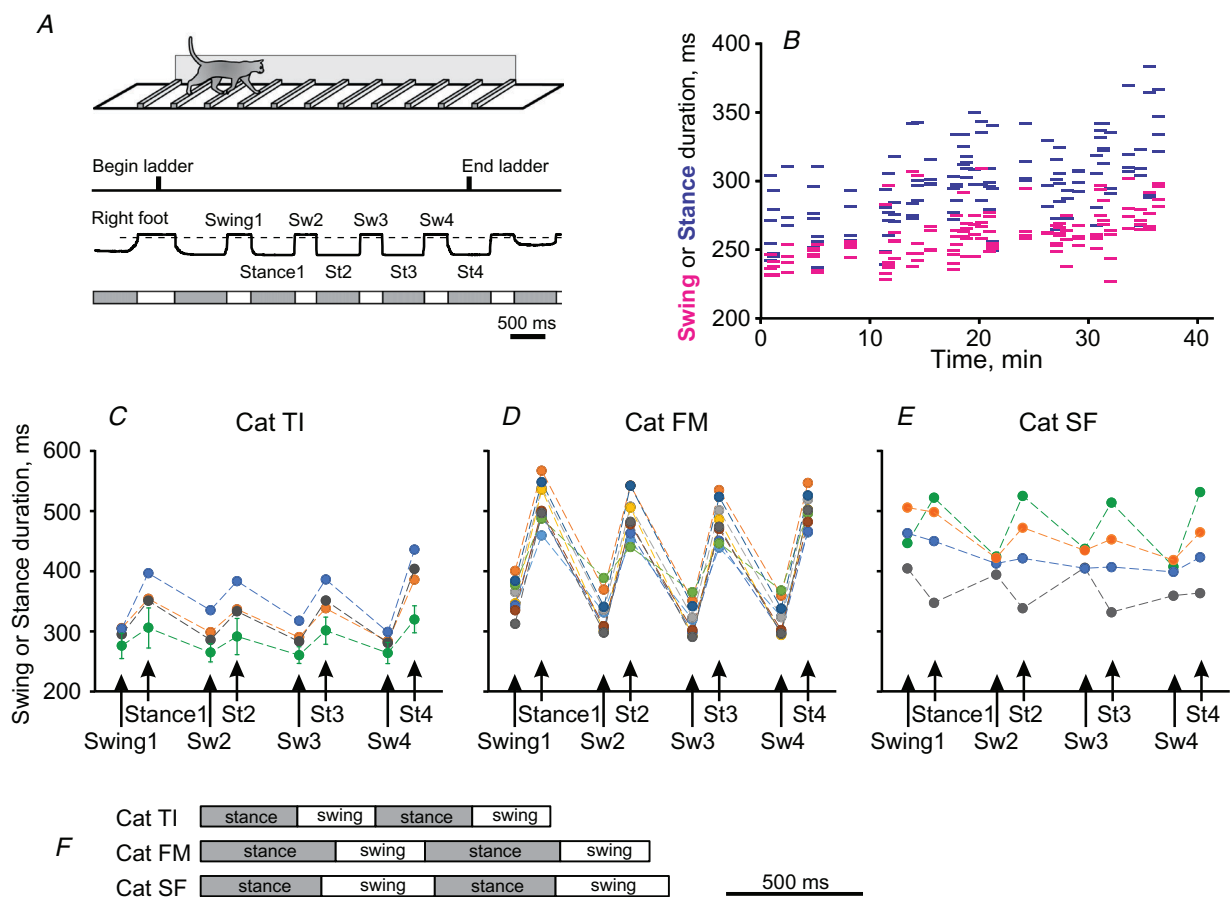


Figure 2. Unperturbed (control) strides in three cats

A, a cat walking on the ladder (top) and example recording and measurement of the right foot strides (cat TI). Same conventions as in Fig. 1. B, durations of swings (magenta) and stances (blue) during 34 unperturbed passages of one recording session (cat TI). For each passage, the duration of four swings and four stances of the right foot are plotted, aligned on the start time of the passage, determined by the 'begin ladder' sensor. C–E, duration of swings and stances during different recording sessions with three cats. In each plot, symbols of the same colour connected with a dashed line show the durations of swings and stances during unperturbed passages, averaged for each recording session (20–61 passages per session). In C, green symbols with error bars (SD) show averaged data from B. For other sessions in C–E, error bars are omitted for clarity. Data are from cat TI (four sessions; C), cat FM (nine sessions; D) and cat SF (four sessions; E). Note the different durations of strides in the three cats and uniform patterns of strides in cats TI and FM, but highly variable patterns of strides in cat SF. F, duration of unperturbed stances and swings of three cats, gross averaged over all recording sessions and strides.

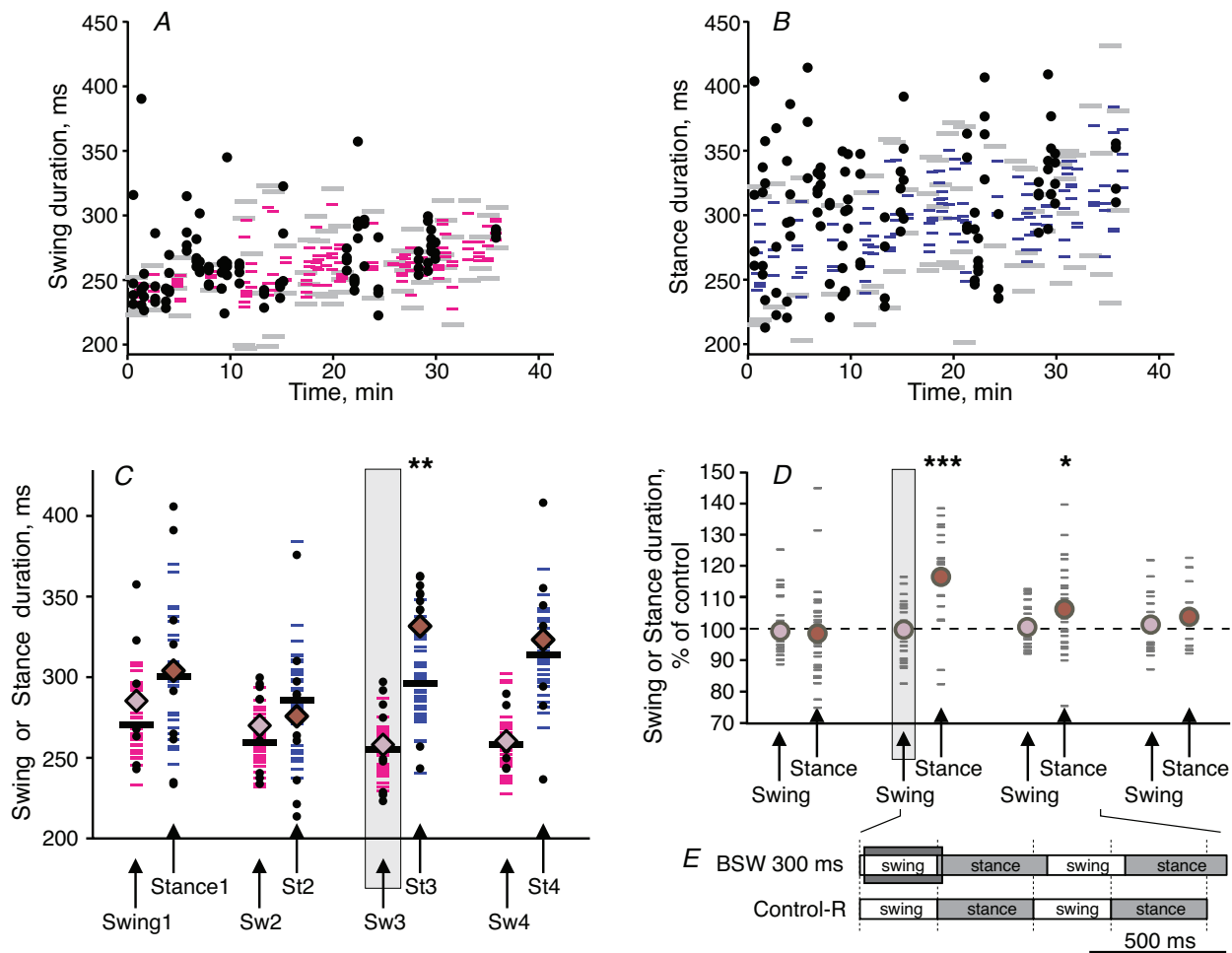


Figure 3. Brief interruption of visual input can affect the duration of strides on the ladder

A and B, duration of swings (A) and stances (B) during passages perturbed by a 300 ms period of darkness (black circles) and unperturbed (coloured dashes). Light grey dashes show the mean + 2SD and mean – 2SD for each passage. Darkness started either at the beginning of a swing (BSW; 19 passages) or the beginning of a stance (BST; eight passages). Dark-perturbed passages were interleaved with unperturbed control runs. Data from each passage are aligned on the start time of the passage. Data for cat T1, from the same recording session as in Fig. 2B. Only passages included in the final analyses are shown. C, duration of swings (Sw) and stances (St) during passages perturbed by 300 ms of darkness starting at the beginning of swing 3 ($n = 10$, black circles; large diamond symbols show averages, with lighter fill for swings and darker fill for stances) and during unperturbed passages ($n = 34$, dashes) for comparison; data are from A and B. Grey vertical bar highlights swing 3, during which the light was turned off. The stance 3 after swing 3 in the dark was longer than in control conditions (** $P = 0.00111$). D, summary data from all experiments with 300 ms BSW darkness (31 passages). The duration of each swing and stance was normalized to the average duration of the respective swing or stance during unperturbed passages of the same recording session, and data from each passage were aligned by the dark-perturbed swing (grey vertical bar). Dash symbols show individual data points; circles show averages. The stance after the swing in the darkness was longer than in control conditions (** $P < 0.001$), as was the stance of the following stride (* $P = 0.0334$). Note that the y-scale is in percentages of the control value. E, changes of the duration of swings and stances after 300 ms BSW light-off period in milliseconds, calculated as follows. For each swing and stance, the difference in milliseconds from the respective average from unperturbed passages during the same recording session was calculated; data from each passage were aligned by the light-off swing; then the average change of each swing or stance duration was calculated across all recording sessions. For the top plot (BSW 300 ms), the averaged change was added to the gross average of the swing or stance duration for this cat. The black horizontal bar behind the light-off swing indicates the onset and duration of the darkness; note that in this experiment, darkness extended slightly into the following stance. The gross averages of control swings and stances (Control-R) are shown below for comparison. Data in A–E are from cat T1.

Table 2. Database: number of recording sessions and passages for each condition included in the final analysis

Cat	Duration of darkness (ms)	Recording sessions	Unperturbed passages	BSW passages	BST passages	MST passages
FM	200	4	146	42	90	43
	300	3	82	48	36	41
	400	2	60	13	15	13
TI	300	2	77	31	8	4
	400	2	62	21	—	—
SF	300	2	83	21	19	—
	400	2	89	31	11	10

Abbreviations: BST, beginning of a stance; BSW, beginning of a swing; MST, middle of a stance.

colours connected with dashed lines. The cat ran faster during some sessions (e.g. green) and slower during others (e.g. blue); however, the durations of swings and stances were consistent during the four strides within each recording session, and the overall pattern of strides was consistent across sessions. Cat FM also had a highly consistent pattern of walking on the ladder: the pattern of strides was highly consistent across sessions, and the duration of swings and stances was consistent across strides of the same session (Fig. 2D). In contrast, cat SF had a more variable pattern of walking (Fig. 2E). This was reflected in a higher SD of the normalized duration of stances in cat SF compared with cats TI and FM (16.7% compared with 14.9%, $P = 0.00671$; and 13.9%, $P < 0.001$; F -test). The SD of the normalized duration of swings in cat SF was higher than that in cat TI (12.6% vs. 9.8%, $P < 0.001$), but not different from cat FM (12.6% vs. 12.4%, $P = 0.559$). Table 1 shows basic characteristics of unperturbed strides in three cats. Gross averages of the durations of swings and stances (across recording sessions and four strides) were 280 ± 27 and 341 ± 51 ms for cat TI, 328 ± 41 and 488 ± 67 ms for cat FM, and 423 ± 53 and 441 ± 73 ms for cat SF, respectively.

The average pattern of strides for each cat is illustrated graphically in Fig. 2F. Given that all strides had the same length, determined by the distance between the cross-pieces of the ladder, a different duration of strides means a different speed of walking.

Brief interruption of visual input can affect strides on the ladder

Figure 3 shows an example experiment, in which strides on the ladder were affected by a brief period of darkness. It also illustrates procedures used for data processing. In this session with cat TI, light was turned off for 300 ms starting at the beginning of a swing on 19 passages (on nine during swing 2 and on ten during swing 3) or at the beginning of a stance on eight passages (on six during stance 2 and on two during stance 3). The duration of swings and stances

during these 27 passages with light-off periods are shown in Fig. 3A and B (black circles), together with data from unperturbed passages shown with coloured dash symbols as in Fig 2B, for comparison. Passages with 300 ms periods of darkness were interleaved with unperturbed passages. Note that Fig. 3A and B illustrates only passages included in the final analyses according to criteria outlined in the Methods.

Figure 3A and B shows that during light-off passages some swings and stances were longer than during unperturbed passages. Respective data points are located above the large dash symbols, which show the two SD confidence interval from the mean of unperturbed strides (Fig. 3A and B). To reveal which phases of the strides were affected by light-off periods, we conducted the following analyses. For each specific condition, e.g. light off for 300 ms starting at the beginning of swing 3, we compared data from perturbed and unperturbed control passages for each swing and stance. Figure 3C shows that the duration of swing 3, during which the lights were off, was not different from control (257 ± 28 vs. 254 ± 14 ms in control conditions, $P = 0.611$). Notably, 300 ms of darkness covered the end of the swing; hence, the foot landed on the crosspiece in the dark. The duration of the following stance 3, however, increased by 36 ms (or 12%), from 295 ± 23 ms in control conditions to 331 ± 44 ms during perturbed passages ($P = 0.00111$, Student's t -test). Neither the swing nor the stance of the next stride was affected.

Figure 3D summarizes data from all experiments with 300 ms darkness starting in the beginning of swing 2 or swing 3. For this plot, we normalized the duration of swings and stances during dark-perturbed passages by the average duration of a respective swing or stance during unperturbed control passages from the same recording session. Then, we aligned normalized data from perturbed passages by the swing during which the darkness started (grey vertical bar in Fig. 3D). This analysis confirmed that the duration of the swing during which lights were off was same as during unperturbed strides, but the following

stance was prolonged to $117 \pm 13\%$ of control ($n = 30$, $P < 0.001$). During the next stride, the duration of the swing was also normal, but the stance was prolonged to $106 \pm 13\%$ of control ($P = 0.0334$).

To illustrate how the temporal sequence of strides was affected by a 300 ms BSW darkness (Fig. 3E), we performed a similar analysis as in Fig. 3D, but calculated changes in milliseconds instead of as percentages. The calculated average change of the duration of each swing and stance starting from the 'light-off' swing was then added to the sequence of gross-averaged swings and stances. Figure 3E shows results of this analysis (Fig. 3E, BSW 300 ms), with the black bar behind the first swing showing the period of darkness. The sequence of swings and stances in control conditions (Control-R and vertical dashed lines) is shown for reference. In addition to a demonstration that the duration of the swing in the dark did not change and duration of the following stance was greater by 53 ± 40 ms ($P < 0.001$, $n = 30$), Fig. 3E illustrates the total increase of the duration of strides after the perturbation. In this example, the total prolongation was 72 ± 100 ms ($P = 0.00110$) after a 300 ms period of darkness (difference of swings and stances from control, starting with the 'light-off' swing was: -2 ± 25 , $+53 \pm 40$, $+1 \pm 15$ and $+20 \pm 41$ ms). Note that the overall prolongation of the two strides was significantly shorter than the light-off duration. This was observed consistently for any light-off perturbations studied here (results will be presented in Figs. 6 and 7 below). The duration of strides on the ladder was affected by all periods of darkness 300 and 400 ms long and by some that were 200 ms long. The effect depended on both the phase of the stride when the darkness started and the duration of the darkness. These two factors will be considered below.

Effect of visual input interruption on strides depends on timing of the darkness in the step cycle

To reveal how the effect of a brief interruption of visual input on strides depends on the phase of the light-off period within the step cycle, we tested darkness periods starting at the beginning of the swing (BSW, described above), beginning of the stance (BST) or middle of the stance (MST).

Figure 4 shows the effects of 400 ms of darkness starting in different phases of the step cycle in cat FM. A light-off period of 400 ms BSW did not affect the duration of this swing, which was completed in the darkness (Fig. 4A and E; duration $106.7 \pm 32\%$ of control, $P = 0.597$). The following stance was prolonged to $125 \pm 13\%$ of control ($P < 0.001$). The next strides were not affected (Fig. 4A). This effect was qualitatively similar to the effect of 300 ms of darkness BSW described above for cat TI. A period of 400 ms of darkness BST had a more long-lasting effect on strides (Fig. 4B). The stance was prolonged to $130 \pm 12\%$

of control ($P < 0.001$). Notably, the darkness was shorter than the average stance duration during unperturbed passages in these recording sessions (488 ± 67 ms; Fig. 4E). Thus, lights were back on 60–70 ms before the 'normal' beginning of the next swing, but the swing was delayed by ~ 150 ms, or 210–220 ms after the lights were back on (Fig. 4E). Also, the swing and stance of the next stride were longer than in control conditions (Fig. 4B; swing $115 \pm 10\%$ of control, $P < 0.001$; stance $111 \pm 8\%$ of control, $P < 0.001$), and even those of the stride after the next were longer (Fig. 4B; swing $108 \pm 8\%$ of control, $P = 0.00657$; stance $108 \pm 5\%$ of control, $P = 0.0101$). A 400 ms period of darkness MST did not affect the stance duration, meaning that the next swing started in the darkness at the same time as it would during unperturbed passages (Fig. 4C and E; $100 \pm 11\%$ of control, $P = 0.765$). However, this swing and the following stance were both longer than normal (Fig. 4C; swing $118 \pm 21\%$ of control, $P = 0.0112$; stance $121 \pm 11\%$ of control, $P < 0.001$). In the next stride, the swing was not affected ($102 \pm 8\%$ of control, $P = 0.433$), but the duration of stance increased to $107 \pm 8\%$ of control ($P = 0.0464$).

Notably, the effects of 400 ms BSW, BST and MST darkness on swings and stances that were in progress when the lights were turned off or that started in the dark were remarkably diverse. When the lights went off < 20 ms after the start of a swing and covered the rest of it (Fig. 4, BSW), the swing was conducted normally, and the paw was placed on the ladder in the darkness at the same time as it would be in control conditions, with the lights on throughout. In contrast, in MST conditions, when the lights were off ~ 200 –250 ms before the start of the swing and were turned on again in about the middle of the swing (Fig. 4, MST), the swing started as in control conditions, but was prolonged. Thus, the duration of a swing was increased when visual input was denied during the second half of the preceding stance and at the onset of the swing. Stance duration was not affected by the darkness covering its second half (Fig. 4, MST), but the stance was prolonged if the darkness covered its very beginning (Fig. 4; BSW) or started shortly (< 20 ms) after the beginning of the stance (Fig. 4, BST). Together, these results illustrate that the demands on visual input for planning the duration of ongoing swings and stances change during the stride cycle. They also show that a swing after it has started, or a stance after it has started and is half-through, will be executed to the end and 'on time' (as in control conditions) even in the absence of visual input.

Superimposed averages from Fig. 4A–C demonstrate the difference between the effects of a 400 ms period of darkness starting at distinct phases of the step cycle on the duration of swings and stances (Fig. 4D). The strongest perturbation of strides was observed after a 400 ms BST period of darkness: it prolonged the light-off stance, the swing and stance of the next stride, and even the swing and

stance of the stride after the next. As a result, the overall prolongation of strides was largest after the BST darkness (Fig. 4E; BST 400 ms, 233 ± 96 ms longer than control, $P < 0.001$). In contrast, neither MST nor BSW darkness had an effect on the stance or swing during which the lights were turned off. A 400 ms MST period of darkness prolonged both the swing and stance of the next stride. A 400 ms BSW period of darkness prolonged only the next stance. The strides after the MST darkness were 158 ± 95 ms longer than control ($P < 0.001$; Fig. 4E, MST 400 ms) and after the BSW darkness 124 ± 210 ms longer than control ($P = 0.0604$; Fig. 4E, BSW 400 ms).

In other cats (TI and SF), effects of long darkness (300 or 400 ms) were qualitatively similar to those described above for cat FM. Strides on the ladder were always affected, and the effects depended on the phase of the

stride during which the lights were turned off (see Fig. 6 below).

Thus, an interruption of visual input for 400 ms in any tested stride phase (BSW, BST or MST) prolonged strides; however, the strength of the perturbation and details of which swings and/or stances were affected depended on the phase of the step cycle in which the lights were turned off. This indicates that the demand on visual information and the ability of visuomotor systems to compensate for its interruption change within the step cycle.

Duration of visual input interruption needed to affect strides

Next, we asked how shorter periods of darkness would affect strides on the ladder, and for how long the visual

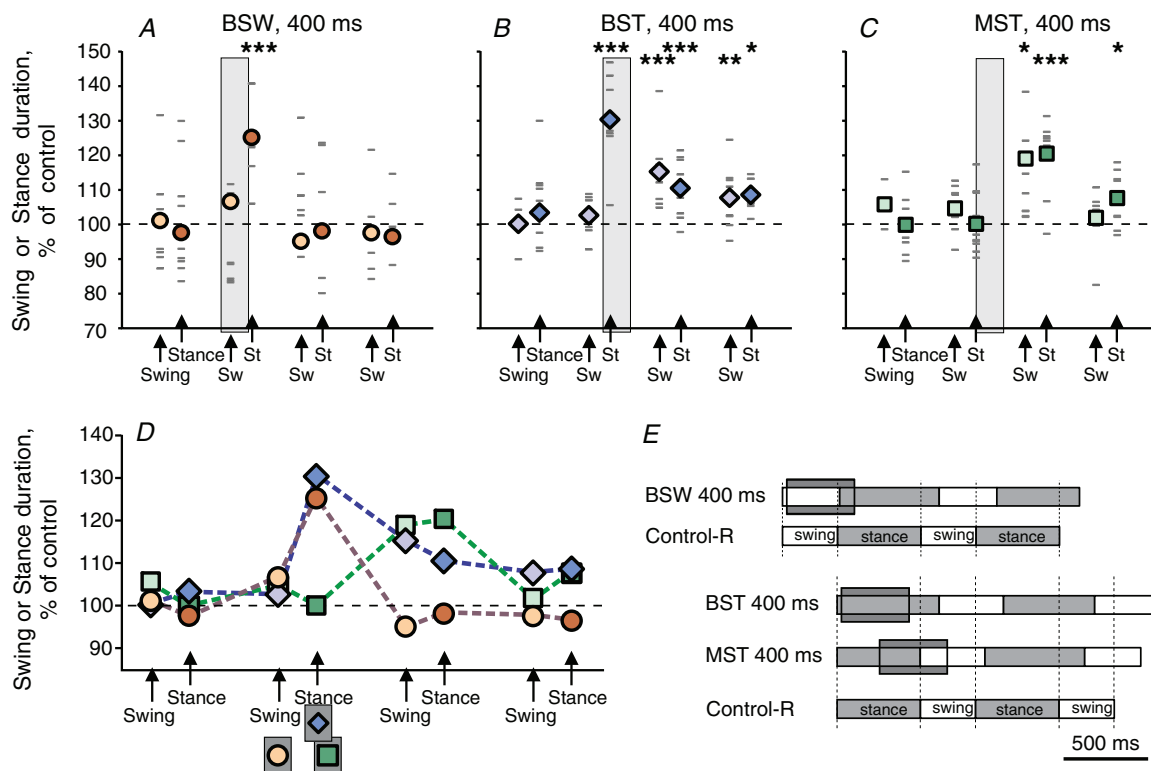


Figure 4. Effect of interruption of visual input depends on timing of the darkness within the step cycle

A–C, changes of the duration of swings and stances after 400 ms of darkness that started at the beginning of a swing (A; BSW; 11 passages), at the beginning of a stance (B; BST; 13 passages) or in the middle of a stance (C; MST; 13 passages). Swing and stance durations were normalized to respective controls, and data from each passage were aligned by the beginning of the light-off swing or stance, as in Fig. 3D (grey vertical bar). Dashes show data for individual swings and stances; large symbols show averages for swings (lighter fill) and stances (darker fill). Significant difference from control is indicated as follows: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$. D, average changes of the duration of swings and stances after 400 ms of BSW (circles), BST (diamonds) and MST (squares) periods of darkness; data from A–C, superimposed for comparison; averages for each condition expressed as a percentage of the control value are connected with a dashed line for clarity. Symbols in grey rectangles below the plot indicate the swing or stance during which the light was turned off in the respective experiments. E, changes of the strides after 400 ms of BSW, BST or MST periods of darkness. Black horizontal bars behind the light-off stance or swing indicate the onset and duration of the darkness. The duration of unperturbed swings and stances (Control-R) is shown for comparison. Data in A–E are from cat FM.

input had to be denied to affect strides. Figure 5 shows results of experiments with 200, 300 and 400 ms of darkness starting in the middle of stance (MST) for cat FM. A 200 ms MST period of darkness had no effect on strides on the ladder (Fig. 5A). This period of darkness was contained within stance, covering its second half. Longer periods of darkness, 300 and 400 ms, extended over the end of the stance into the following swing (Fig. 5E). However, neither of these conditions affected the duration of the light-off stance; hence, with the 300 and 400 ms MST darkness, the swing started in the dark. The duration of this swing increased to $111 \pm 8\%$ of control after the 300 ms MST period of darkness (Fig. 5B; $n = 41$, $P < 0.001$) and to $118 \pm 21\%$ of control after the 400 ms MST (Fig. 5C; $n = 13$, $P = 0.0112$). The 400 ms period of darkness also prolonged the stance of the next stride (Fig. 5C; $121 \pm 11\%$ of control, $P < 0.001$) and

the stance of the following stride ($107 \pm 8\%$ of control, $P = 0.0464$), as described above (Fig. 5C; same data as in Fig. 4C).

The emergence and gradual increase of the effect on strides with increasing duration of MST darkness from 200 to 300 and to 400 ms is summarized in Fig. 5D and E. The effects first appeared with the 300 ms of darkness, and with 400 ms the magnitude of the effects on swings and stances increased, in addition to the number of prolonged swings and stances (Fig. 5D). In comparison to control conditions, the duration of stances and swings changed, starting from the 'light-off' stance, by -5 ± 55 , $+7 \pm 39$, $+2 \pm 50$ and -7 ± 27 ms after 200 ms of darkness; by -6 ± 47 , $+36 \pm 27$, $+16 \pm 42$ and $+5 \pm 22$ ms after 300 ms of darkness; and by $+3 \pm 47$, $+53 \pm 63$, $+97 \pm 48$ and $+6 \pm 22$ ms after 400 ms of darkness (significance as in Fig. 5A–C). Respectively, the overall prolongation of

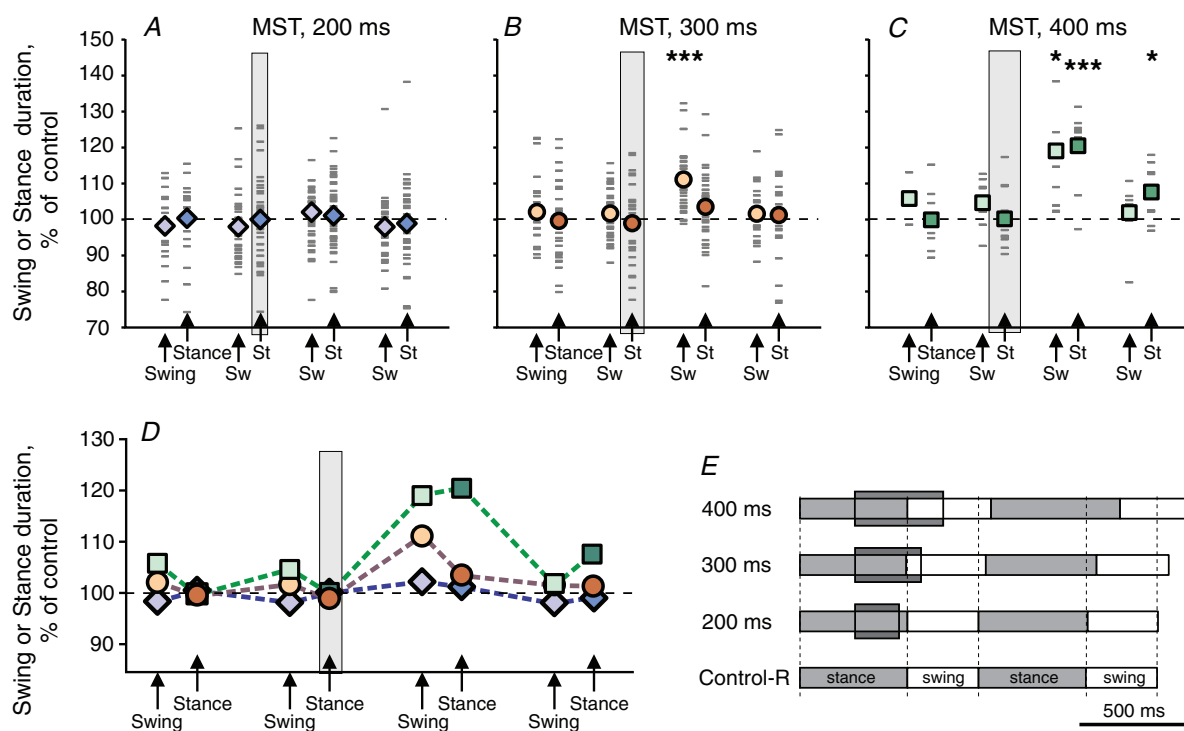


Figure 5. Emergence and increase of the effect of interruption of visual input on strides with increasing duration of the darkness

A–C, changes in the duration of swings and stances after darkness that started in the middle of a stance (MST) and lasted for 200 ms (A; 43 passages), 300 ms (B; 41 passages) or 400 ms (C; 11 passages). Swing and stance durations were normalized to the respective controls, and data from each passage were aligned by the beginning of the light-off stance (grey vertical bar). Dashes show data for individual swings and stances; large symbols show averages for swings (lighter fill) and stances (darker fill). Significant difference from control is indicated as follows: * $P < 0.05$; *** $P < 0.001$. D, average changes of the duration of swings and stances after MST periods of darkness lasting 200 ms (diamonds), 300 ms (circles) and 400 ms (squares); data from A–C, superimposed for comparison; averages for each condition are connected with a dashed line for clarity. E, changes of the strides after MST periods of darkness lasting 200, 300 or 400 ms. Changes for each swing or stance were calculated in milliseconds, relative to the gross average for the cat, as in Fig. 3E. Black horizontal bars behind the light-off stance indicate the onset and duration of the darkness. Note that the 200 ms period of darkness was completely within the stance, whereas the 300 and 400 ms dark periods extended into the following swing. Bottom plot (Control-R) shows duration of unperturbed swings and stances for comparison. Data in A–E are from cat FM.

strides was small after 300 ms MST darkness (49 ± 102 ms increase relative to control conditions, $P = 0.0176$) and larger after 400 ms of darkness (158 ± 95 ms increase relative to control conditions, $P < 0.001$ vs. control, $P = 0.0018$ vs. 300 ms effect; Fig. 5E).

The effects of darkness starting in other phases of the step cycle (BSW and BST) also depended on the duration of the darkness, with longer darkness affecting strides more (Fig. 6). A 200 ms period of darkness slightly prolonged the stance if started at its beginning (BST, by $4.9 \pm 12\%$ or 23 ± 60 ms, $n = 90$, $P < 0.001$) or in the beginning of the preceding swing (BSW, by $5.2 \pm 10\%$ or 26 ± 52 ms, $n = 42$, $P = 0.00294$). A 300 ms BST period of darkness prolonged both the lights-off stance and the following swing (stance by $10.6 \pm 11\%$ or 53 ± 56 ms, $n = 36$, $P < 0.001$; swing by $6.3 \pm 10\%$, or 21 ± 35 ms, $P = 0.00185$), with the total increase of the duration of that stride being 74 ± 83 ms ($P < 0.001$). The effect of a 400 ms BST period of darkness was even stronger, increasing the duration of the light-off stance by 109 ± 73 ms, the following swing by 55 ± 37 ms (total increase of the stride by 164 ± 63 ms, $P < 0.001$), and even affecting the next strides. A 300 ms BSW period of darkness led to a combined increase of the light-off swing and the following stance duration by 60 ± 75 ms ($P < 0.001$) and 400 ms BSW by 109 ± 134 ms ($P = 0.0133$). Given that the effects of 200 ms of darkness were small, it was tested extensively only in cat FM. In cat TI, a 300 ms period of darkness

starting at BST had no effect on strides, but it prolonged stances when started in other phases of the step cycle (Fig. 6). A 400 ms BSW period of darkness had stronger effect than 300 ms BSW on the duration of the following stance ($129.4 \pm 21\%$ of control or 104 ± 79 ms longer than control, $n = 21$, $P < 0.001$; vs. $117.1 \pm 15.5\%$ of control or 53 ± 40 ms longer than control, $n = 31$, $P < 0.001$). In cat SF, strides on the ladder were prolonged by 300 and 400 ms of darkness starting in any of the tested stride phases (BSW, BST or MST; Fig. 6).

Interruption of visual input affects stances more strongly than swings

Figure 7 summarizes the observed effects of brief periods of denial of visual input on swings and stances during locomotion on the ladder. Brief periods of darkness (200 or 300 ms) affected only the current and the following swing and/or stance. A 400 ms period of darkness could also prolong later strides (e.g. 400 ms BST or MST; Fig. 4); however, this effect could also be attributable to compensatory processes adjusting the gait after a major disturbance (by ~ 150 ms or more during the first affected stride), rather than reflecting a direct effect of lights-off 1.5 s ago.

Brief periods of darkness had stronger effects on stances than on swings. Both the duration of stances increased more than the duration of swings, and the length of the

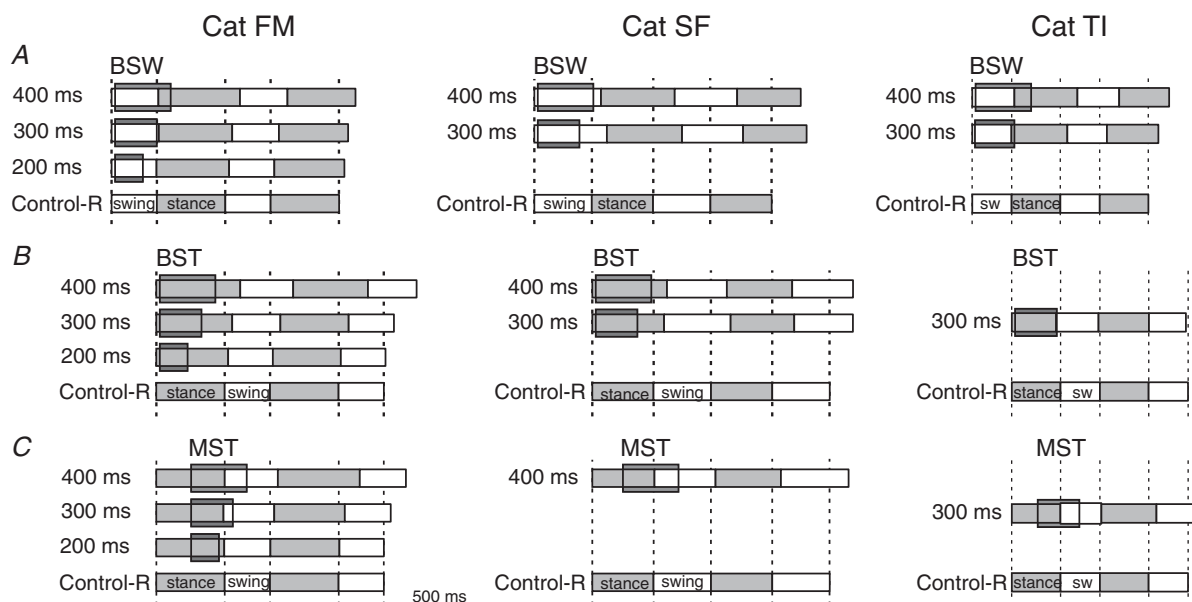


Figure 6. Summary of the effects of a brief interruption of visual input on strides in three cats

Changes of strides after interruption of visual input by brief periods of darkness that started at the beginning of a swing (A; BSW), at the beginning of a stance (B; BST) or in the middle of a stance (C; MST). The timing and duration of the darkness are indicated by black boxes; the duration of darkness (in milliseconds) is also indicated on the left of each sequence. In each panel, the bottom plot (Control-R) shows the sequence of unperturbed strides of the right paw; for each cat, the duration of swing and stance are gross averages across all unperturbed passages from all recording sessions. Dashed vertical lines show the duration of control strides for reference.

time window during which a stance could be influenced was longer than the time window for influencing the swing (Fig. 7A and B). Figure 7A helps to estimate the time window during which a brief interruption of visual input can influence the duration of stance. A period of darkness that started in the middle of the preceding stance caused a strong increase of the duration of the current stance if it extended well into the swing; shorter periods of darkness, with little or no extension into swing, had no effect or marginal effect on the stance duration (Fig. 7A; upper-left MST). Stance duration also increased when the lights went off at the beginning of the preceding swing (Fig. 7A, BSW). Similar increases of the stance duration were caused by turning the lights off at its beginning, with longer periods of darkness having a stronger effect (Fig. 7A, BST). The duration of the current stance was not affected by a darkness starting in its middle, even when the darkness extended beyond the end of stance into swing (Fig. 7A, lower-right MST). Thus, the time window in which periods of brief darkness could increase the duration of the stance started at the end of the previous stance, included the preceding swing and extended until the middle of the current stance.

Swings were affected less than stances (Fig. 7B). Very short, 200 ms periods of darkness starting at any of the tested phases of the stride (BSW, BST or MST) did not change the duration of swing. The duration of swing was

prolonged by 300 and 400 ms of darkness during the preceding stance that either covered a large part of the stance from its beginning (Fig. 7B, BST) or started in the middle of stance and extended into the beginning of the current swing (Fig. 7B, MST). Swing duration was increased only a little by 300 or 400 ms of darkness starting at the beginning of swing, despite the fact that the darkness covered the rest of the swing and the moment of landing the paw on the ladder (Fig. 7B, BSW). Thus, the time window for the brief darkness to extend the duration of a swing started around the middle of the preceding stance and continued into the current swing, whereby darkness BSW had to cover most of it to cause a minor extension.

The time windows during which brief periods of darkness increase the duration of stances and swings characterize time windows during which visual information for the respective movements is collected. Thus, visual information for planning a stance (and/or a swing of the other forelimb) is collected starting from the end of the previous stance, during the preceding swing and until the middle of the current stance. Visual information for planning a swing is collected starting from the middle of the preceding stance and continues into the current swing. Several features of these windows are worth noting. First, they are not homogeneous. Dependence of the effects of 400 ms of darkness on the phase of the step cycle was considered above (Fig. 4).

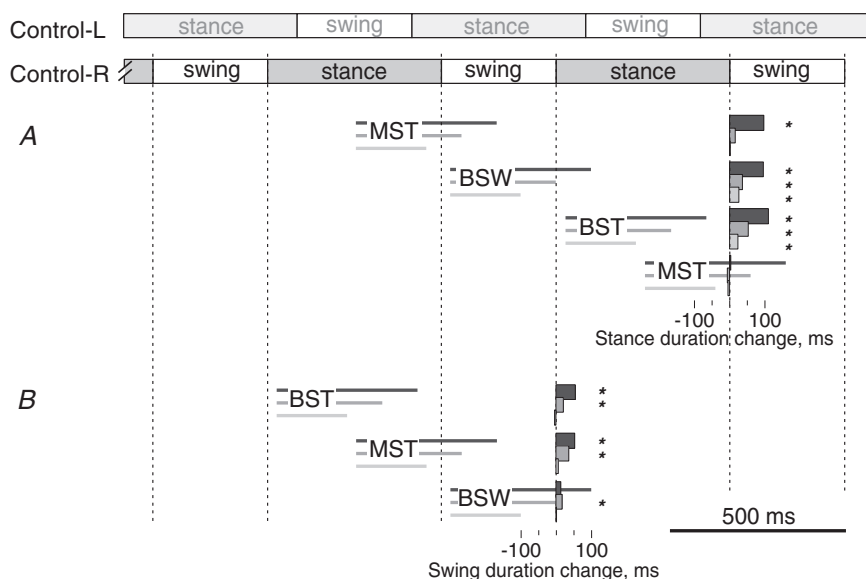


Figure 7. Summary of the effects of short periods of darkness on swings and stances during locomotion on the ladder

Top: sequence of strides of the right (Control-R) and left (Control-L, pale) forelimb during unperturbed passages. *A*, changes of the duration of stance, relative to control conditions (in milliseconds), after short periods of darkness. Changes are aligned at one stance to facilitate comparison and are shown as light, medium and dark grey bars for 200, 300 and 400 ms of darkness, respectively. Asterisks to the right of the bars mark significant changes. The light-off periods that caused these changes are shown as thin bars in the respective stance or swing. *B*, summary of the effects of short periods of darkness on swing duration. Same representation as in *A*. *A* and *B* show data from cat FM.

Figure 7 illustrates such dependence also for 300 ms of darkness. Moreover, an abrupt transition from very small or absent effects on strides of 200 ms of darkness to consistent effects of 300 ms of darkness and to pronounced and strong effects of 400 ms of darkness indicates that these windows are patchy or that brief periods of denial of visual input could be compensated. These two options are not mutually exclusive. In the scenario of patchy, non-continuous windows, 200 ms periods of denial of visual input would fall within less sensitive parts of the windows. By extension, this suggests 'hot spots' for acquisition of visual information within these windows. In the compensation scenario, visual information missed during the darkness could be acquired, in part, during the remaining portion of the window and during an

additional time of prolonged swings and stances. The compensation scenario is consistent with the fact that any prolongation of the strides is shorter than the duration of the period of darkness causing this prolongation.

The effects of brief darkness can be related to gaze fixations in cats walking on a ladder, studied in our previous work (Rivers *et al.* 2014; Zubair *et al.* 2019). Figure 8 illustrates the relationship between affected swings and stances, on the one hand, and typical distances to the sites of gaze fixations and occurrence of fixations during the step cycle, on the other. At the beginning of a swing, cats typically do not look at the target for foot landing (rung 5 in Fig. 8A), and darkness that started at the beginning of a swing (BSW) had little effect on its duration. Instead, BSW darkness of any duration (200, 300 or 400 ms) prolonged

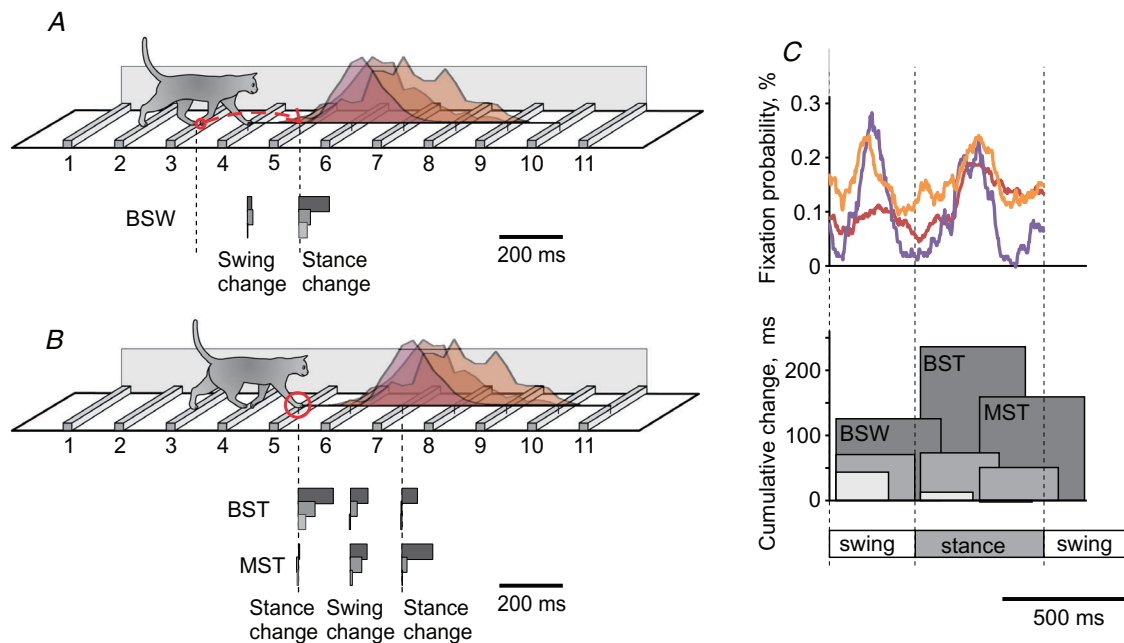


Figure 8. Relationship between the effects of short periods of darkness on strides and typical distances and phases of gaze fixations

A and B, relationship between the effects of brief periods of darkness and typical targets of gaze fixations. A, schematic diagram of a cat at the beginning of a right-forelimb swing from rung 3 to rung 5, with superimposed distributions of distances to targets of gaze fixations relative to the head in three cats (data from Rivers *et al.* 2014, reprocessed). Horizontal bar plots below the scheme show changes of the swing and stance duration (in milliseconds), relative to control strides, after 200, 300 and 400 ms darkness starting at the beginning of a swing (BSW; light, medium and dark grey bars, respectively). B, schematic diagram of a cat at the beginning of a right-forelimb stance on rung 5, with superimposed distributions of distances to targets of gaze fixations from the head (same data as in A). Bar plots below the scheme show changes of the stance, swing and next stance duration (in milliseconds), relative to control strides, after periods of darkness starting at the beginning of the stance (BST) and middle of the stance (MST). Changes after 200, 300 and 400 ms of darkness are shown as light, medium and dark grey bars, respectively. Note that at MST, the cat's head will move forwards halfway to rung 6, and the typical distance of gaze fixations will move forwards correspondingly. C, relationship between cumulative effects of brief darkness (cumulative change of the duration of two swings and two stances, starting from the 'dark-on' swing or stance) and probability of gaze fixations during the step cycle (gaze data from Zubair *et al.* 2019, reprocessed). For reference, the sequence of unperturbed strides of the right paw is shown below the plot in the bottom panel and with dashed vertical lines. In the cumulative change plot, the location and length of each rectangle show the timing and duration of the brief darkness; the height of the rectangle shows the cumulative effect of the brief darkness on strides. The upper panel shows the probability of gaze fixation, as a percentage, against the phase of the step cycle for three cats.

the following stance (Fig. 8A). Darkness starting at the beginning of stance (BST) on rung 5 prolonged this stance (Fig. 8B), and 300 or 400 ms BST darkness also prolonged the following swing to rung 7, on which cats often fixate during the stance on rung 5 (Fig. 8B; 22, 41 and 11% of fixations in three cats); 400 ms BST darkness also prolonged the next stance on rung 7. The MST darkness did not affect the 'light-off' stance, and 200 ms MST had no effect at all. Longer MST darkness of 300 and 400 ms extended the following swing onto rung 7 and the next stance on it (Fig. 8B). Again, this was the rung on which cats could fixate during the second half of stance on rung 5 (Fig. 8B; 10, 11 and 5% of fixations in three cats). Thus, brief darkness mostly affected swings and stances up to two rungs ahead from cat's head, which were within the close portion of the range of gaze fixations. In the more distant range of gaze fixations (e.g. >60 cm, 55, 25 and 78% of fixations in three cats), stepping was not affected, indicating that acquisition of visual information to compensate for the 'outage' caused by the brief darkness was completed by then.

Earlier work showed that the probability of gaze fixations changes within the step cycle (Fig. 8C, top plot; data from Zubair *et al.* 2019, reprocessed). To assess whether the dependence of the prolongation of strides on the timing of the darkness reported here (Figs 4 and 6) has a relationship to the probability of fixations at different phases of the step cycle, we plotted the cumulative change of the duration of two strides (two swings and two stances) against the timing of the darkness within the step cycle (Fig. 8C, middle panel). The plot illustrates a qualitative relationship between the effects of 400 ms darkness and probability of gaze fixations. The 400 ms BST period of darkness that covered most of the fixations during stance had the strongest effect on the stride duration. Cumulative effects of shorter periods (200 and 300 ms) of darkness did not show such a clear relationship to the probability of fixations, probably because they affected mostly one swing or stance, and the loss of visual information during such a short period of darkness could be compensated effectively.

Discussion

Our results show that brief interruptions of visual input can rapidly influence the ongoing locomotor behaviour in cats walking on a horizontal ladder and that the demand for visual input changes dynamically during the step cycle. During some phases of the cycle, a brief 200 ms denial of visual input could be tolerated fully without any changes to the strides (200 ms MST), whereas during other phases it caused changes of the stride duration (200 ms BST or BSW). The effects of longer periods of visual input denial, which always prolonged stances and/or swings, also depended on the phase of the darkness

onset. Thus, there are critical phases within the stride cycle for acquiring visual information, and interruption during these phases cannot be compensated without increasing the duration of strides and slowing down the walking. Collection of visual information for final planning of a swing starts in the middle of the preceding stance and continues into the beginning of the current swing. For a stance (and/or a swing of the other paw), visual information is collected starting from the end of the previous stance and until the middle of the current stance. The effects of brief denial of visual input on strides are closely related to the pattern of gaze behaviour, described previously in cats (Rivers *et al.* 2014; Zubair *et al.* 2019). The fact that any compensatory increase of the duration of strides was always shorter than the duration of the denial of visual input suggests that there is a redundancy in acquisition of visual information during walking on the ladder in normal conditions.

Possible causes of prolongation of swings or stances

In cats walking on the ladder, brief denial of visual input can increase the duration of one or two, sometimes three stances and/or swings. This finding is consistent with several lines of evidence from earlier studies. Cats typically look two to four steps ahead when walking on the ladder (Fig. 8; Rivers *et al.* 2014) or down a cluttered alley (Fowler & Sherk, 2003). In a cluttered alley, cats can make one or two, sometimes up to four steps after the light is turned off (Wilkinson & Sherk, 2005). Also, humans look two to three steps ahead when walking on complex terrains (e.g. Matthis *et al.* 2018).

What are possible causes of an increase of the duration of swings and/or stances after a brief denial of visual input? The primary reason for a prolongation might be the requirement for additional time to acquire visual information that was missed during the denial period. Given that visual information is used for planning future steps, such a scenario predicts that affected could be not only the steps that are covered by a darkness period, but also future swings or stances that start after the light is back on. Our results support this prediction. This interpretation is consistent with conclusions of earlier work on the effects of limiting visual information during locomotion in complex environments, both in cats (e.g. Sherk & Fowler, 2001; Wilkinson & Sherk, 2005) and in humans (e.g. Laurent & Thomson, 1988; Hollands & Marple-Horvat, 1996). In a broader context, slowing down of strides due to interruption of visual input could be caused by an overall slowing down of visual processing, communicating results to motor systems and respective slowing down of the creation of motor programs. In such scenario, availability of visual information is a bottleneck for the speed of ongoing motor planning and

locomotion, as suggested for human walking on rough terrain (Matthis *et al.* 2018). This idea is compatible with results of experiments in cats, who reduced the speed of walking along a cluttered alley as illumination changed from bright light to dim light and to strobe lighting (Sherk & Fowler, 2001). In addition to these direct effects, i.e. increase of the duration of strides for catching up with acquiring and processing 'missed' visual information, large prolongation of strides could lead to compensatory adjustments of locomotion owing to biomechanical constraints on changes of the walking speed.

In contrast, missing visual feedback directly involved in execution of ongoing motor programs is not a likely cause of prolongation of strides. If that were the case, only the strides covered by darkness would be affected, and swings would be affected more strongly than stances. Our results do not support these predictions. In addition, analysis of gaze shows that cats do not look at the crosspiece on which the paw in swing will be landing, but rather look ahead (Fig. 8; Fowler & Sherk, 2003; Rivers *et al.* 2014). Attentional changes and reaction to the novelty of turning the lights off and on were also unlikely to be the major causes of the observed increase in stride duration. Cats were extensively trained, familiar with all aspects of the experiments, and accomplished numerous passages through the walking chamber, during which the lights were turned off briefly. Moreover, novelty reaction predicts immediate effects on the swings and stances during which the lights were turned off and on, and independence of such effects on the duration of darkness and the phase of its onset. Neither of these predictions is supported by our data.

Thus, we conclude that an increase of the duration of strides in our experimental conditions is caused by the reduced availability of visual information for planning future strides and reflects the need for additional time to compensate for the acquisition of visual input and its processing.

How is vision used during accurate stepping?

Vision is required for accurate stepping on a complex terrain, because during long periods of darkness (seconds; Beloozerova & Sirota, 2003) cats stop walking on a ladder when the light goes off. Humans also need to see obstacles to avoid them (reviewed by Patla, 1997). In contrast, vision is not crucial for locomotion on a flat surface because cats continue to walk on a flat surface even after the lights go off (Beloozerova & Sirota, 2003).

In theory, vision can be used for planning strides and motor programs for accurate stepping on the crosspieces of the ladder and/or for execution of motor programs, i.e. using direct visual guidance of movements, such as

dynamic adjustment of the movement using dynamically updated visual estimation of the distance between the paw and the stepping target. The following lines of evidence show that vision is used for planning of strides, but not for execution of the locomotor program. (i) A swing, when started, is completed in darkness. For example, 400 ms BSW and some 300 ms BSW periods of darkness covered the end of the swing and landing of the paw, but the swing was completed successfully nevertheless and was of about same duration as in control conditions (Fig. 7). (ii) During a swing, cats do not look with their central vision at the crosspiece on which the paw is going to land (Fig. 8; Fowler & Sherk, 2003; Rivers *et al.* 2014). This crosspiece may still be within cat's peripheral visual field; however, results of Wilkinson & Sherk (2005) show that cats are using central vision during locomotion on a cluttered surface. (iii) A swing can start in darkness, e.g. 400 ms MST and some of 300 ms MST periods of darkness covered the swing onset. Moreover, the swing in these conditions was started on time, as it would start normally in full light (Fig. 7). (iv) Darkness starting mid-stance and covering the onset of a swing (MST 400 ms) or covering most of stance and ending shortly before swing onset (BST 400 ms) makes the swing longer. Notably, darkness starting immediately after the swing onset (BSW 200, 300 or 400 ms) has no effect or minimal effect on its duration (Fig. 7). Taken together, these results show that execution of a current swing is not guided visually but is already programmed before it starts.

When vision is needed: dynamics of the demand on visual information and planning movement during accurate stepping

Prolongation of swings and stances depended on both the phase of the stride cycle at which the darkness started and its duration. The observed effects of the denial of visual input on swings and stances were closely related to the typical pattern of gaze behaviour. When walking on the ladder, cats typically make fixations around the middle of swing and the middle of stance (Zubair *et al.* 2019), mostly on the second and third, and less often the fourth crosspiece ahead (Fig. 8; Rivers *et al.* 2014). Our results show that stepping on the crosspieces in the close range of fixations (first and second, <60 cm ahead) is most affected by brief denial of visual input, supporting the notion that visual information for the next steps is acquired during these fixations.

Darkness starting at the beginning of the right paw swing from rung 3 to rung 5 (Fig. 8A) had little effect on that swing. Gaze data (Fig. 8A; Rivers *et al.* 2014) showed that while making the swing, cats did not look at rung 5, which was the target for paw landing. Thus, the motor program for this swing was already in place

and was executed despite interruption of visual input. The duration of the next stance on rung 5 was increased by 26, 36 or 95 ms after BSW 200, 300 or 400 ms, respectively. The self-selected gait was then restored, and stepping on rungs 6 (left paw) and 7 (right paw) was as in control passages, indicating that this prolongation was sufficient for compensatory collection and processing of the required visual information.

Darkness starting at the beginning of stance on rung 5 (Fig. 8B) prolonged that stance by 23, 53 or 109 ms after BST 200, 300, or 400 ms, respectively. A minor prolongation by 23 ms was sufficient for a complete compensation of visual input denied by a 200 ms BST. After 300 ms BST, also the next swing to rung 7 was slightly extended before the self-selected pace was restored. After a strong disruption by 400 ms BST (>160 ms cumulative increase of the duration of stance and swing), cats were not able to return to their normal pace until after two strides, either because disruption of the visual input was too severe or owing to limitations imposed by biomechanics of cat locomotion. Darkness starting in the middle of stance had no effect on the duration of that stance, indicating that the motor program for the next swing to rung 7 was already in place at that time, and the swing started on time, as it would with continuous visual input. Notably, 200 ms MST had no effect on walking at all, showing that visual input during this period was either not used for planning future steps or that its absence could be compensated completely. The 300 or 400 ms MST period of darkness prolonged the following swing to rung 7 (Fig. 8B), and 400 ms MST also prolonged the next stance at rung 7, before the cat returned to the self-selected pace.

An overall pattern of the effects of brief periods of denial of visual input on the duration of swings and stances shows a clear correspondence with typical targets for gaze fixations (Fig. 8). Immediate swings at the crosspieces at which cats did not look were not affected, whereas stepping on the crosspieces at which they typically fixate was affected when visual input was interrupted by darkness. At the same time, the need for compensatory acquisition of visual information after a brief period of darkness was restricted to the crosspieces within the near range of gaze fixations, because only one swing and/or one stance were typically prolonged.

The above considerations also suggest the redundancy of the acquisition of visual information for accurate stepping. Any interruptions of visual input for 200–400 ms were rapidly compensated by severalfold shorter (few dozens of milliseconds) increases of the duration of swings and stances, after which the normal self-selected speed of walking was typically restored. Stepping on the crosspieces that were the second and third ahead of the cat at the onset of darkness was prolonged. Notably, at least some information about these crosspieces must

have been collected before, during the stride before the dark-perturbed one, because cats sometimes fixate at the fourth crosspiece ahead (Fig. 8). They also often glance over the crosspieces using constant gaze before fixating (Zubair *et al.* 2019). This information might be used for creating a coarse locomotor program but was not sufficient for finalizing motor programs for accurate stepping on the second and third crosspieces ahead, which required additional, actualized visual information for their fine-tuning.

Outlook and open questions

The results of the present study open a number of new perspectives for further research into visuomotor coordination. The first group of questions is related to acquisition of visual information that was missed during denial of visual input. Would the denial lead to compensatory increase of gaze fixations or constant gaze, or both, and would their dependence on step cycle be affected? If yes, which crosspieces of the ladder will be preferred targets for additional fixations or scans by the constant gaze? Addressing these questions will help to reveal which specific pieces of missed visual information were so crucial for maintaining the accuracy of stepping that strides had to be prolonged to acquire them, and whether fixations or constant gaze would be the preferred acquisition strategy. Another group of questions concerns neuronal mechanisms and pathways mediating visual influence on locomotion. At what level of the motor system does denial of visual input interfere with locomotor programs to prolong stances and swings? How is the activity of neurons in the locomotor network affected by the denial of visual input? A particularly interesting aspect here concerns the neuronal mechanisms of the prolongation of swings and stances that are made after the lights were turned on again. In other words, how does a compensatory delay in acquisition and processing of visual information translate into slowing down of future strides? Addressing these questions will shed light on temporal aspects of the interaction between neurons in visual and motor structures and temporal constraints on using vision for locomotion. Finally, there is an interesting implication for cognitive processes taking place during locomotion. Denial of visual input during certain phases of stride cycle (200 ms MST) has no effect on strides. Does this interval represent an optimal time window for looking around while walking? At least, absence of effects on strides suggests that during this period the visual system is not serving the needs of locomotion and could be used for other tasks. A speculative prediction here is that performance on visual cognitive tasks, e.g. a visual discrimination task, might be enhanced during the second

half of stance or, more generally, depend on the timing of the task within the step cycle. Research into stride-phase dependence of cognition might shed light on the temporal aspects of the interaction between the ventral (What?) pathway serving visual cognition and the dorsal (Where?) pathway serving the needs of motor systems.

References

- Beloozerova IN, Farrell BJ, Sirota MG & Prilutsky BI (2010). Differences in movement mechanics, electromyographic, and motor cortex activity between accurate and non-accurate stepping. *J Neurophysiol* **103**, 2285–2300.
- Beloozerova IN & Sirota MG (1993). The role of the motor cortex in the control of accuracy of locomotor movements in the cat. *J Physiol* **461**, 1–25.
- Beloozerova IN & Sirota MG (2003). Integration of motor and visual information in the parietal area 5 during locomotion. *J Neurophysiol* **90**, 961–971.
- Chu KMI, Seto SH, Beloozerova IN & Marlinski V (2017). Strategies for obstacle avoidance during walking in the cat. *J Neurophysiol* **118**, 817–831.
- de Carvalho WD, Rosalino LM, Dalponte JC, Santos B, Harumi Adania C & Lustosa Esbérard CE (2015). Can footprints of small and medium sized felids be distinguished in the field? Evidences from Brazil's Atlantic Forest. *Trop Conserv Sci* **8**, 760–777.
- Fowler GA & Sherk HA (2003). Gaze during visually-guided locomotion in cats. *Behav Brain Res* **139**, 83–96.
- Grundy D (2015). Principles and standards for reporting animal experiments in *The Journal of Physiology and Exp Physiol*. *J Physiol* **593**, 2547–2549.
- Guenther E & Zrenner E (1993). The spectral sensitivity of dark- and light-adapted cat retinal ganglion cells. *J Neurosci* **13**, 1543–1550.
- Gunter R (1951). The absolute threshold for vision in the cat. *J Physiol* **114**, 8–15.
- Hollands MA & Marple-Horvat DE (1996). Visually guided stepping under conditions of step cycle-related denial of visual information. *Exp Brain Res* **109**, 343–356.
- Hollands MA, Marple-Horvat DE, Henkes S & Rowan AK (1995). Human eye movements during visually guided stepping. *J Mot Behav* **27**, 155–163.
- LaMotte RH, & Brown JL (1970). Dark adaptation and spectral sensitivity in the cat. *Vision Res* **10**, 703–716.
- Lankheet MJ, Rowe MH, van Wezel RJ & van de Grind WA (1996). Horizontal cell sensitivity in the cat retina during prolonged dark adaptation. *Vision Neurosci* **13**, 885–896.
- Laurent M & Thomson JA (1988). The role of visual information in control of a constrained locomotor task. *J Mot Behav* **20**, 17–37.
- Matthis JS, Barton SL & Fajen BR (2017). The critical phase for visual control of human walking over complex terrain. *Proc Natl Acad Sci U S A* **114**, E6720–E6729.
- Matthis JS, Yates JL & Hayhoe MM (2018). Gaze and the control of foot placement when walking in natural terrain. *Curr Biol* **28**, 1224–1233.e5.
- Nguyen CT, Iyer GS, Volgushev MA & Beloozerova IN (2019). Denial of visual information during swing phase of the stride prolongs the following stance in cats. *Neuroscience Meeting Planner*, Program No. 227.07, Society for Neuroscience, Chicago, IL. Online. <https://www.abstractsonline.com/pp8/#!/7883/presentation/52428>
- Patla AE (1997). Understanding the roles of vision in the control of human locomotion. *Gait and Posture* **5**, 54–69.
- Pryor K (1975). *Lads before the wind: adventures in porpoise training*. New York: Harper & Row.
- Rivers TJ, Sirota MG, Guttentag AI, Ogorodnikov DA, Shah NA & Beloozerova IN (2014). Gaze shifts and fixations dominate gaze behavior of walking cats. *Neuroscience* **275**, 477–499.
- Sherk H & Fowler GA (2001). Visual analysis and image motion in locomoting cats. *Eur J Neurosci* **13**, 1239–1248.
- Skinner BF (1938) *The behavior of organisms: an experimental analysis*. New York: Appleton-Century-Crofts.
- Wilkinson EJ & Sherk HA (2005). The use of visual information for planning accurate steps in a cluttered environment. *Behav Brain Res* **164**, 270–274.
- Zubair HN, Chu KMI, Johnson JL, Rivers TJ & Beloozerova IN (2019). Gaze coordination with strides during walking in the cat. *J Physiol* **597**, 5195–5229.

Additional information

Data availability statement

The data supporting the findings of this study are available from the corresponding author (M.V.; maxim.volgushev@uconn.edu) or senior author (I.N.B.; ibeloozerova3@gatech.edu) upon request.

Competing interests

We have no competing interests to report.

Author contributions

Experiments were performed in the Motor Systems Neurophysiology laboratory headed by Dr Irina N. Beloozerova at the Barrow Neurological Institute in Phoenix, AZ, USA. M.V., C.T.N. and I.N.B. designed the research; C.T.N. and I.N.B. performed the experiments; M.V., C.T.N., G.S.I. and I.N.B. analysed the data; M.V. and I.N.B. wrote the manuscript. All authors approved the final version of the manuscript and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. All persons designated as authors qualify for authorship, and all those who qualify for authorship are listed.

Funding

This work was supported by the National Institutes of Health the National Institute of Neurological Disorders and Stroke (grant

R01 NS-058659 to I.N.B.), the National Eye Institute (grant R21 EY033071 to I.N.B. and M.V.) and the National Science Foundation (grant 1912557 to I.N.B.).

Acknowledgements

This study would not have been possible without the contribution of a great group of enthusiastic high school students and Arizona State University undergraduates who have contributed numerous hours to training cats, acquisition of the data and their preliminary analyses: Meghan Foe, Sandy Seto, Hai Tran, Humza Zubair, Kevin Chu, David Tze and Alicia Wu. We are grateful to Mr Peter Wettstein for exceptional engineering assistance.

Keywords

cat, ladder, locomotion, vision, visuomotor integration, walking on complex surfaces

Supporting information

Additional supporting information can be found online in the Supporting Information section at the end of the HTML view of the article. Supporting information files available:

Peer Review History

Statistical Summary Document