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Night vision by cuttlefish enables changeable camouflage

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SUMMARY

Because visual predation occurs day and night, many predators must have good night vision. Prey therefore exhibit antipredator behaviours in very dim light. In the field, the giant Australian cuttlefish (*Sepia apama*) assumes camouflaged body patterns at night, each tailored to its immediate environment. However, the question of whether cuttlefish have the perceptual capability to change their camouflage at night (as they do in day) has not been addressed. In this study, we: (1) monitored the camouflage patterns of *Sepia officinalis* during the transition from daytime to night-time using a natural daylight cycle and (2) tested whether cuttlefish on a particular artificial substrate change their camouflage body patterns when the substrate is changed under dim light (down to starlight, 0.003 lux) in a controlled light field in a dark room setting. We found that cuttlefish camouflage patterns are indeed adaptable at night: animals responded to a change in their visual environment with the appropriate body pattern change. Whether to deceive their prey or predators, cuttlefish use their excellent night vision to perform adaptive camouflage in dim light.

Key words: crepuscular, visual perception, low light, visual predation, *Sepia officinalis*, scotopic vision.

INTRODUCTION

Camouflage body patterns in cephalopods have to date been studied mostly in daytime, both in the field and in the laboratory (e.g. Hanlon et al., 2005; Hanlon et al., 2008; Hanlon and Messenger, 1988; Kelman et al., 2007; Mäthger et al., 2006; Norman et al., 2001), partly because of the human diurnal life style but also because of the extra challenge of recording behavioural data under low-light conditions without impacting and therefore changing an animal's natural behaviour. Hanlon et al. reported the first evidence from field studies that the giant Australian cuttlefish *Sepia apama* uses camouflage body patterns at night (Hanlon et al., 2007). Hanlon and colleagues used non-invasive red lighting that did not affect the behaviour of the cuttlefish, whose visual system is tuned to the green parts of the spectrum (Bellingham et al., 1998; Brown and Brown, 1958; Marshall and Messenger, 1996), but that provided sufficient illumination for video recording the camouflage body patterns.

Cephalopods (squid, cuttlefish and octopus) show a variety of nocturnal and diurnal behaviours (Hanlon and Messenger, 1996). Even within cuttlefish species, there is some variation. The giant Australian cuttlefish *S. apama* appears to be primarily diurnal/crepuscular. All reproductive signalling behaviour ceases at night and is replaced by camouflage to avoid detection by predators (Hanlon et al., 2007). *Sepia officinalis*, by contrast, has been reported to be cathemeral, active both day and night (Watanuki et al., 2000); some of their physiological processes undergo diurnal cycles, suggesting that physical activity might be increased at night (Denton and Gilpin-Brown, 1961; Mark et al., 2007). Irrespective of diurnal activity changes, cuttlefish night vision must be well developed (either to detect prey or to avoid becoming prey), and we might therefore expect their camouflage body patterns to be fine tuned and changeable, even at night. In fact, dolphins preferentially forage in dim light conditions and their diets include cephalopods (e.g. Pusineri et al., 2007; Silva, 1999). Certainly, a large proportion of the world's animals are active in low-light conditions, either at night or at greater depths in the sea (Warrant and Locket, 2004). Only in recent years has there been an increasing research interest in vision and the behaviour of animals that are active in low-light conditions (e.g. Chuang et al., 2008; Penteriani et al., 2006; Pirhofer-Walzl et al., 2007; Pusineri et al., 2007; Sazima and Uieda, 1979; Silva, 1999; Warrant, 2004; Warrant, 2008; Warrant and Locket, 2004).

In cuttlefish, camouflage body patterns have been grouped into patterns that function by background matching ('Uniform' and 'Mottle' patterns) and those that function presumably by disruptive colouration (see Cott, 1940; Hanlon et al., 2009; Stevens and Merilaita, 2009a; Stevens and Merilaita, 2009b). A Uniform pattern is defined by a uniform light or dark pattern that is distributed equally across the body of the animal (i.e. little or no contrast). Mottle patterns are characterized by light and dark 'splotches' that are distributed across the body (Chiao et al., 2010). In a Disruptive pattern, the animal shows large transverse and longitudinal light and dark components that tend to disrupt the animal's body outline (Hanlon et al., 2009). Uniform patterns can be evoked on uniform substrates, such as uniform artificial computer print outs, or fine sand with little or no contrast (e.g. Allen et al., 2009; Allen et al., 2010; Mäthger et al., 2008). Cuttlefish show Mottle patterns on small-scale moderate-to-high contrasting substrates, such as small black and white checkerboards or natural substrates with small particles (check or particle size 3 to 12% of the size of the animal's White square component, "a rectangular area centered on the dorsal mantle") (Hanlon and Messenger, 1988) (for details, see Barbosa et al., 2004; Barbosa et al., 2007; Chiao et al., 2010; Mäthger et al., 2008). Disruptive patterns are evoked on large-scale high-

contrasting substrates with defined edges, such as large black and white checkerboards or natural rocks (white check or white rock size 40 to 120% of the size of the animal's White square) (Barbosa et al., 2007; Mäthger et al., 2008; Zylinski et al., 2009).

Hanlon et al. (Hanlon et al., 2007) recorded the camouflage body patterns of 71 giant Australian cuttlefish at night and noted that four were Uniform, 33 were Mottle and 34 were Disruptive. The respective backgrounds were not analysed in their study but it appeared that the camouflage body patterns were tailored to the different backgrounds. Furthermore, their study did not address whether cuttlefish change body patterns in response to changes in their visual environment at night.

In this study, we addressed two questions. (1) Do cuttlefish *S. officinalis* continue to camouflage as natural light levels change during the transition from daytime to night-time? (2) In very dim light, is this camouflage behaviour adaptable, i.e. can the animal actively analyse visual information from its surroundings to choose an appropriate camouflage body pattern?

MATERIALS AND METHODS

Animals and substrates

Cuttlefish *Sepia officinalis* (Linnaeus 1758) were maintained at the Marine Resources Center at the Marine Biological Laboratory in Woods Hole, MA, USA. Twenty-two cuttlefish with mantle lengths between 4.0 and 5.7 cm were used, 12 in experiment 1 and 10 in experiment 2. Animals were tested on artificial substrates known to evoke the three major body patterns (Barbosa et al., 2007; Hanlon et al., 2007): Uniform (solid grey), Mottle (high-contrast checkerboard with check size approximately 5% of the size of the animals' White square areas, hereafter 'small check') and Disruptive (high-contrast checkerboard with check size approximately 100% the size of the animals' White square areas, hereafter 'large check'). The substrates were presented to the cuttlefish on both the floor and the wall of the arena.

Experiment 1: natural light

A shallow tray was filled with seawater and equipped with twelve small arenas each 13 cm in diameter. Owing to their proximity to each other, the arenas received similar amounts of ambient light. The seawater was changed periodically without disturbing the animals to maintain a consistent temperature and dissolved oxygen content. Experiments began mid-afternoon, approximately 2–3 h before sunset and ended approximately 2–3 h after sunset. Each photograph was accompanied by light measurements near the animal's arena using an Extech Easyview EA30 light meter (giving lux values; Fig. 1A, grey lines, one line for each of three nights measured). The light intensity measurements obtained in this way were reliable to as low as approximately 0.05 lux, but not lower (limited by light meter sensitivity). Therefore, experiment 2 was more trustworthy than experiment 1 regarding light intensities during which adaptive camouflage is accomplished. A flash photograph was taken of each animal every twenty minutes using a SONY HDR-HC1 video camera. Photography was assisted by a 'night mode' function that uses infrared light to focus the lens; the animals did not appear to be disturbed by the flash (i.e. they did not move, ink or change body patterns in response to the flash). Sixteen images and light measurements were taken each night; one substrate was tested per night for a total of three experimental nights (grey, small check and large check substrates to evoke Uniform, Mottle and Disruptive body patterns, respectively) over a period of five days to minimize natural changes in day length and light availability.

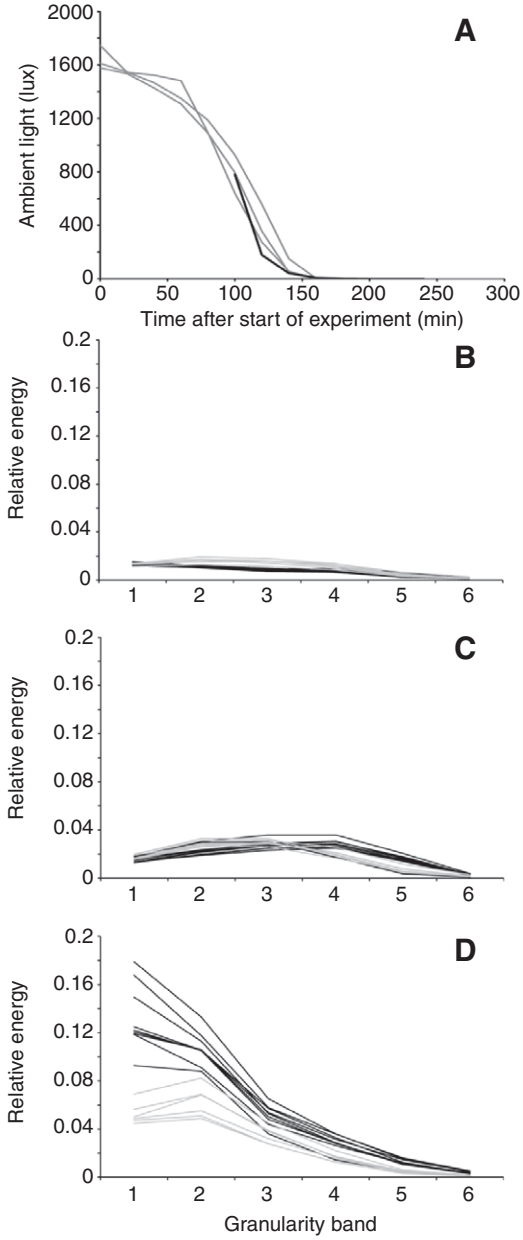

Fig. 1. (A) Light levels were similar among nights where natural light was measured (experiment 1, grey lines) and where controlled light was calculated (experiment 2, black line). In experiment 1, natural light was measured every 20 min on each of three nights (one night of measurements for each substrate type). On the night the grey substrate (evokes Uniform) was tested, the range of measureable light was 1578–0.6 lux; six measurements were taken beyond the sensitivity of our light meter (<0.05 lux). On the night the small check substrate (evokes Mottle) was tested, the range of measurable light was 1612–0.05 lux; six measurements were taken beyond the sensitivity of our light meter (<0.05 lux). On the night the large check substrate (evokes Disruptive) was tested, the range of measureable light was 1746–1.52 lux; seven measurements were taken beyond the sensitivity of our light meter (<0.05 lux). In experiment 2, lighting conditions were controlled using filters to mimic the natural decrease in light as the day proceeds through dusk to night. The light intensity under different combinations of filters was calculated and plotted (black line) with the lighting conditions measured in experiment 1. (B–D) Curves from granularity statistics for: (B) grey substrate (evokes Uniform), (C) small check substrate (evokes Mottle) and (D) large check substrate (evokes Disruptive). Each line is the mean of 12 animals at each measurement time. For simplicity and because there was no trend between curve shape and time, curves are shown before sunset (black lines) and after we reached the limit of sensitivity of our light meter (<0.05 lux, grey lines).

Experiment 2: controlled light

Ten cuttlefish were tested in a dark room under controlled light conditions designed to imitate the natural decrease in light (compare the black line to the grey lines in Fig. 1A). Light levels were changed by covering a white light source with sheets of 0.6 neutral density filters (LEE Filters #210, Burbank, CA, USA); each sheet decreased the light by 75%. During the first 75 min, one filter was added every 15 min to decrease the light by 75% at each time step. During the last 45 min, two filters were added every 15 min to decrease the amount of light by 150%. In total, the light was decreased in eight steps: 780 lux (light source with no filters; typical late afternoon), 178 lux (early evening), 42 lux (late evening), 9 lux (twilight), 2 lux (deep twilight), 0.2 lux (full moon), 0.01 lux (quarter moon), 0.003 lux (starlight) (Fig. 1A, black line).

One cuttlefish was placed in each of two arenas approximately 13 cm in diameter and the substrate was presented on both the floor and the wall of the arena. After acclimating to the arena, each cuttlefish was photographed following the same method described for experiment 1. Under the lowest light intensity (0.003 lux), the substrate and arena wall were changed to present visual cues known to stimulate a different body pattern. Before the experiment commenced, a second substrate and wall combination was concealed under an initial substrate and wall combination. Substrates and walls were changed by carefully removing the top substrate to expose the second substrate. Substrate combinations that evoked the following body pattern categories were tested: grey (evokes Uniform) substrate to small check (evokes Mottle) and to large check (evokes Disruptive); small check (evokes Mottle) to grey (evokes Uniform) and to large check (evokes Disruptive); large check (evokes Disruptive) to grey (evokes Uniform) and to small check (evokes Mottle). Animals were allowed to spend 5 min on the new substrate, then were photographed as above.

Data analysis

Images from both experiment 1 and experiment 2 were analysed using the automated grading method introduced by Barbosa et al. (Barbosa et al., 2008). In brief, each animal was cut from its background and warped to conform to a standard template. The image was then band-pass filtered to gather spatial frequency information over six energy bands. These 'granularity statistics' were averaged for all 12 (experiment 1) or 10 (experiment 2) animals for each time period. The average statistics were then graphed in curves that described the body pattern (Fig. 1B–D) (see also Barbosa et al., 2008; Chiao et al., 2010).

Granularity statistics from experiment 2 were converted into measures of mean granularity for each animal under each lighting condition. In brief, the mean granularity (*MG*) is used to quantify the shape of the granularity curve and is calculated using:

$$MG = \frac{\sum_{g=1}^{6} gS(g)}{TE},$$

where g is the granularity band number (1–6), S is the relative energy (granularity statistic) and TE is total spectrum energy, the sum of granularity bands 1–6. For more details, please see Chiao et al. (Chiao et al., 2009; Chiao et al., 2010).

The distributions of mean granularity statistics for each light level were tested for sphericity using Mauchly's test of sphericity and were compared using a repeated measures ANOVA. When this ANOVA was significant, the distributions were compared with *post hoc* pairwise comparisons of marginal means with a Bonferroni correction. All statistical analyses were performed in SPSS 11.5.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

Experiment 1: natural light

Grey substrate (evokes Uniform)

Granularity statistics showed little variation in cuttlefish Uniform body pattern under different natural light conditions. The curves for body patterns shown under measureable amounts of light (1578 lux to 0.6 lux) were very flat, indicating low energy in each of the spatial

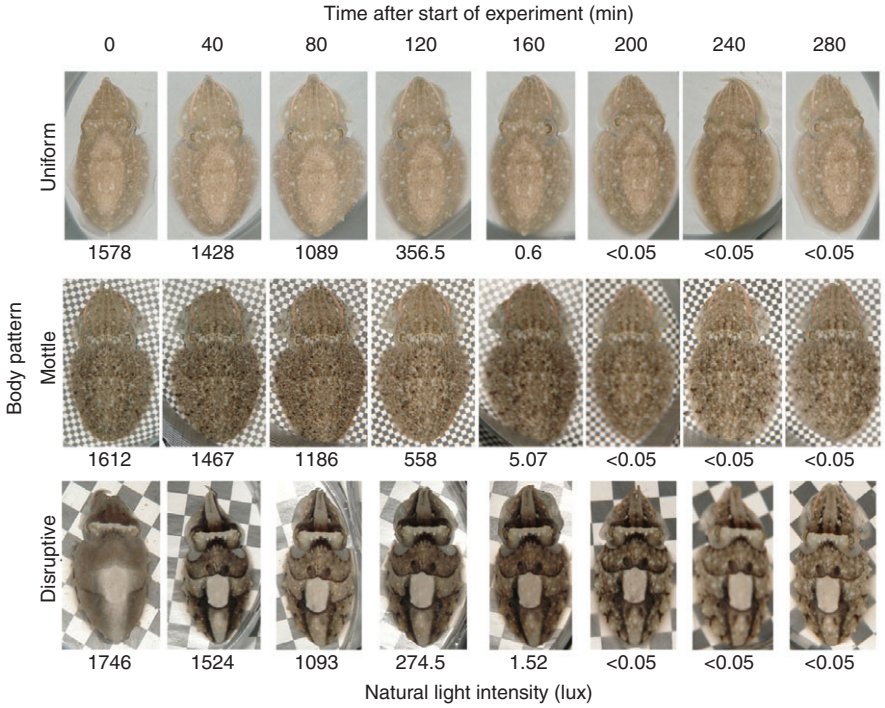

Fig. 2. Results from experiment 1. Representative animals for Uniform, Mottle, and Disruptive body patterns at different intensities of natural light. To keep the figure concise, images shown are odd-numbered data points, i.e. taken at time 0, 40, 80 min, etc., indicated above each column. Below each image is the light intensity in lux measured immediately before the photograph was taken.

frequency bands (Fig. 1B, black lines). The curves for body patterns shown in dim light (less than 0.05 lux) were essentially identical (Fig. 1B, grey lines). This curve shape is consistent with the known characteristics of Uniform body patterns (e.g. Barbosa et al., 2008; Chiao et al., 2010). Examination of the photographs taken during this experiment revealed no obvious change in Uniform body pattern (Fig. 2).

Small check substrate (evokes Mottle)

Granularity statistics showed very slight variation in cuttlefish Mottle body pattern under different natural light conditions. The curves for body patterns shown under measureable amounts of light (1612 lux to 0.05 lux; Fig. 1C, black lines) peaked in granularity bands 3 and 4, whereas curves for body patterns that were shown in dim light (less than 0.05 lux) peaked in granularity bands 2 and 3 (Fig. 1C, grey lines). This result was very subtle, however, and we deem it not important because we find that Mottle body patterns typically translate into curves that peak between granularity bands 2 and 4 (e.g. Barbosa et al., 2008; Chiao et al., 2010); that is, both types of curves found here are characteristic for Mottle body patterns. Examination of the photographs taken during this experiment revealed no obvious change in Mottle body pattern (Fig. 2).

Large check substrate (evokes Disruptive)

Granularity statistics showed some variation in cuttlefish Disruptive body pattern under different natural light conditions. The curves for body patterns shown under measureable amounts of light (1746 lux to 1.52 lux; Fig. 1D, black lines) peaked in granularity band 1 and had slightly higher amplitudes than the curves for body patterns measured in very dim light (less than 0.05 lux; Fig. 1D, grey lines). Regardless of amplitude, all curves had the characteristic shape for Disruptive body patterns, with most energy in bands 1 and 2, followed by a steep negative slope in bands 3–6 (e.g. Barbosa et al., 2008; Chiao et al., 2010). Examination of the photographs taken during this experiment revealed some changes in Disruptive body pattern (Fig. 2). For example, measurements that were taken during the darkest time periods revealed many animals with Mottle regions along the side of the mantle (Fig. 2, right side). Many distinctive Disruptive components persisted throughout the experiment [see Hanlon and Messenger (Hanlon and Messenger, 1988) for a definition of the components]. After the light meter reached the limit of its sensitivity (Fig. 1D, grey lines), all 12 animals showed strong expression of the White head bar, 10 animals showed the Dark anterior head bar, nine animals showed strong expression of the White square, and nine animals showed the White posterior triangle.

Experiment 2: controlled light

Grey substrate (evokes Uniform)

In these trials, animals were presented with a uniform grey substrate that was changed to either a large check (evokes Disruptive) or a small check (evokes Mottle) substrate at the dimmest light intensity (0.003 lux). All 10 animals maintained a Uniform body pattern on the grey substrate under all experimental light conditions. The granularity statistics produced curves that were characteristic of

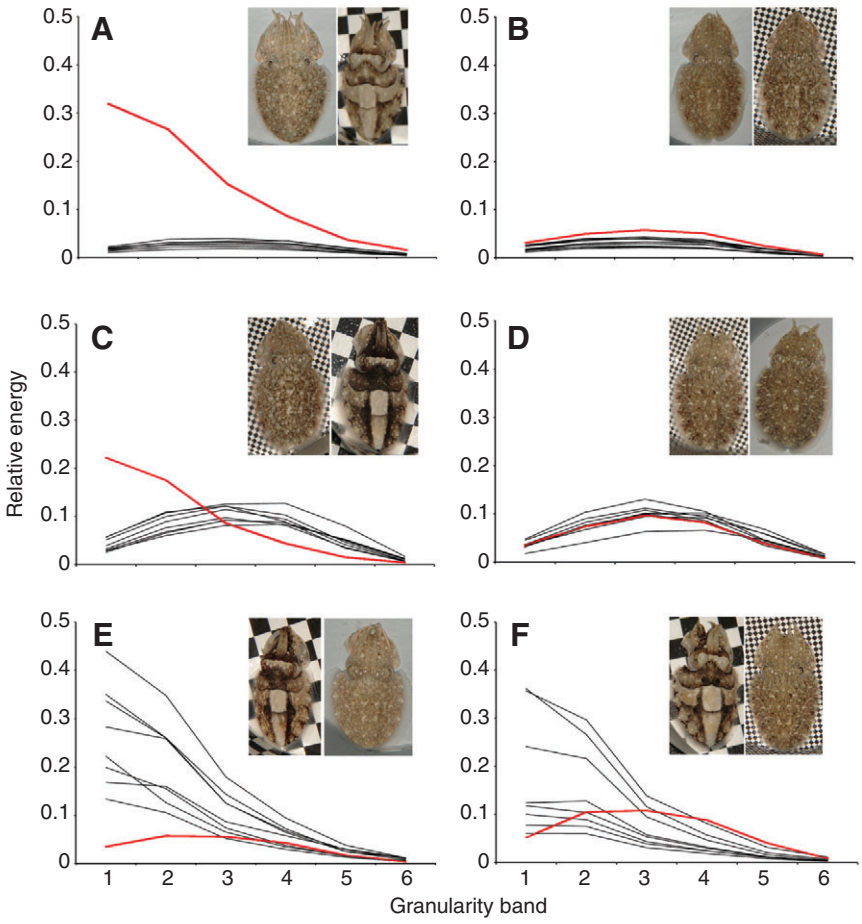

Fig. 3. Results from experiment 2. Curves from granularity statistics and representative images for six body pattern changes evoked by substrate changes at a light intensity of 0.003 lux (starlight): (A) grey to large check (Uniform to Disruptive); (B) grey to small check (Uniform to Mottle); (C) small check to large check (Mottle to Disruptive); (D) small check to grey (Mottle to Uniform); (E) large check to grey (Disruptive to Uniform); and (F) large check to small check (Disruptive to Mottle). For simplicity and because there was no trend between curve shape and time, curves are shown before (black lines) and after (red lines) the substrate was switched. Each of the black lines is the average granularity curve for all 10 animals at one of the eight steps as the light was decreased using filters (discussed in the Materials and methods). The red line is the average granularity curve for all 10 animals after the substrate was switched at a light intensity of 0.003 lux. Representative images are body patterns on the same cuttlefish before (left) and after (right) the substrate was changed.

Uniform body patterns, i.e. flat, with low energy in all six granularity bands (Fig. 3A,B, black lines).

Grey (evokes Uniform) to large check (evokes Disruptive)

After the substrate was changed to a large check substrate, cuttlefish showed a Disruptive body pattern. The granularity statistics produced a curve with high energy in bands 1 and 2 and a steep negative slope, characteristic of Disruptive body patterns (Fig. 3A, red line). This was confirmed by image data (Fig. 3A, images).

Mauchly's test of sphericity indicated that sphericity was not violated for these data (P=0.080). A repeated measures ANOVA for within-subjects effects was significant ($F_{8,72}$=11.556, P<0.01). *Post hoc* pairwise comparisons of marginal means with a Bonferroni correction revealed that the mean granularity (a quantification of the shape of the granularity curve) was not significantly different for animals on the initial substrate, grey, under eight light levels. After the substrate was switched to large check, the mean granularity of the body pattern on the new substrate was significantly different from the mean granularity of the body patterns on the initial substrate under all light conditions (P<0.014 for each comparison; Fig. 4A).

Grey (evokes Uniform) to small check (evokes Mottle)

After the substrate was changed to a small check substrate, cuttlefish showed a Mottle body pattern. The granularity statistics produced a curve that was very similar to that of the Uniform animals but with slightly more energy in band 3 than for the Uniform animals (Fig. 3B, red line). Examination of the image data revealed Mottle body patterns with slightly higher contrast light and dark patches than were observed with the Uniform body patterns (Fig. 3B, images).

Mauchly's test of sphericity indicated that sphericity was not violated for these data (P=0.318). A repeated measures ANOVA for within-subjects effects was not significant ($F_{8,72}$=0.776, P=0.625). This test suggested that there was no difference between the mean granularity for body patterns shown when the cuttlefish sat on the initial substrate, grey, under eight different light levels and the mean granularity for body patterns shown after the substrate was switched to small checks (Fig. 3B, Fig. 4B).

Small check substrate (evokes Mottle)

In these trials, animals were presented with a small check substrate that was changed to either a large check (evokes Disruptive) or to a grey (evokes Uniform) substrate at the dimmest light intensity (0.003 lux). All ten animals maintained a Mottle body pattern on the small check substrate under all experimental light conditions. The granularity statistics produced curves that were characteristic of Mottle body patterns with peaks in bands 3 and 4 (Fig. 3C,D, black lines).

Small check (evokes Mottle) to large check (evokes Disruptive)

After the substrate was changed to a large check substrate, cuttlefish showed a Disruptive body pattern. The granularity statistics produced a curve with high energy in bands 1 and 2 and a steep negative slope, characteristic of Disruptive body patterns (Fig. 3C, red line). This was confirmed by image data (Fig. 3C, images).

Mauchly's test of sphericity indicated that sphericity was not violated for these data (P=0.441). A repeated measures ANOVA for within-subjects effects was significant ($F_{8,72}$=33.307, P<0.01). *Post hoc* pairwise comparisons of marginal means with a Bonferroni

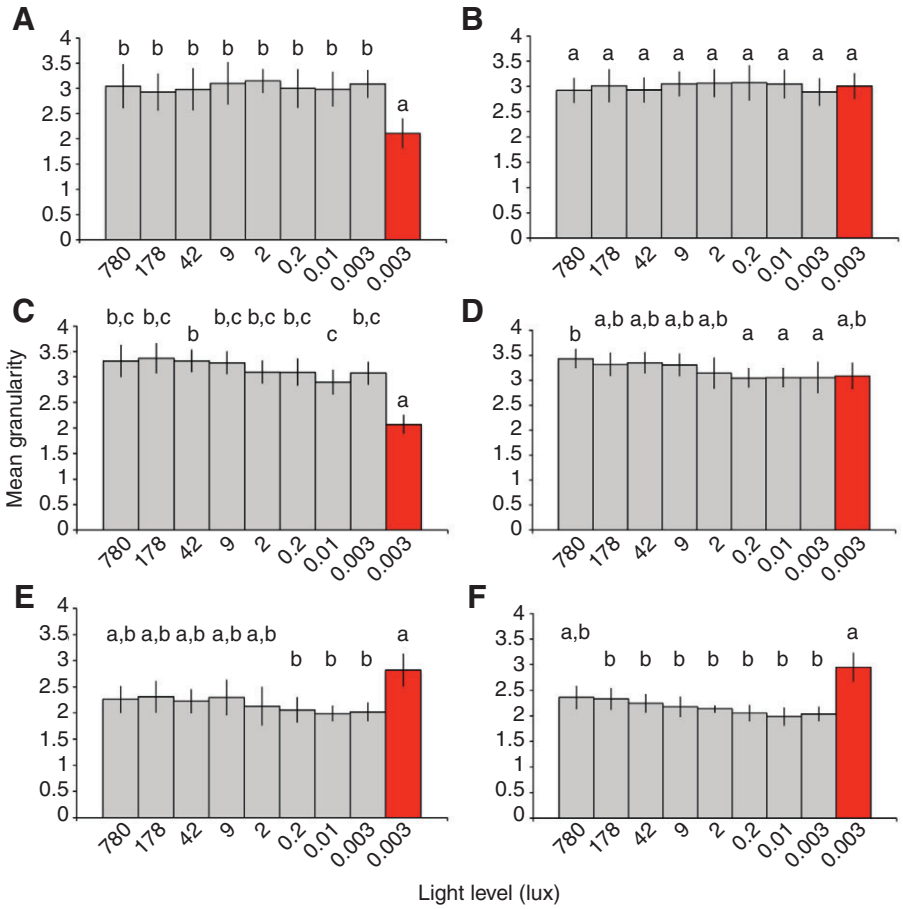

Fig. 4. Results from experiment 2. Plot of the average of the mean granularity statistics for each of eight light levels on the initial substrate (grey bars) and after the substrate was switched (red bars). (A) Grey to large check (Uniform to Disruptive); (B) grey to small check (Uniform to Mottle); (C) small check to large check (Mottle to Disruptive); (D) small check to grey (Mottle to Uniform); (E) large check to grey (Disruptive to Uniform); (F) large check to small check (Disruptive to Mottle). Within each graph, different letters denote significantly different distributions. Error bars are ±s.d.

correction revealed that the mean granularity was not significantly different for animals on the initial substrate, small checks, under eight light levels, with one exception. The mean of the distribution of calculated mean granularity under 42 lux was significantly different from the mean of the distribution of calculated mean granularity under 0.01 lux (P=0.004; Fig. 4C). After the substrate was switched to large checks, the mean granularity of the body pattern on the new substrate was significantly different from the mean granularity of the body patterns on the initial substrate under all light conditions (P<0.001 for each comparison; Fig. 4C).

Small check (evokes Mottle) to grey (evokes Uniform)

After the substrate was changed to a grey substrate, cuttlefish showed little change in body pattern. The granularity statistics produced a curve that was indistinguishable from the curves for Mottle body patterns. Examination of image data revealed body patterns that were very similar before and after the substrate switch (Fig. 3D, images).

Mauchly's test of sphericity indicated that sphericity was violated for these data (P=0.003). Therefore, the Greenhouse–Geisser correction was applied to the repeated measures ANOVA for within-subjects effects. This ANOVA was significant ($F_{3.3,29.8}$=5.209, P=0.004). *Post hoc* pairwise comparisons of marginal means with a Bonferroni correction revealed that the mean granularity was not significantly different for animals on the initial substrate, small checks, under eight light levels, with three exceptions. The mean of the distribution of calculated mean granularity under 780 lux was significantly different from the mean of the distribution of calculated mean granularity under 0.02 lux (P=0.007), 0.01 lux (P=0.041) and 0.003 lux (P=0.044; Fig. 4D). After the substrate was switched to grey, the mean granularity of the body pattern on the new substrate was not significantly different from the mean granularity of the body patterns on the initial substrate under any of the eight light conditions (P>0.305 for each comparison; Fig. 3D, Fig. 4D).

Large check (evokes Disruptive) substrate

In these trials, animals were presented with a large check substrate that was changed to either a small check (evokes Mottle) or to a grey (evokes Uniform) substrate at the dimmest light intensity (0.003 lux). All 10 animals maintained a Disruptive body pattern on the large check substrate under all experimental light conditions. Under all light conditions, the granularity statistics produced curves that were characteristic of Disruptive body patterns with peaks at bands 1 and 2 and a steep negative slope (Fig. 3E,F, black lines).

Large check (evokes Disruptive) to grey (evokes Uniform)

After the substrate was changed to a grey substrate, cuttlefish showed a Uniform body pattern. The granularity statistics produced a curve that showed low energy in all six bands, characteristic of Uniform body patterns (Fig. 3E, red line). This was confirmed by image data (Fig. 3E, images).

Mauchly's test of sphericity indicated that sphericity was violated for these data (P<0.001). Therefore, the Greenhouse–Geisser correction was applied to the repeated measures ANOVA for within-subjects effects. This ANOVA was significant ($F_{2.9,26.2}$=10.219, P<0.001). *Post hoc* pairwise comparisons of marginal means with a Bonferroni correction revealed that the mean granularity was not significantly different for animals on the initial substrate, large checks, under eight light levels. After the substrate was switched to grey, the mean granularity of the body pattern on the new substrate was significantly different from the initial substrate under three lighting conditions: 0.2 lux (P=0.001), 0.01 lux (P=0.001) and 0.003 lux (P=0.001; Fig. 3E; Fig. 4E).

Large check (evokes Disruptive) to small check (evokes Mottle)

After the substrate was changed to a small check substrate, cuttlefish showed a Mottle body pattern. The granularity statistics produced a curve with a peak in band 3, characteristic of Mottle body patterns (Fig. 3F, red line). This was confirmed by image data (Fig. 3F, images).

Mauchly's test of sphericity indicated that sphericity was not violated for these data (P=0.119). A repeated measures ANOVA for within-subjects effects was significant ($F_{8,72}$=23.456, P<0.001). *Post hoc* pairwise comparisons of marginal means with a Bonferroni correction revealed that the mean granularity was not significantly different for animals on the initial substrate, large checks, under eight light levels. After the substrate was switched to small checks, the mean granularity of the body pattern on the new substrate was significantly different from the mean granularity of the body patterns on the initial substrate under all light conditions (P<0.023 for all comparisons; Fig. 4F), except one, 780 lux (P=0.103).

DISCUSSION

Here, we present the first evidence from experimental data that cuttlefish *S. officinalis* continue to camouflage themselves under light conditions comparable to night-time levels and that this behaviour is adaptive, i.e. the camouflage patterns are changeable as they are during daylight and crepuscular periods (e.g. Allen et al., 2010; Chiao and Hanlon, 2001; Kelman et al., 2007; Mäthger et al., 2006).

Although there are no data available on the energy requirements of adaptive camouflage body patterning, it is likely that cuttlefish would save energy by not expressing camouflage body patterns if it were not necessary. Because they assess their visual environment and continue to camouflage in very dim light, we can speculate that these animals might use this behaviour (1) to avoid predators with keen night vision and (2) to increase their own hunting success at night (cuttlefish are carnivorous predators). In particular, *S. officinalis* has been suggested to be active day and night (Denton and Gilpin-Brown, 1961; Mark et al., 2007; Watanuki et al., 2000). For other species that are less active at night, such as *S. apama* (Aitken and O'Dor, 2005), it is likely that nocturnal camouflage behaviour is an anti-predator tactic (Hanlon et al., 2007). Cephalopods as a group show a range of nocturnal and diurnal activity patterns, and we hypothesize that nocturnal or deep-sea cephalopods that dwell in very little light use their adaptable body patterning to avoid predators and/or increase their hunting success.

The visual abilities of nocturnal and deep-sea animals are remarkable, ranging from high sensitivity to dim light by a variety of means [such as large eyes, pupils and photoreceptors, tapeta to aid photon capture, neural summation, large visual receptive field, specialized foveas, etc. (for reviews, see Denton, 1990; Warrant, 2004; Warrant, 2008; Warrant and Locket, 2004)] to excellent motion detection even though only a few photons are available for vision (Krapp and Hengstenberg, 1996; Warrant, 2004; Warrant, 2008). Despite the fact that many animals sacrifice colour vision for increased sensitivity to low light, colour vision has been found in nocturnal hawkmoths and geckoes, as well as in some deep-sea fish and cephalopods, which opens the possibility that colour vision under low-light conditions might not be as unusual as previously thought (Denton and Locket, 1989; Kelber et al., 2002; Partridge et al., 1988; Roth and Kelber, 2004). Other remarkable adaptations include the use of polarized moon and night-time sky light for navigation in some insects (Dacke et al., 2003a; Dacke et al., 2003b) and the body curling

behaviour of mesopelagic animals in response to threatening stimuli in dim light (Robison, 1999). Considering how successful these and many other nocturnal and deep-sea animals are, the physiological and morphological adaptations that go alongside their low-light active life styles are perhaps not surprising (although certainly impressive).

In our experiments, there was little change in camouflage body pattern when the substrate was changed from small check (evokes a Mottle body pattern) to uniform grey (evokes a Uniform body pattern), and *vice versa* (Fig. 3B,D, images and statistical analyses). As there is a trade-off between resolution and sensitivity (Land and Nilsson, 2002), one potential reason for this might be that the low number of photons available at 0.003 lux affects the visual acuity of the cuttlefish and the animal cannot resolve fine details, such as those of a small check substrate, even though the contrast between the black and white checks is high (approximately 80%). A high-fidelity match to the background might, however, not be necessary. Our knowledge of the visual abilities of cephalopod predators is still patchy (e.g. Losey et al., 2003; Marshall et al., 2003), and because predator visual acuity is also compromised at such low photon levels (possibly even to the same degree), general background matching at night could be a satisfactory camouflage mechanism.

Whether or not cuttlefish can discern a high-contrasting small-scale background (such as a substrate with check sizes approximately 5% of the animal's White square component) at a light intensity of 0.003 lux remains to be established. Even though cephalopod camouflage is a visually driven behaviour and we can make inferences about the relationship between visual cue and camouflage body pattern, we cannot say with absolute certainty that because a cuttlefish does not respond to a particular visual cue with a body pattern, the animal cannot discern that particular cue. A different approach to measuring spatial resolution, such as an experiment taking advantage of the optomotor response, might answer this question (e.g. Groeger et al., 2005; Messenger, 1970).

In experiment 2, we lowered the light intensity to 0.003 lux, which is equivalent to starlight levels on land. Light intensity in the sea drops to levels well below this value, depending on water type, depth and time of day (Denton, 1990; Jerlov, 1976; Warrant and Locket, 2004). Future work might expand on our study and scrutinize some of the other 700+ cephalopod species that occupy the world's oceans, examining how they have adapted their various life styles to low-light environments.

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