The LINNEAN SOCIETY of London BIOLOGICAL Journal of the Linnean Society

Biological Journal of the Linnean Society, 2012, **105**, 359–368. With 4 figures

Spectral composition and visual foraging in the three-spined stickleback (Gasterosteidae: *Gasterosteus aculeatus* L.): elucidating the role of ultraviolet wavelengths

INGOLF P. RICK*, DOMINIQUE BLOEMKER and THEO C. M. BAKKER

Institute for Evolutionary Biology and Ecology, University of Bonn, An der Immenburg 1, 53121 Bonn, Germany

Received 27 May 2011; revised 1 August 2011; accepted for publication 1 August 2011

Visual signalling can be affected by both the intensity and spectral distribution of environmental light. In shallow aquatic habitats, the spectral range available for visually mediated behaviour, such as foraging, can reach from ultraviolet (UV) to long wavelengths in the human visible range. However, the relative importance of different wavebands in foraging behaviour is generally unknown. Here, we test how the spectral composition of ambient light influences the behaviour of three-spined sticklebacks (*Gasterosteus aculeatus*) when foraging for live cladoceran *Daphnia magna*. Although paying particular attention to the UV waveband, we measured the foraging preferences of sticklebacks for prey presented under four different spectral conditions. These conditions selectively removed UV (UV–), short-wave (SW–), mid-wave (MW–) or long-wave (LW–) light from the entire spectrum. The absence of UV and long wavelengths strongly reduced prey attractiveness for *G. aculeatus* compared with conditions without short-wave and mid-wave light. To control for potential light habitat preferences in the main experiment, we conducted a further choice experiment without prey stimuli. Fish in these trials did not discriminate significantly between the different spectral conditions. When comparing both experiments, it was observed that, although filter preferences for MW– and LW– conditions were virtually consistent, they differed at shorter wavelengths, with a reduced preference for UV– conditions and, at the same time, an increased preference for SW– conditions in the presence of prey. Thus, prey choice seems to be strongly affected by visual information at the short-wave end of the spectrum. The foraging preferences were also mirrored by the chromatic contrast values between prey and the experimental background, as calculated for each lighting condition using a series of physiological models on stickleback perception. *Biological Journal of the Linnean Society*, 2012, **105**, 359–368.

ADDITIONAL KEYWORDS: color vision – foraging behaviour – light environment – predator–prey interactions.

INTRODUCTION

Environmental lighting conditions are of crucial importance in visually mediated behaviour in fish. For instance, the intensity of ambient light has been shown to affect intraspecific communication (e.g. Endler, 1987) as well as foraging behaviour (Guthrie & Muntz, 1993; Hart & Gill, 1994). In a foraging context, variation in light intensity can influence prey selection (Confer *et al.*, 1978) and consumption rates (Connaughton, Epifanio & Thomas, 1994; Macy, Sutherland & Durbin, 1998; Ryer & Olla, 1999). However, the efficiency of visual prey detection is not dependent solely on differences in light intensity, but is also influenced by variation in the spectral composition of light, which, in turn, is strongly affected by the relative abundance of dissolved and suspended matter in aquatic habitats (Lythgoe, 1972). Consequently, the diversity in spectral distribution in shallow waters can be higher than that found in

*Corresponding author. E-mail: irick@evolution.uni-bonn.de

terrestrial habitats (Endler, 1997). Furthermore, the spectral range of some shallow aquatic habitats can be very broad (Baker & Smith, 1982), ranging from very short wavelengths (ultraviolet, UV: 300–400 nm) to longer wavelengths in the human visible waveband (400–700 nm). Numerous fish species living in surface waters are able to perceive light of a broader wavelength range, including UV (Losey *et al*., 1999), and many fish possess UV colour components that are often involved in intraspecific interactions, such as mate choice (e.g. Kodric-Brown & Johnson, 2002; Rick & Bakker, 2008a).

The visibility of visual signals depends on both the intensity and visual contrast that is generated against the natural background (Lythgoe, 1968). Consequently, in visually hunting predators, foraging can depend on the prey's contrast with the background, which can be extended to shorter wavelengths in species with UV sensitivity. UV vision has been found to be important in foraging, especially in terrestrial vertebrates and invertebrates. For example, common kestrels (*Falco tinnunculus* L.) use UV-reflective vole scent marks when hunting for prey (Viitala *et al*., 1995), and foraging redwings (*Turdus iliacus* L.) are attracted to UV-reflective bilberries (Siitari, Honkavaara & Viitala, 1999). In fish, evidence that UV light contributes to foraging performance is rather scarce. However, in larval fish of some species, UV is thought to be involved in the location and capture of zooplankton prey (Loew *et al*., 1993; Browman, Novales-Flamarique & Hawryshyn, 1994).

Although depending largely on the spectral range of the natural light environment, visual sensitivity in the three-spined stickleback underlies substantial intraspecific variation, ranging from populations inhabiting red-shifted habitats, in which visual perception is limited to longer wavelengths (Cronly-Dillon & Sharma, 1968; McDonald & Hawryshyn, 1995), to clear water populations, in which vision is also extended into the UV as a result of a fourth UV-sensitive cone receptor (Rowe *et al*., 2004). With regard to the latter case, reproductively active male and female sticklebacks in some populations possess pronounced UV-reflecting skin regions (Rick, Modarressie & Bakker, 2004; Rowe *et al*., 2004) and UV signals provide important cues in visual communication (Modarressie, Rick & Bakker, 2006; Rick, Modarressie & Bakker, 2006; Rick & Bakker, 2008b, c).

In this study, although focusing on the potential importance of UV wavelengths compared with other parts of the spectrum, we explored how differences in spectral composition affect foraging performance in sticklebacks. A choice experiment was conducted in which live cladoceran *Daphnia magna* (Straus) were presented as prey individuals under four different spectral conditions, with each selectively removing blocks from the entire UV–visible range between 300 and 700 nm. We determined how the manipulation of the spectral content of the prey's light environment influences the foraging choice of sticklebacks, and how this can be classified when taking into account the stickleback's perception of chromatic and achromatic contrast between prey and the visual background. In the absence of data on spectral sensitivity for fish from our study population, we modelled visual perception for a range of hypothetical stickleback cone contributions.

MATERIAL AND METHODS

Experimental subjects

Samples of sticklebacks were collected in September 2009 from a shallow pond near the Institute for Evolutionary Biology and Ecology in Bonn, Germany (50°73′N, 7°07′E). After being housed in a single group for 3 months in an outside stock tank (700 l), individual fish measuring 5.0 ± 0.2 cm in standard length were isolated into single aquaria ($30 \times 20 \times 20$ cm^3, 12 l) with internal filter aeration in the laboratory. The non-reproductive fish were maintained at 17 ± 2 °C under an 8 h : 16 h light : dark illumination cycle provided by fluorescent tubes simulating natural skylight conditions including UV (True Light, 'Natural Daylight' 5500, 36 W, 1200 mm). Fish were individually housed for 3 weeks before being used in the experiment and were fed *ad libitum* with frozen red chironomid larvae once daily. Feeding was stopped 2 days before the experiment to ensure that test individuals were hungry during the trials. As prey organisms in the experiment, we used laboratory-bred *Daphnia magna*, which belong to the natural prey spectrum of sticklebacks.

Experimental set-up

Foraging preferences were tested using a cross-shaped choice chamber which has been described in detail elsewhere (Rick & Bakker, 2008a), but with minor changes in the present study (Fig. 1). One stickleback in the central arena was given the choice between four groups of 10 *Daphnia* each, which were presented in small chambers ($8 \times 20 \times 2.5$ cm^3) in the stimulus tanks. The test fish was able to view each stimulus shoal through a cut-out window (8×10 cm^2). A marked zone (9×9 cm^2) in front of each stimulus chamber served as the attention zone. The central arena and the four stimulus compartments were filled with water up to a height of 10 cm. Prey appearance was manipulated by placing four different colour filters between the test fish and prey shoals. These filters removed discrete wave bands and are referred

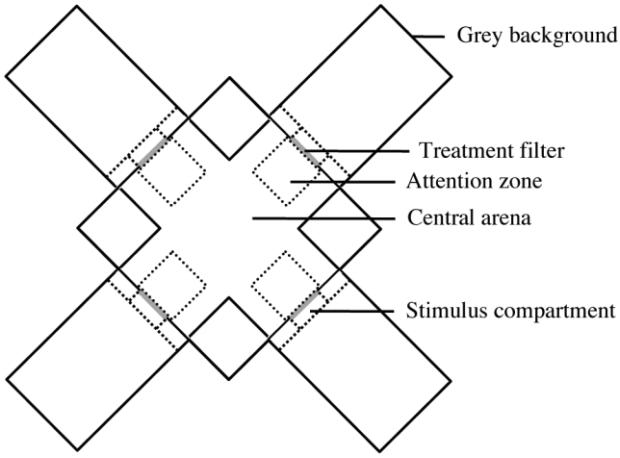

Figure 1. Experimental apparatus used to test stickleback foraging preferences. The set-up consists of a central choice arena and four attention zones (dotted lines in central area) from where the test fish was able to view the prey shoals in the stimulus compartments through the four different treatment filters (grey lines). The side walls and the back wall of each stimulus tank (i.e. the visual background), as well as the inner walls of the central arena, consisted of opaque abrased grey plastic partitions (full lines), whereas the stimulus compartments were surrounded by ultraviolet (UV)-transparent plexiglas (dotted lines around stimulus compartments).

to as UV-blocking (UV–), short-wave-blocking (SW–), mid-wave-blocking (MW–) and long-wave-blocking (LW–) (Lee Filter No. 229, Rosco Supergel Filters 14, 339 and 73, respectively; for transmission curves, see Fig. 2A). The set-up was illuminated by four fluorescent tubes (True Light, 'Natural Daylight' 5500, 36 W, 1200 mm) which were arranged as described in Rick & Bakker (2008a). The brightness transmission (300–700 nm) of the four treatment filters was measured spectrophotometrically (see below) and adjusted between the four filters using multiple layers of filter material, so that the exact ratio of quantal flux for the four light treatments (UV– : SW– : MW– : LW–) was 1.19 : 1.13 : 1.15 : 1.00.

Experimental procedure

For the experimental trials, 16 test fish were divided into four groups of four fish. Individuals within each group were assigned the same four stimulus shoals, but viewed them behind exchanged filters following a randomized design. Four trials were conducted per day, with each trial consisting of a 10-min acclimatization period with opaque partitions (20×20 cm^2) placed between the test fish and prey, an observation phase with lifted opaque partitions, which lasted until the fish had frequented all four attention zones (maximum period of 10 min), and a 10-min test phase in which fish behaviour was recorded. Prey choice was measured as the time spent by the test fish (entire body) in the four attention zones during the test phase.

Control experiment

To control for general preferences for the four light environments, independent of foraging behaviour, a supplemental control experiment was conducted with 17 additional test fish. Experimental trials were performed analogous to the foraging experiment, except that prey stimuli were not present.

All trials were recorded from above with a webcam, and videos were analysed blindly without knowledge of the trial type and filter positions.

Visual modelling

We questioned how differences in the reflectance of prey and visual background between the colour treatments might translate into differences in the relative response of stickleback visual pigments and thus affect the chromatic and achromatic background contrast of prey as perceived by the stickleback's eye. Therefore, we included reflectance data of prey and the experimental background, transmission data of the optical filters used, data on downwelling irradiance measured in the experimental set-up, and stickleback cone pigment absorbance spectra, as well as lens transmission properties, in a visual model.

Standardized reflectance scans of five *Daphnia* were recorded with a spectrophotometer (Avantes AVS-USB2000) connected to a deuterium–halogen light source (Avantes DH-2000) for illumination. A bifurcated, 200-μm, small-tip fibre-optic with unidirectional illumination and recording was held at a 90° angle to the body surface. For measurements, *Daphnia* were removed from their stock tank and placed on a piece of black fabric in order to reduce light transmission and scattering caused by translucent parts of the body. Scans were collected from the dorsal carapace region. Reflectance was measured relative to a 98 % Spectralon white standard over the range 300–700 nm at about 0.5-nm resolution in wavelength (Fig. 2B). Data were recorded with Spectrawin 5.1 (Avantes) and imported into Microsoft Excel. Fifteen measurements were made in succession, averaged for the sample region without changing the probe contact. Using the same measurement protocol, we measured the spectral reflectance from the visual background in the stimulus compartments, which consisted of abrased grey plastic partitions (Fig. 2B). The spectral transmission of the four treatment filters was determined by measuring the reflectance relative to the white standard, with the

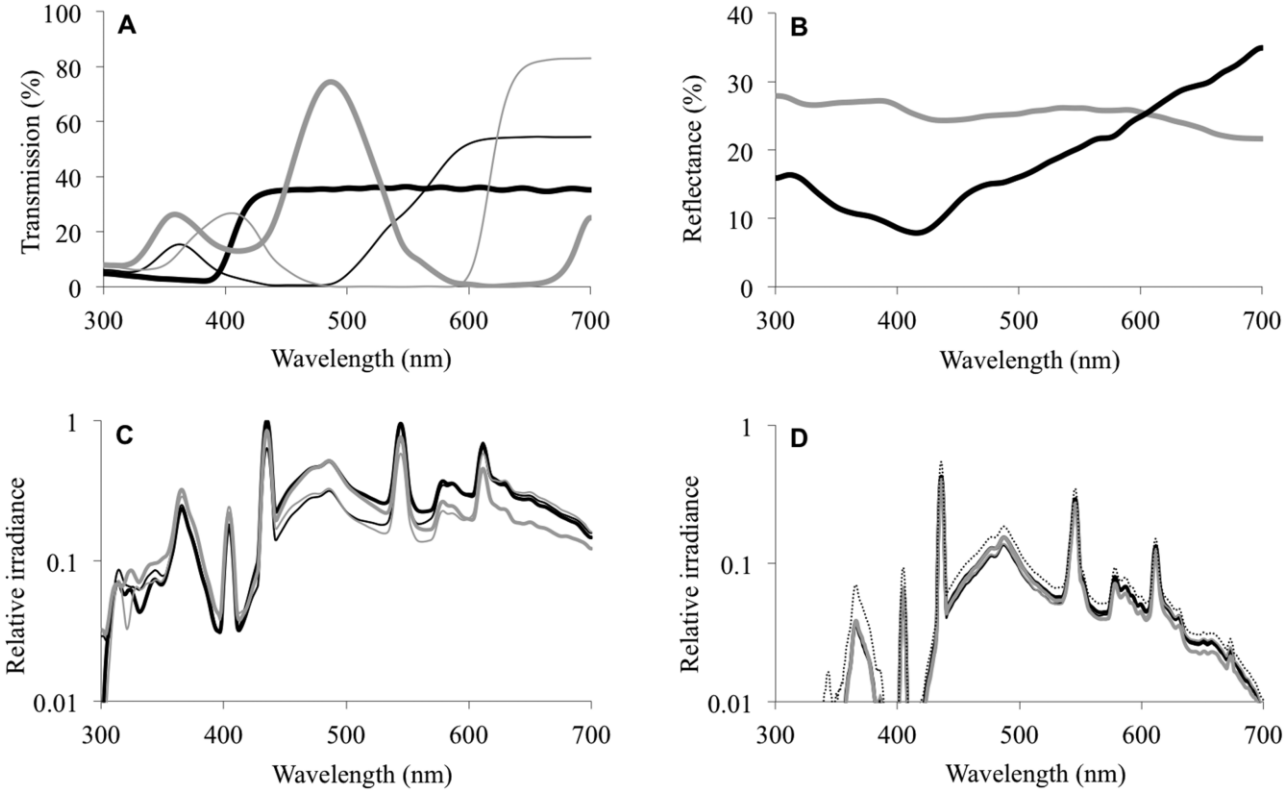

Figure 2. A, Spectral transmission of the ultraviolet-blocking (UV–, thick black line), short-wave-blocking (SW–, thin black line), mid-wave-blocking (MW–, thin grey line) and long-wave-blocking (LW–, thick grey line) treatment filters. B, Mean reflectance of the dorsal carapace region of five *Daphnia magna* (black line) and the visual background (grey line) used in the experimental set-up. C, Relative downwelling irradiance measured in the UV– (thick black line), SW– (thin black line), MW– (thin grey line) and LW– (thick grey line) stimulus chambers. A logarithmic *y* axis is used because of the large emission peaks produced by the fluorescent lighting. As downwelling irradiance was measured, the spectra display a combination of unfiltered light from above and through the back side, as well as filtered light collected through the front side of each chamber. D, Relative sidewelling (dotted line) and upwelling irradiance collected for the UV– (thick black line), SW– (thin black line), MW– (thin grey line) and LW– (thick grey line) conditions. The spectra of the sidewelling irradiance for the different lighting conditions strongly overlap each other, so that only an averaged spectrum is plotted for simplicity. Relative irradiance was calculated proportional to the highest peak intensity attained when considering all lines of sight.

reflection probe attached perpendicular to the filter or, where required, to multiple layers of filter material located on the white reference (Fig. 2A).

We measured irradiance (light collected from a whole hemisphere) with the probe pointing upwards for downwelling light (Fig. 2C). For comparison, we also collected upwelling irradiance with the probe pointing downwards, as well as sidewelling irradiance by holding it parallel to the ground and pointing away from the test fish arena towards the stimulus background (Fig. 2D). All measurements were conducted with an Avantes CC-UV/VIS cosine corrector placed in the stimulus compartments next to the relevant optical filter. Irradiance was calibrated against an Avantes NIST traceable application standard.

The coloration of *Daphnia* and of the experimental background were quantified as perceived through the stickleback visual system. To do this, published cone absorbance maxima (Rowe *et al*., 2004) and parameters provided in Govardovskii *et al*. (2000) were used to determine spectral sensitivity functions for a hypothetical tetrachromatic model of stickleback vision including UV-sensitive (UVS), short-wave-sensitive (SWS), medium-wave-sensitive (MWS) and long-wave-sensitive (LWS) cone receptors. We also modelled the visual responses for different scenarios involving three cone types (SWS, MWS, LWS) and two cone types (MWS, LWS). For reasons of clarity, these calculations were all based on cone absorbance maxima given by Rowe *et al*. (2004). Absolute cone

catches Q for each receptor type i were derived using the equation:

$$Q_i = \int_{\lambda 300}^{\lambda 700} R_i(\lambda) S(\lambda) I(\lambda) \mathrm{d}\lambda$$

where λ is the wavelength, R_i represents the spectral sensitivity of cone type i, $S(\lambda)$ is the mean reflectance of the visual stimuli (*Daphnia*, grey background) and $I(\lambda)$ corresponds to the spectrum of downwelling irradiance in the stimulus compartments (Fig. 2C), summed across wavelengths between 300 and 700 nm (Endler & Mielke, 2005). The spectral sensitivity R_i of each cone type was calculated as:

$$R_i = P_i(\lambda) L(\lambda) T(\lambda)$$

where P_i denotes the normalized absorbance of cone type i, $L(\lambda)$ is the lens transmission of non-reproductive fish from the same population (I. P. Rick, unpubl. data) and $T(\lambda)$ is the transmission of the vertically mounted optical filters (UV–, SW–, MW– or LW–) located in the light path between the test fish and the prey shoals. As prey perception of sticklebacks in our experimental set-up was restricted to short signalling distances and occurred at low water depths, the absorption and scatter of water were not included in our calculations.

We used a photoreceptor noise-limited colour discrimination model (Vorobyev & Osorio, 1998) to determine the relative contrast Δf for each receptor type i under each of the four light conditions as the natural logarithm of the absolute quantum catches for prey coloration (P), which is normalized against the visual background (B):

$$\Delta f_i = \ln\left(\frac{Q_{i\mathrm{P}}}{Q_{i\mathrm{B}}}\right)$$

The perceptual distance ΔS, which describes the chromatic contrast between prey and the experimental background, was calculated for a tetrachromatic visual system as:

$$\Delta S^2 = \frac{\left[\begin{array}{l}(e_1e_2)^2(\Delta f_4 - \Delta f_3)^2 + (e_1e_3)^2(\Delta f_4 - \Delta f_2)^2 + \\ (e_1e_4)^2(\Delta f_2 - \Delta f_3)^2 + (e_2e_3)^2(\Delta f_4 - \Delta f_1)^2 + \\ (e_2e_4)^2(\Delta f_3 - \Delta f_1)^2 (e_3e_4)^2(\Delta f_2 - \Delta f_1)^2\end{array}\right]}{\left[(e_1e_2e_3)^2 + (e_1e_2e_4)^2 + (e_1e_3e_4)^2 + (e_2e_3e_4)^2\right]}$$

for a trichromatic visual system as:

$$\Delta S^2 = \frac{\left[e_1^2(\Delta f_3 - \Delta f_2)^2 + e_2^2(\Delta f_3 - \Delta f_1)^2 + e_3^2(\Delta f_1 - \Delta f_2)^2\right]}{\left[(e_1e_2)^2 + (e_1e_3)^2 + (e_2e_3)^2\right]}$$

and for a dichromatic visual system as:

$$\Delta S^2 = \frac{(\Delta f_1 - \Delta f_2)^2}{(e_1^2 + e_2^2)}$$

Colour signals that appear similar to a receiver result in small ΔS values, whereas large ΔS values correspond to highly contrasting signals (Vorobyev & Osorio, 1998). We considered the signalling noise e_i for each receptor type to depend only on neural noise (Håstad, Victorsson & Ödeen, 2005):

$$e_i = \sqrt{\frac{\omega}{\eta_i}}$$

where ω is the Weber fraction and η_i is the relative density of the receptor type i in the retina. As behavioural data on sensitivity thresholds and information on cone proportions of sticklebacks are lacking, we chose a Weber fraction value of 0.05 as a conservative measure of visual performance (Vorobyev *et al*., 1998). For tetrachromatic models, we used hypothetical cone ratios of 1 : 1 : 2 : 2 and 1 : 1 : 1 : 1 for the UVS, SWS, MWS and LWS cones. Furthermore, we used cone ratios of 1 : 2 : 2 and 1 : 1 : 1 (SWS : MWS : LWS) for trichromatic models and a ratio of 1 : 1 (MWS : LWS) for a dichromatic model. Differences in cone ratios did not lead to differences in the qualitative results for the respective models.

In addition to spectral variation, sticklebacks may have based their foraging behaviour during the experimental trials on differences in brightness contrast between prey and the experimental background. We assumed that the double cones are responsible for luminance detection (Hart *et al*., 2000) and calculated the achromatic contrast ΔQ between the *Daphnia* prey and the background for each light condition by dividing the summed cone catches of the MWS and LWS cones for the prey stimulus by the summed mid-wave and long-wave cone excitation for the background.

As more detailed psychophysical data on stickleback visual perception are lacking, our models are only approximate. However, they should be sufficient to illustrate differences in the chromatic and achromatic background contrast of prey between light habitats that differ considerably in spectral content with reference to the visual system under study.

Statistics

For analysis, we used the R 2.11.0 software package (R-Development-Core-Team, 2010). Data for the foraging experiment were normally distributed accord-

ing to Shapiro–Wilk tests, whereas data for the control experiment were square-root transformed to reach normality. Linear mixed-effects models ('lme', package 'nlme'; Pinheiro *et al.*, 2009) were fitted to measure differences in preference between the four filter treatments. The relative attention time (time spent in one attention zone/time spent in all attention zones) was used as dependent variable, the treatment filter was the fixed factor and test fish identity was the random factor. A likelihood-ratio test (LRT) assessed whether the removal of the fixed factor caused a significant decrease in model fitting. Hence, degrees of freedom always differ by unity. The reported P value of the model refers to the increase in deviance when the fixed factor was removed (F statistics). When comparing filter preferences between both the foraging and control experiment, an interaction term between the treatment filter and experiment type was included in the model. All P values given were based on two-tailed tests.

RESULTS

Foraging experiment

In the foraging trials, sticklebacks showed distinct prey choice behaviour over the duration of each trial, as they frequently detected, approached and tried to attack the *Daphnia* behind all four treatment filters. *Gasterosteus aculeatus* discriminated significantly between the four filter treatments (full model LRT: $\chi^2 = 10.19$, d.f. = 1, $P = 0.017$; Fig. 3A). *Post-hoc* tests revealed that the test fish spent a significantly smaller amount of time in front of prey presented behind the UV– relative to the SW– filter (LRT: $\chi^2 = 4.56$, $P = 0.033$), and a significantly shorter time in front of the UV– relative to the MW– prey shoals (LRT: $\chi^2 = 8.92$, $P = 0.003$). Furthermore, the fish significantly preferred prey stimuli viewed under MW– conditions relative to stimuli shown under LW– conditions (LRT: $\chi^2 = 5.41$, $P = 0.020$) and tended to prefer prey under SW– conditions relative to LW– conditions (LRT: $\chi^2 = 2.85$, $P = 0.091$). There was no significant difference in attention time between SW– and MW– conditions ($\chi^2 = 0.10$, $P = 0.753$) or between UV– and LW– conditions ($\chi^2 = 0.24$, $P = 0.627$).

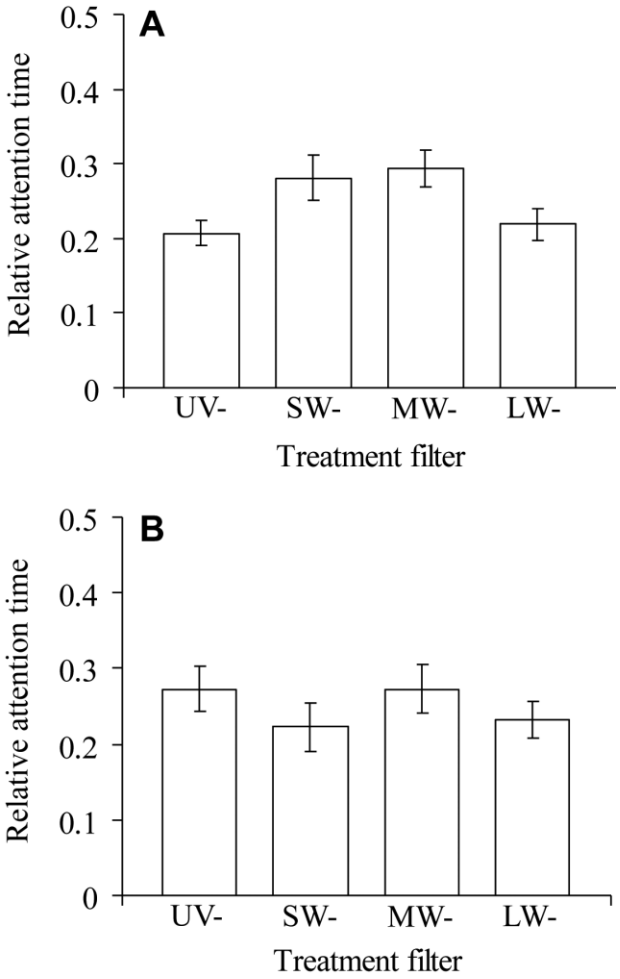

Figure 3. A, Mean relative attention time ± SEM spent by 16 sticklebacks in front of *Daphnia* shoals presented behind ultraviolet-blocking (UV–), short-wave-blocking (SW–), mid-wave-blocking (MW–) and long-wave-blocking (LW–) filters during the foraging trials. B, Mean relative attention time ± SEM spent by 15 sticklebacks in front of empty chambers presented behind UV–, SW–, MW– and LW– filters during the control trials.

Control experiment

In the control experiment, two of the 17 trials had to be discarded as the test fish did not frequent all four attention zones during the acclimatization period. Fish in the control trials showed no significant discrimination between the four light habitats (full model LRT: $\chi^2 = 2.812$, $P = 0.422$; Fig. 3B). When comparing both experiments, the preferences of fish in the foraging trials were not significantly different from the light habitat preferences in the control trials, although a tendency was apparent (full model LRT: interaction between filter treatment and experiment type, $\chi^2 = 6.685$, $P = 0.083$).

Visual modelling

The reflectance spectra of *D. magna* revealed higher reflectance values at UV wavelengths relative to short wavelengths, followed by an increase in reflectance towards longer wavelengths (Fig. 2B). The diffuse reflectance at UV wavelengths was low, suggesting that we measured the true reflectance of the carapace rather than an artefact of scattered short-wave light.

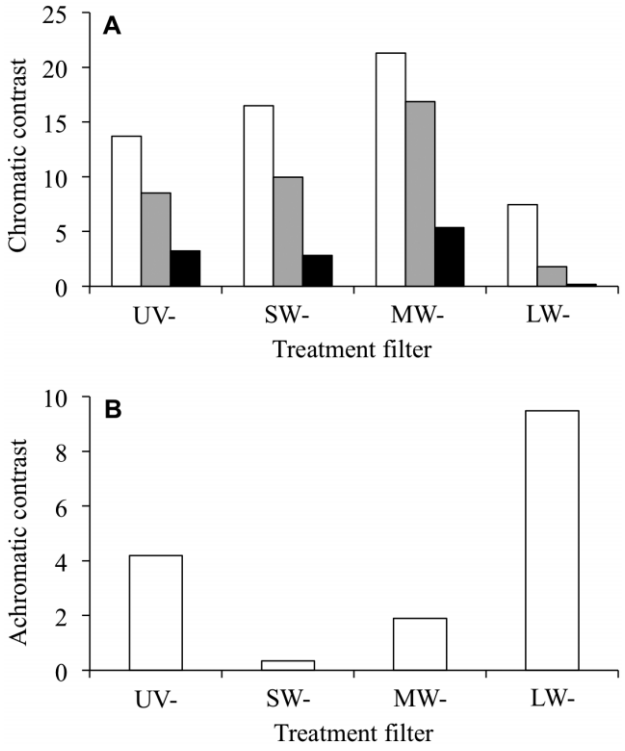

Figure 4. A, Chromatic contrast of *Daphnia* prey against the experimental background for the ultraviolet-blocking (UV–), short-wave-blocking (SW–), mid-wave-blocking (MW–) and long-wave-blocking (LW–) conditions for hypothetical tetrachromatic (white bars), trichromatic (grey bars) and dichromatic (black bars) visual models. Contrast calculations were based on a cone ratio of 1 : 1 : 2 : 2 [UV-sensitive (UVS) : short-wave-sensitive (SWS) : medium-wave-sensitive (MWS) : long-wave-sensitive (LWS)] for the tetrachromatic model, 1 : 2 : 2 (SWS : MWS : LWS) for the trichromatic model and 1 : 1 (MWS : LWS) for the dichromatic model. B, Achromatic contrast of prey against the visual background for the four experimental lighting conditions.

The chromatic contrast ΔS of *Daphnia* against the experimental background as seen through the stickleback's eye was lowest for LW– conditions, followed by UV– and SW– conditions, and highest for the MW– treatment filter, when modelled for both a tetrachromatic and trichromatic colour space (Fig. 4A). In comparison, for the dichromatic model, overall chromatic contrast values were notably lower. Here, LW– conditions generated the lowest contrast, followed by SW– conditions, whereas UV– conditions revealed a slightly higher contrast value and MW– conditions the highest contrast (Fig. 4A).

The achromatic background contrast of prey was lowest for the most attractive SW– and MW– conditions, followed by the UV– condition, and highest for the LW– condition (Fig. 4B).

DISCUSSION

The outcome of the choice experiment shows that differences in the spectral composition of ambient light affect stickleback foraging behaviour. The removal of UV (UV–) and red wavelengths (LW–) significantly reduced prey attractiveness, whereas the absence of short-wave (SW–) and mid-wave (MW–) light had a comparably lower effect on prey choice. We thus demonstrated, for the first time, that UV is of relative importance in a foraging task, which is contrary to previous studies on zebra finches (*Taeniopygia guttata* Vieilott) (Maddocks, Church & Cuthill, 2001) and guppies (*Poecilia reticulata* Peters) (White *et al*., 2005), where a similar approach of blocking certain wavebands did not reveal a significant reduction in foraging behaviour under UV-absent conditions relative to the removal of other wavelengths. Furthermore, in a series of foraging experiments on sticklebacks, no significant difference between UV-present and UV-absent conditions was found with regard to both feeding preferences and foraging efficiency (Modarressie & Bakker, 2007). However, the authors showed that sticklebacks attacked prey faster when the visual background lacked UV reflection relative to a UV-reflecting background. The discrepancy between this finding and the outcome of the present study may be explained by differences in the experimental design, as prey individuals in the former study were presented in front of a background with a much stronger overall reflectance than in the present study. In addition, the fact that only the UV content of the visual background was removed in the former study is different from the present study, in which illumination lacked UV and thus, in addition, the visual appearance of prey.

The filter preference of fish in the control trials was not affected significantly by the light habitat alone and was not significantly different from the filter preference found for the foraging experiment. However, filter preferences tended to differ between the two experiments, suggesting that behavioural decisions in the foraging trials were more probably based on prey perception. This was more pronounced for shorter wavelengths, with lower values for UV– and higher values for SW– conditions in the presence of prey, and vice versa without prey stimuli. By contrast, preferences for MW– and LW– conditions were rather similar in both the foraging and control trials, so that prey choice seems to be strongly affected by short-wave visual information. Moreover, it is possible that the sample size in the control experiment during which test fish were not stimulated by prey was too small to draw stronger conclusions.

The results of the foraging experiment are in accordance with studies on avian foraging behaviour,

which demonstrated that UV cues are used for prey detection, discrimination and recognition (Viitala *et al*., 1995; Church *et al*., 1998; Siitari *et al*., 1999). In some UV-sensitive fish species, the UV waveband is assumed to be involved in foraging by improving the overall detection of short-wave-scattering planktivorous prey species or by enhancing prey contrast (Loew *et al*., 1993; Browman, Novales-Flamarique & Hawryshyn, 1994). Zooplankters, such as *Daphnia*, show UV-absorbing body parts which may contrast with the UV of the downwelling spacelight, particularly when viewed in the upward direction (Novales-Flamarique & Browman, 2001; White *et al*., 2005). In addition, our reflectance measurements revealed that *D. magna*, which were used as prey items in the present study, also possess body parts of higher UV reflectance (Leech & Johnsen, 2006), which could also contribute to prey contrast with a visual background. We thus used a physiological model of stickleback perception to calculate the chromatic and achromatic contrast of *Daphnia* when horizontally observed against the grey experimental background behind the four different spectral conditions used in the choice trials. We found our measurements of chromatic contrast to be roughly consistent with the preferences found in the choice experiment, with lower background contrasts under the less attractive UV– and LW– conditions relative to a higher contrast value, particularly for the preferred MW– condition. Calculations based on visual systems incorporating three (SWS, MWS, LWS) or four (UVS, SWS, MWS, LWS) cone types led to comparable contrast values, suggesting that the presence of an additional UV cone type is not required to explain our results. In comparison, when modelling data for only two cone types (MWS, LWS), the chromatic background contrast of prey did not match well with the prey preferences in the foraging experiment. In summary, in order to make safer assumptions, more detailed data for the spectral sensitivity of fish from our study population, as well as for the environmental lighting conditions, are needed.

When comparing the achromatic background contrast of prey with choice behaviour in the foraging experiment, it became apparent that the preferred conditions (SW–, MW–) generated a lower achromatic contrast, and vice versa. One may argue that the fish spent more time in front of filters blocking the short-wave and mid-wave spectral parts because of a prolonged detection and investigation of prey based on luminance cues under these conditions. However, studies on the preferences of visually foraging planktivorous fish indicate that more conspicuous prey in terms of colour, contrast, size and movement should evoke a stronger predator response (Endler, 1978). Consequently, the time spent by test fish in front of prey is more likely to be a proxy of prey preference elicited by chromatic cues rather than prey assessment based on luminance perception. However, as brightness can have profound effects on visual foraging in fish (e.g. Confer *et al*., 1978), further work assessing the relative contribution of chromatic and achromatic perception in stickleback visual foraging is necessary in order to draw stronger conclusions.

Our results corroborate the view that visual perception in the UV and long-wave spectral regions contributes strongly to stickleback foraging behaviour, and that it is potentially based on an enhanced chromatic background contrast of prey generated across these wavebands. However, it is important to note that our visual model is only based on the reflectance properties of *Daphnia*, and omits light transmission through translucent parts of the body; this may also influence the outcome of the present study and should be addressed in further investigations.

The impact of an increased UV chromatic contrast on visual interactions has been demonstrated for the Australian crab spider (*Thomisus spectabilis* Doleschall), where it is used to attract hymenopteran prey by exploiting the prey's UV sensitivity (Heiling, Herberstein & Chittka, 2003; Heiling *et al*., 2005). Furthermore, UV vision in birds has been found to be of importance in bird–fruit interactions, in such a way that the chromatic contrast of fruit signals relative to their visual background is enhanced for a visual system shifted towards the UV part of the spectrum, at least under bright light conditions (Schaefer, Schaefer & Vorobyev, 2007).

Alternatively, our finding that the attractiveness of prey is reduced when UV and long wavelengths are missing could be caused by a decrease in motion perception under these conditions, which is assumed to be strongly dependent on long-wavelength information (Schaerer & Neumeyer, 1996; Krauss & Neumeyer, 2003), but may also be attributed to UV wavelengths (Rubene *et al*., 2010).

It cannot be completely ruled out that the avoidance of prey presented under UV– conditions is caused by a reduced polarization contrast, as UV-polarized light is used for the detection of planktonic prey at least in salmonid fish (e.g. Novales-Flamarique & Browman, 2001). Further experimental work using polarizing filters is required to clarify in what way the UV spectral part is involved in visual foraging in three-spined sticklebacks.

Our results are widely consistent with an analogous experiment on female mating preferences in three-spined sticklebacks, in which blocking the UV and long-wavelength components of male nuptial coloration produced the greatest reduction in male attractiveness (Rick & Bakker, 2008a). Taken together, these results may suggest the existence of a

sensory bias mechanism which is especially affected by visual information in the very short (UV) and very long ('red') parts of the stickleback's visible range. The latter sensory bias has already been assumed for this species (Smith *et al*., 2004). Nevertheless, to obtain more detailed information on a potential UV-related perceptual bias, future work should consider visual foraging preferences for a wider range of prey colours and objects than used in the present study.

In summary, our study suggests that the very short-wave (UV) parts of the stickleback visible spectrum are of relative importance in visual foraging decisions in this species, which is at least relevant for fish from our study population. Whether the effects of the artificially manipulated lighting conditions can also be applied to predator–prey interactions under a range of natural light environments in aquatic habitats requires further investigation.

ACKNOWLEDGEMENTS

We are grateful to Marion Mehlis, Meike Hiermes, Sebastian Baldauf and Timo Thünken for discussions and comments on the manuscript. We thank Marion Mehlis and Meike Hiermes for catching sticklebacks at the field-site.

REFERENCES

Baker KS, Smith RC. 1982. Bio-optical classification and model of natural-waters 2. *Limnology and Oceanography* **27:** 500–509.

Browman HI, Novales-Flamarique I, Hawryshyn CW. 1994. Ultraviolet photoreception contributes to prey search behavior in 2 species of zooplanktivorous fishes. *Journal of Experimental Biology* **186:** 187–198.

Church SC, Bennett ATD, Cuthill IC, Partridge JC. 1998. Ultraviolet cues affect the foraging behaviour of blue tits. *Proceedings of the Royal Society B: Biological Sciences* **265:** 1509–1514.

Confer JL, Howick GL, Corzette MH, Kramer SL, Fitzgibbon S, Landesberg R. 1978. Visual predation by planktivores. *Oikos* **31:** 27–37.

Connaughton VP, Epifanio CE, Thomas R. 1994. Effects of varying irradiance on feeding in larval weakfish (*Cynoscion regalis*). *Journal of Experimental Marine Biology and Ecology* **180:** 151–163.

Cronly-Dillon J, Sharma SC. 1968. Effect of season and sex on the photopic spectral sensitivity of the threespine stickleback. *Journal of Experimental Biology* **49:** 679–687.

Endler JA. 1978. A predator's view of animal color patterns. *Evolutionary Biology* **11:** 319–364.

Endler JA. 1987. Predation, light-intensity and courtship behavior in *Poecilia reticulata* (Pisces, Poeciliidae). *Animal Behaviour* **35:** 1376–1385.

Endler JA. 1997. Light, behavior and conservation of forest-dwelling organisms. In: Clemmons JR, Buchholz R, eds. *Behavioural approaches to conservation in the wild*. Cambridge: Cambridge University Press, 330–356.

Endler JA, Mielke PW. 2005. Comparing entire colour patterns as birds see them. *Biological Journal of the Linnean Society* **86:** 405–431.

Govardovskii VI, Fyhrquist N, Reuter T, Kuzmin DG, Donner K. 2000. In search of the visual pigment template. *Visual Neuroscience* **17:** 509–528.

Guthrie DM, Muntz WRA. 1993. Role of vision in fish behaviour. In: Pitcher TJ, ed. *Behaviour of teleost fishes*, 2nd edn. London: Chapman & Hall, 89–128.

Hart NS, Partridge JC, Bennett ATD, Cuthill IC. 2000. Visual pigments, cone oil droplets and ocular media in four species of estrildid finch. *Journal of Comparative Physiology A* **186:** 681–694.

Hart PJB, Gill AB. 1994. Evolution of foraging behaviour in the threespine stickleback. In: Bell MA, Foster SA, eds. *The evolutionary biology of the threespine stickleback*. Oxford: Oxford University Press, 85–113.

Håstad O, Victorsson J, Ödeen A. 2005. Differences in color vision make passerines less conspicuous in the eyes of their predators. *Proceedings of the National Academy of Sciences* **102:** 6391–6394.

Heiling AM, Cheng K, Chittka L, Goeth A, Herberstein ME. 2005. The role of UV in crab spider signals: effects on perception by prey and predators. *Journal of Experimental Biology* **208:** 3925–3931.

Heiling AM, Herberstein ME, Chittka L. 2003. Pollinator attraction – crab-spiders manipulate flower signals. *Nature* **421:** 334.

Kodric-Brown A, Johnson SC. 2002. Ultraviolet reflectance patterns of male guppies enhance their attractiveness to females. *Animal Behaviour* **63:** 391–396.

Krauss A, Neumeyer C. 2003. Wavelength dependence of the optomotor response in zebrafish (*Danio rerio*). *Vision Research* **43:** 1273–1282.

Leech DM, Johnsen S. 2006. Ultraviolet vision and foraging in juvenile bluegill (*Lepomis macrochirus*). *Canadian Journal of Fisheries and Aquatic Sciences* **63:** 2183–2190.

Loew ER, McFarland WN, Mills EL, Hunter D. 1993. A chromatic action spectrum for planktonic predation by juvenile yellow perch, *Perca flavescens*. *Canadian Journal of Zoology-Revue Canadienne de Zoologie* **71:** 384–386.

Losey GS, Cronin TW, Goldsmith TH, Hyde D, Marshall NJ, McFarland WN. 1999. The UV visual world of fishes: a review. *Journal of Fish Biology* **54:** 921–943.

Lythgoe JN. 1968. Visual pigments and visual range underwater. *Vision Research* **8:** 997–1012.

Lythgoe JN. 1972. The adaptation of visual pigments to the photic environment. In: Dartnall HJA, ed. *Handbook of sensory physiology*. New York: Springer-Verlag, 566–603.

Macy WK, Sutherland SJ, Durbin EG. 1998. Effects of zooplankton size and concentration and light intensity on the feeding behavior of Atlantic mackerel *Scomber scombrus*. *Marine Ecology Progress Series* **172:** 89–100.

Maddocks SA, Church SC, Cuthill IC. 2001. The effects of

the light environment on prey choice by zebra finches. *Journal of Experimental Biology* **204:** 2509–2515.

McDonald CG, Hawryshyn CW. 1995. Intraspecific variation of spectral sensitivity in threespine stickleback (*Gasterosteus aculeatus*) from different photic regimes. *Journal of Comparative Physiology A* **176:** 255–260.

Modarressie R, Bakker TCM. 2007. A limited role for ultraviolet radiation when threespine sticklebacks (*Gasterosteus aculeatus*) prey upon *Daphnia*. *Canadian Journal of Fisheries and Aquatic Sciences* **64:** 1573–1580.

Modarressie R, Rick IP, Bakker TCM. 2006. UV matters in shoaling decisions. *Proceedings of the Royal Society B: Biological Sciences* **273:** 849–854.

Novales-Flamarique I, Browman HI. 2001. Foraging and prey-search behaviour of small juvenile rainbow trout (*Oncorhynchus mykiss*) under polarized light. *Journal of Experimental Biology* **204:** 2415–2422.

Pinheiro JC, Bates D, DebRoy S, Sarkar D. 2009. nlme: linear and nonlinear mixed effects models. *R package* v. 3.1-93.

R-Development-Core-Team. 2010. *R: a language and environment for statistical computing*. Vienna: R Foundation for Statistical Computing.

Rick IP, Bakker TCM. 2008a. Color signaling in conspicuous red sticklebacks: do ultraviolet signals surpass others? *BMC Evolutionary Biology* **8:** 189.

Rick IP, Bakker TCM. 2008b. Males do not see only red: UV wavelengths and male territorial aggression in the threespined stickleback (*Gasterosteus aculeatus*). *Naturwissenschaften* **95:** 631–638.

Rick IP, Bakker TCM. 2008c. UV wavelengths make female three-spined sticklebacks (*Gasterosteus aculeatus*) more attractive for males. *Behavioral Ecology and Sociobiology* **62:** 439–445.

Rick IP, Modarressie R, Bakker TCM. 2004. Male threespined sticklebacks reflect in ultraviolet light. *Behaviour* **141:** 1531–1541.

Rick IP, Modarressie R, Bakker TCM. 2006. UV wavelengths affect female mate choice in three-spined sticklebacks. *Animal Behaviour* **71:** 307–313.

Rowe MP, Baube CL, Loew ER, Phillips JB. 2004. Optimal mechanisms for finding and selecting mates: how threespine stickleback (*Gasterosteus aculeatus*) should encode male throat colors. *Journal of Comparative Physiology A* **190:** 241–256.

Rubene D, Hastad O, Tauson R, Wall H, Odeen A. 2010. The presence of UV wavelengths improves the temporal resolution of the avian visual system. *Journal of Experimental Biology* **213:** 3357–3363.

Ryer CH, Olla BL. 1999. Light-induced changes in the prey consumption and behavior of two juvenile planktivorous fish. *Marine Ecology Progress Series* **181:** 41–51.

Schaefer HM, Schaefer V, Vorobyev M. 2007. Are fruit colors adapted to consumer vision and birds equally efficient in detecting colorful signals? *American Naturalist* **169:** 159–169.

Schaerer S, Neumeyer C. 1996. Motion detection in goldfish investigated with the optomotor response is 'color blind'. *Vision Research* **36:** 4025–4034.

Siitari H, Honkavaara J, Viitala J. 1999. Ultraviolet reflection of berries attracts foraging birds. A laboratory study with redwings (*Turdus iliacus*) and bilberries (*Vaccinium myrtillus*). *Proceedings of the Royal Society B: Biological Sciences* **266:** 2125–2129.

Smith C, Barber I, Wootton RJ, Chittka L. 2004. A receiver bias in the origin of three-spined stickleback mate choice. *Proceedings of the Royal Society B: Biological Sciences* **271:** 949–955.

Viitala J, Korpimaki E, Palokangas P, Koivula M. 1995. Attraction of kestrels to vole scent marks visible in ultraviolet-light. *Nature* **373:** 425–427.

Vorobyev M, Osorio D. 1998. Receptor noise as a determinant of colour thresholds. *Proceedings of the Royal Society B: Biological Sciences* **265:** 351–358.

Vorobyev M, Osorio D, Bennett ATD, Marshall NJ, Cuthill IC. 1998. Tetrachromacy, oil droplets and bird plumage colours. *Journal of Comparative Physiology A* **183:** 621–633.

White EM, Church SC, Willoughby LJ, Hudson SJ, Partridge JC. 2005. Spectral irradiance and foraging efficiency in the guppy, *Poecilia reticulata*. *Animal Behaviour* **69:** 519–527.