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The role of UV in crab spider signals: effects on perception by prey and predators

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Summary

Australian crab spiders *Thomisus spectabilis* sit on the petals of flowers and ambush prey such as honeybees. White-coloured *T. spectabilis* reflect in the UV (UV+ spiders) and previous research has shown that their presence, curiously, attracts honeybees to daisies. We applied an UV-absorber (Parsol®) to create UV-absorbing (UV–) spiders that did not reflect any light below 395 nm wavelength. These physical changes of visual signals generated by crab spiders caused honeybees to avoid flowers with UV– spiders on their petals. They also affected the perception of UV– spiders by honeybees and a potential avian predator (blue tits). Compared to UV+ spiders, UV– spiders produced less excitation of the UV-photoreceptors in honeybees and blue tits, which translated into a reduced UV-receptor contrast and a reduced overall colour contrast between UV– spiders and daisy petals. Our results reveal that a clean physical elimination of reflection in the UV range affects perception in predators and prey and ultimately changes the behaviour of prey.

Key words: *Thomisus spectabilis*, *Apis mellifera*, communication, vision, colour signal, ultraviolet.

Introduction

Sensitivity in the ultraviolet (UV) range of the electromagnetic light spectrum is common in animal visual systems and occurs in all major taxonomic groups (for a review, see Tovée, 1995). Among invertebrates, sensitivity in the UV has been reported for insects (e.g. Briscoe and Chittka, 2001), crustaceans (e.g. Marshall et al., 1996) and spiders (e.g. Walla et al., 1996). UV vision is also widespread in many vertebrate taxa, including fish (Neumeyer, 1998), reptiles (e.g. Fleishman et al., 1993), birds (e.g. Andersson et al., 1998; Cuthill et al., 2000) and, to a lesser extent, in mammals (Jacobs, 1992). Behavioural studies have demonstrated that sensitivity in the UV is often associated with specific behaviours towards UV-reflecting stimuli. Mate choice is often affected by UV-reflecting body parts, such as wing patterns in butterflies (Knüttel and Fiedler, 2001), plumage parts in birds (Cuthill et al., 2000), or UV-reflecting skin in reptiles (Fleishman et al., 1993). In flower-naive honeybees, UV reflectance is only attractive when it is paired with blue reflectance in artificial flowers (Giurfa et al., 1995; Menzel and Shmida, 1993). Moreover, UV light attracts them in the context of a flight response when they attempt to escape into open space (Menzel and Greggers, 1985).

Photospectrometry allows us to measure the reflectance of objects, and advances in neuroethology allow us to calculate the effects of the reflectance on the perceptual system of some receivers (Peitsch et al., 1992; Chittka, 1996; Hart et al., 2000). The UV component in visual signals has increasingly attracted the attention of scientists (e.g. Hunt et al., 2001; Shi and Yokoyama, 2003; Kellie et al., 2004). UV light, however, typically affects more than one receptor type, and thus can have multiple effects on the visual systems of receivers. This arises because the sensitivity spectra of different visual receptors often overlap, and specifically because the sensitivity of long-wavelength receptors extends into the UV (Stavenga et al., 1993). Spectral sensitivity curves need to overlap in order to convey colour information optimally (Chittka, 1996). It is therefore important to analyse the effects of physical changes in the electromagnetic reflectance of signals on every visual receptor of receivers.

Furthermore, visual signals can only be perceived if they are distinguishable from background noise (Chittka et al., 1994; Endler, 1999), and their visibility depends on the ambient light conditions and their contrast against the background colour (Endler, 1991, 1993, 1999; Vorobyev and Osorio, 1998; Fleishman and Persons, 2001; Spaethe et al., 2001; Heindl and Winkler, 2003). Insects, for example, respond to visual signals based on the contrast between an object and the environment, involving all types of photoreceptors (e.g. Briscoe and Chittka, 2001).

A thorough study of visual signals must therefore trace the effects of light reflectance on photoreceptor excitations and calculate the contrast of colour stimuli against background

colour. Several authors have taken this approach, using known values for receptor sensitivity to calculate the relative excitations of different photoreceptors by a colour stimulus and, based on these, the colour contrast between a stimulus and the background (Chittka, 1996, 2001; Endler and Théry, 1996; Andersson et al., 1998; Osorio et al., 1999; Spaethe et al., 2001; Théry and Casas, 2002; Théry et al., 2005). We followed this approach by studying the signalling communication between Australian crab spiders *Thomisus spectabilis* and two types of prey, European honeybees *Apis mellifera* (Heiling et al., 2003) and Australian native bees *Australoplebia australis* (Heiling and Herberstein, 2004). The spiders ambush pollinating insects on flowers, and are visually perceived by bees (Heiling et al., 2003; Heiling and Herberstein, 2004). Honeybees prefer to land on flowers with crab spiders sitting on them rather than unoccupied flowers (Heiling et al., 2003). Australian native bees are also attracted to spider-occupied flowers, but unlike the introduced European bees, do not land on them (Heiling and Herberstein, 2004). We found that, in contrast to European crab spiders (Chittka, 2001; Théry and Casas, 2002; Théry et al., 2005), *T. spectabilis* reflects more light in the UV than the flowers do (Heiling et al., 2003). UV-reflecting white flowers are rare in nature (Chittka et al., 1994) and therefore white, UV reflecting spiders will appear conspicuous on most flowers. They attract honeybees to flowers by creating a pronounced UV contrast and consequently a pronounced overall colour contrast (Heiling et al., 2003). The latter result suggests that the spiders' UV reflection is largely responsible for the bees' attraction to spider-occupied flowers. Here, we test this assumption by removing UV reflection from *T. spectabilis* with an UV-absorbent substance and observing the response of honeybees. We predict that the manipulation will make spider-occupied flowers less attractive to honeybees. Furthermore, we demonstrate how such a manipulation is perceived by the visual system of honeybees and also a potential predator, a passerine insectivorous bird.

Materials and methods

Study animals and collection sites

Crab spiders *Thomisus spectabilis* Dolesch 1858 (Thomisidae) were collected in November 2002 in Brisbane, Australia. The spiders were maintained in the laboratory in plastic cups, water-sprayed daily and fed a weekly diet of live crickets (*Acheta domestica*) and fruit flies (*Drosophila sp.*). They were kept under a 12 h:12 h light:dark cycle with the temperature ranging from 20 to 25°C. Honeybees (*Apis mellifera* L.), a natural prey of *T. spectabilis*, were available from a hive maintained on Macquarie University campus.

Manipulation of spider colour

To investigate whether the UV-reflection of *T. spectabilis* affects the response of honeybees, we applied a mixture of two different UV light-absorbing chromophores on adult female spiders. The chromophores, both common ingredients in sunscreens, were 2-ethylhexyl-*p*-methoxycinnamate (Parsol® MCX), an UV-B light absorber, and 4-tert-butyl-4′-methoxydibenzoylmethane (Parsol® 1789), an UV-A light absorber. The spiders (N=28) were briefly brushed with the mixture. By covering the spiders' body surfaces using Parsol® (DSM, Heerden, The Netherlands), we were able to cut off any reflectance of light below 395 nm (Fig. 1).

Experimental procedure

To investigate whether artificially removing the UV-reflectance of naturally coloured *T. spectabilis* affects the response by honeybees to spiders on flowers, we performed a choice experiment under natural daylight conditions. We offered honeybees the choice between two daisies, one of them occupied by a spider and the other one vacant. Spiders were anaesthetised with carbon dioxide to eliminate any influence of spider behaviour on the choice of honeybees. They were placed onto the petals of a randomly selected daisy, according to the natural position of adult female *T. spectabilis* on radially symmetric flowers (A. M. Heiling, personal observation). We used white daisies (*Chrysanthemum frutescens*, Asteraceae) in our study, as they are a common substrate of *T. spectabilis* (Heiling et al., 2004). The daisies were randomly selected and their petals cut to equalise the diameter to 30 mm. They were placed in black plastic lids (diameter=4 cm) and covered with Glad Wrap™, a clear wrap foil that consists of polypropylene and is permeable to all wavelengths of light above 300 nm, with less than 5% attenuation. Glad Wrap™ (The Clorox Company, Oakland, CA, USA) removes olfactory cues emanating from the flower. In previous studies in which honeybees were presented with a choice of flowers covered by Glad Wrap™ (e.g. Heiling et al., 2004), the bees still landed readily on a covered flower. We covered the daisies and spiders, as flower odours (Heiling et al., 2004) and possibly the smell of Parsol® affect honeybee choice. Each pair of lids containing the flowers was placed horizontally on a rectangle (18 cm×13 cm) of black cardboard, with a distance of 8 cm between the flower centres. This arrangement replaced a feeding station for honeybees, offering 25% sucrose solution. We recorded the first visit of a honeybee on either of the two flowers and then removed the bee from the population. As the

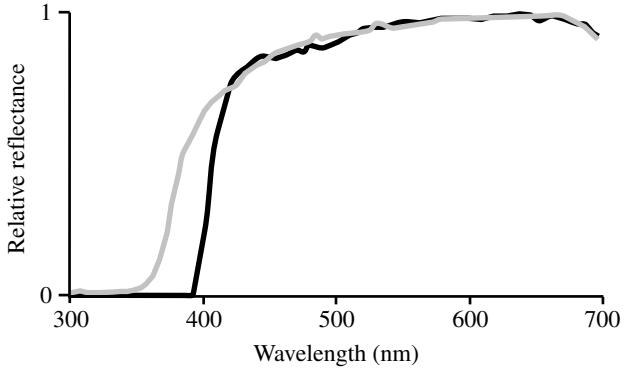

Fig. 1. Relative reflectance of manipulated UV-absorbing spiders (black curve, N=28) and naturally white spiders (grey curve, N=25; data taken from Heiling et al., 2003).

size of a spider might affect its signalling impact on a flower, we weighed each individual to the nearest 0.1 mg.

Colour analyses of T. spectabilis *and* C. frutescens

We measured spider and flower reflectance using a USB 2000 spectrometer with a PX-2-pulsed xenon light source attached to a PC running OODBase32 software (Ocean Optics Inc., Dunedin, FL, USA). The measurements covered the range from 300 nm to 700 nm. Each spider and flower was measured six times and the median value taken for further calculations. We calculated the relative receptor excitation values (E) for the different types of photoreceptors of honeybees, which have peak sensitivities in the UV, the blue and the green (for methods, see Chittka, 1996; Briscoe and Chittka, 2001). Receptor voltage signals E were also calculated for passerine insectivorous birds (blue tits), which have tetrachromatic vision, with their receptor sensitivities peaking in the UV (UVS), blue (SWS), green (MWS) and red (LWS; Hart, 2001).

We included blue tits as a model for avian predators, even though this particular species is not a natural predator of *T. spectabilis*. The spiders are often predated upon by other species of passerine songbirds, such as noisy miners *Manorina melanocephala* (A. M. Heiling, personal observation), but the receptor sensitivities of these have not been studied. All passeriform birds studied so far possess a tetrachromatic set of cones, with little interspecific variation in the tuning of photopigments (Bowmaker et al., 1997; Cuthill et al., 2000; Hart, 2001). Among 12 different passerines studied, for example, the wavelengths of maximum absorbance ranged from only 355-380 nm for the UV pigment, 440–454 nm for the short-wave pigment, 497–504 nm for the medium-wave pigment, and 557–567 for the long-wave pigment (summarised in Hart, 2001). The blue tit thus serves as a typical example of a passerine predator of crab spiders.

The calculations of E-values generate the proportion of the maximum potential excitation in each receptor type. Based on the E-values, we determined the colour loci in the hexagon colour space of honeybees (Chittka et al., 1994) and of blue tits (a tetrahedron; Goldsmith, 1990). For honeybee vision, we illustrated the colour space, which is based on two colour opponent processes (Backhaus, 1991) and shows how the colour of the spiders and flowers is perceived (Chittka, 1996). Specifically, a colour's angular position in the colour hexagon indicates a bee-subjective hue, while increasing distance from the centre of the hexagon indicates increasing spectral purity or saturation.

We used the colour coordinates in the colour spaces of honeybees and blue tits to calculate the Euclidean distances. These calculations were performed for each spider-flower combination used in the experiments. Euclidean distance in the colour hexagon is correlated with the colour contrast as perceived by the bee receiver of visual signals (Chittka, 1996; Théry et al., 2005). This approach takes into consideration the colour opponent processes that influence how the brain integrates a colour signal (Chittka, 1996).

The identification of UV, blue and green through a bee's eye relies on different neuronal channels (Giurfa and Lehrer, 2001). An object seen at an area subtending at least 5° (and no more than 15°) is perceived by the green receptor of bees (Giurfa and Lehrer, 2001; Spaethe et al., 2001). For a bee to perceive signals using all three spectral receptor types, the stimulus must subtend an area of at least 15°, which corresponds to 59 ommatidia of its compound eye. Hence, compared to green contrast, colour contrast is perceived from a shorter distance to an object. Moreover, the sensitivity of bees in the UV is 16 times higher compared to the sensitivity in the blue and in the green (Helversen, 1972). The sensitivities of photoreceptors are adjusted to the quantity of light reflected from the predominant background. Due to the low reflectance of UV from green foliage background (Chittka et al., 1994), the UV receptor is relatively more sensitive. Similarly, the sensitivities of the four cone types of passerine birds (UVS, SWS, MWS and LWS) peak in different regions of the light spectrum, with a combination of MWS and LWS receptors (double cones) used for detecting achromatic contrast between objects and all four types of cones responsible for the detection of colour contrast (e.g. Hart et al., 2000). For these reasons, we compared not only the overall contrast between spiders and daisies from the view of honeybees and blue tits, but also the specific contrasts for the different receptor types.

We used exact binomial P-tests to assess honeybee choice between flowers occupied by UV– spiders or UV+ spiders *vs* vacant flowers. Independent t-tests were used to compare spider mass, E-values and contrasts between UV– spiders and UV+ spiders. The t-tests considered the variances within groups, and in the case of unequal variances, we used independent t-tests that output fractional degrees of freedom (for methods, see Satterthwaite, 1946). Furthermore, we used Mann–Whitney U-tests to compare colour contrasts between spider-flower combinations that were chosen or rejected by honeybees.

Results

White spiders treated with UV-absorbing Parsol® (UV– spiders) did not differ significantly in mass from naturally white spiders (UV+ spiders) used in a former study that tested the effect of the presence of a spider on honeybees' choice of flowers (Heiling et al., 2003; mean mass ± S.D. = 0.122±0.029 g, N=28 and 0.133±0.039 g, N=25, respectively; t_{51}=–1.113, P=0.271).

Effect of a spider's presence on the choice of honeybees

The presence of both UV– spiders and UV+ spiders clearly affected the response of European honeybees, but in different ways. While the presence of UV+ spiders attracted honeybees to flowers (Heiling et al., 2003; Fig. 2), UV– spiders deterred them. When given the choice between a daisy occupied by a UV– spider and a vacant daisy, bees clearly preferred the vacant daisy over the spider occupied one (exact binomial P=0.0257, N=28; Fig. 2).

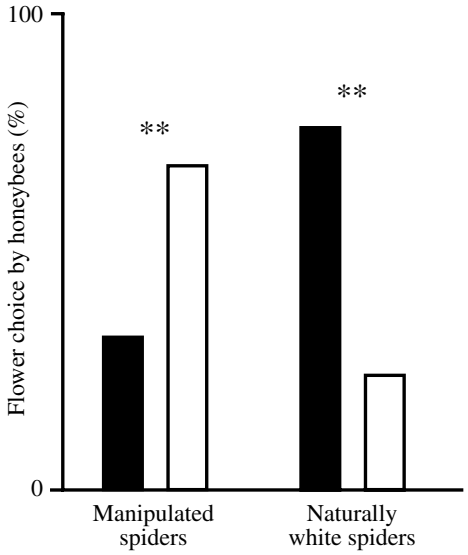

Fig. 2. The responses of honeybees when presented with a choice between a flower occupied by a manipulated UV-absorbing crab spider and a vacant flower (left; *N*=28) and a choice between a flower occupied by a naturally white crab spider and a vacant flower (right; *N*=25; data taken from Heiling et al., 2003). The data show the percentage of times that honeybees first landed on the spider-occupied flower (black bars) or first landed on the vacant flower (white bars). Each bee was tested only once. **P<0.01.

UV-absorbing and naturally white spiders from the view of honeybees and blue tits

Compared to UV+ spiders, UV– spiders reflected less light in the UV, but surprisingly more light above 400 nm (Fig. 1). Plotting spider and flower colours in bee colour space revealed that the colour of UV+ spiders was more distinct from the colour of daisy petals than the colour of UV– spiders (Fig. 3). The receptor excitation values for UV– spiders were significantly lower than those for UV+ spiders in the UV, but higher in the blue and in the green range of the spectrum (Table 1). Moreover, the drop in the excitation of UV receptors for UV– spiders was much larger than the increase on the excitation of blue and green receptors (Table 1). Similarly, UV+ spiders and UV– spiders differed in the maximum potential excitation of blue tit receptor cones, with *E*-values being significantly lower in the UV, but higher in the blue, in the green and the red (Table 1), and again, the drop in the excitation of UV receptors was much larger than the increase in excitation of the other three receptors (Table 1).

The colour of UV+ spiders (described in Heiling et al., 2003; *N*=25) and UV– spiders (*N*=28) generated different contrasts against the petals of white daisies (Fig. 4). For honeybees, the UV– spiders against the white petals of daisies created a lower contrast in the UV ($t_{31.12}$=9.746, P<0.001). However, there were no differences in blue contrast and green contrast between UV+ spiders and daisies and UV– spiders and daisies (t_{51}=0.38, P=0.722 and t_{51}=0.362, P=0.719, respectively; Fig. 4).

The overall colour contrast, which incorporates the entire spectrum visible to bees, was lower between UV– spiders and daisies than between UV+ spiders and daisy petals ($t_{37.19}$=–14.141, P<0.001; Fig. 4; see also Heiling et al., 2005). Although the colour space (Fig. 3) indicates similarity of UV– spiders and daisies, the colour contrast generated by UV– spiders was still pronounced and lay well above the detection threshold of 0.01 (Dyer and Chittka, 2004).

Furthermore, in both choice experiments using UV+ spiders and UV– spiders, there was no difference in colour contrast between spider–flower combinations that were chosen and those that were rejected by honeybees (Median ± Qi, Qs=0.152±0.126, 0.177, Mann–Whitney U=54, P=0.849, N=25 and Median ± Qi, Qs=0.0363±0.0197, 0.0472, Mann–Whitney U=67, P=0.27, N=28).

From the view of blue tits, UV– spiders created a lower

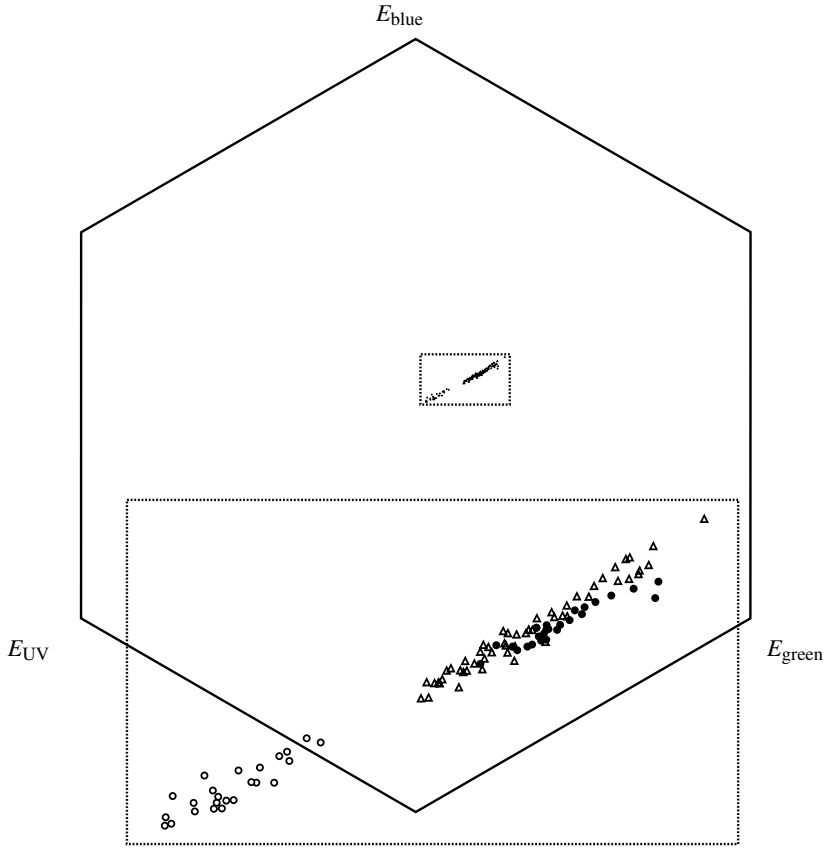

Fig. 3. Illustration of spider- and flower colour in the colour hexagon of honeybees, calculated for UV-absorbing spiders (black circles, *N*=28), natural spiders (white circles, *N*=25; data for calculation taken from Heiling et al., 2003), and the petals of daisies (white triangles, *N*=53). The small window represents the actual position of colours in the hexagon, shown in detail in the enlarged window.

contrast in the UV receptor than UV+ spiders (t_{51}=–10.701, P<0.001), but a similar contrast in the blue ($t_{48.34}$=0.178, P=0.86), green ($t_{46.71}$=0.276, P=0.784), and red receptors ($t_{44.61}$=0.775, P=0.443; Fig. 4). This combination of spiders and daisies also generated a lower overall colour contrast (t_{51}=12.528, P<0.001; Fig. 4). While the UVS-contrast and the overall colour contrast created by UV– spiders on daisies just reached the detection threshold of birds (0.06; Théry and Casas, 2005; Fig. 4), UV+ spiders were well distinguishable from daisy petals by UVS contrast and colour contrast (Fig. 4). The contrasts between both UV+ and UV– spiders and daisies in the blue, the green, and the red were far below the detection threshold of birds (Fig. 4).

Discussion

By treating the body surface of naturally UV-reflecting Australian crab spiders *Thomisus spectabilis* (UV+) with Parsol®, we generated UV-absorbing spiders (UV–). Physically, this resulted in a complete removal of reflection below 395 nm, while leaving increasing reflection of wavelengths above 400 nm. The difference in colour between UV+ spiders and UV– spiders translated into a different response by honeybees, a natural prey of crab spiders. While the presence of UV+ spiders attracted honeybees to the flowers (Heiling et al., 2003), UV– spiders deterred them (present results). Thus it appears that the UV component of this Australian crab spider is crucial in attracting honeybees.

Not surprisingly, our comparison of reflectance data between UV+ spiders and UV– spiders revealed different excitation values in the UV for both the honeybee (E_{UV}) and the bird visual system (E_{UVS}), with UV+ spiders inducing a stronger response by the UV-photoreceptors than UV– spiders. However, our analyses of the visual appearance of crab spiders revealed that UV+ spiders and UV– spiders are not equally perceived beyond the UV spectrum by both honeybees and blue tits. UV+ spiders and UV– spiders also differed in the excitations of the green, the blue and, in the case of birds, the red receptors. For both honeybees and birds, UV+ spiders caused significantly lower excitations of these receptor types than UV– spiders did. To explain these results we performed additional reflectance measurements on five *T. spectabilis*, which revealed an average reflectance of 76% above 400nm before and after the treatment with Parsol®. We found that the increased reflectance of UV– spiders above 400 nm was not caused by the application of Parsol®. Instead, it might have been caused by the housing conditions of spiders. The spiders were fed a diet of crickets and fruit flies, which may have affected their colouration. Moreover, UV+ spiders were kept in the laboratory for a longer period of time than UV– spiders. This means that, at the time of experimentation, UV+ spiders were older than UV– spiders, which might have also affected the reflectance properties of spiders. However, the critical values that bees and birds use to distinguish signals are not the receptor excitation values *per se*, but the colour contrast of the signal against its natural background. Here, our results show no difference between UV+ spiders and UV– spiders in contrast other than in the UV. Thus, we are confident that despite natural noise in spider colour, our treatment with Parsol® only manipulated UV-reflection and that honeybees reacted to this manipulation only.

The elimination of UV-reflection from visual signals in other

Table 1. *Receptor excitation values, calculated for UV-absorbing spiders (UV–) and naturally coloured spiders (UV+) from the view of honeybees and blue tits*

Receptor type	Spiders		Statistics
	UV– spiders	UV+ spiders	
Honeybee system			
UV	0.637±0.004	0.763±0.005	t_{51}=18.731, P<0.001
Blue	0.876±0.001	0.868±0.003	$t_{37.76}$=–2.594, P=0.013
Green	0.841±0.001	0.823±0.003	$t_{33.86}$=–5.265, P<0.001
Bird system			
UVS	0.655±0.007	0.817±0.005	$t_{44.07}$=18.434, P<0.001
SWS	0.882±0.001	0.869±0.002	$t_{36.94}$=–4.538, P<0.001
MWS	0.824±0.001	0.803±0.003	$t_{33.31}$=–5.958, P<0.001
LWS	0.858±0.001	0.841±0.003	$t_{32.09}$=–5.341, P=0.001

Values are means ± S.D.; N=28 (UV–), N=25 (UV+); UV+ data taken from Heiling et al. (2003).

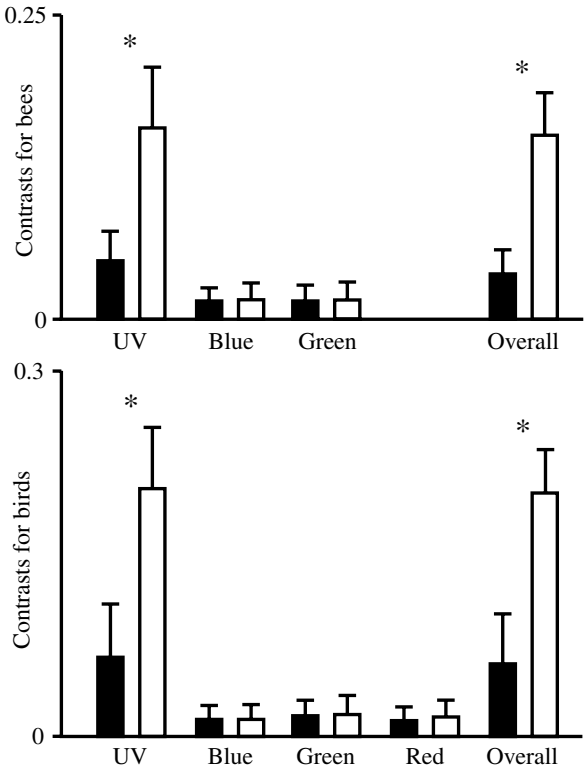

Fig. 4. Visual contrasts generated by UV-absorbing *T. spectabilis* (black bars, N=28) and natural *T. spectabilis* (white bars, N=25; calculation based on data taken from Heiling et al., 2003) on daisy petals from the view of honeybees and blue tits. Values are means ± S.D.; only one S.D. bar is drawn to simplify the graph. *P<0.001.

systems such as in the social interactions in birds affected the behaviour during female–male or male–male interactions (Andersson and Amundsen, 1997; Sheldon et al., 1999; Alonso-Alvarez et al., 2004). Like our results, these studies on birds provide strong evidence that the removal of the UV component from the signal (male plumage) causes the observed changes in behaviour. Our additional photospectrometric analyses and calculations of receptor excitations, however, reveal the aspects of the perceptual changes that may be responsible for these effects.

Different visual systems will not perceive and process the colour of an object equally, if different types of photoreceptors with different sensitivities are involved in colour vision (e.g. Endler, 1990). For example, flowers that reflect in the UV and in the red range of the electromagnetic spectrum, will appear ultraviolet to a UV-blue-green-trichromatic bee and red to our blue-green-red-trichromatic visual system (Chittka et al., 1994). In the visual systems of honeybees and blue tits, the peak spectral absorbance of their photoreceptors lies in different regions of the spectrum. For example, honeybees are maximally sensitive to UV at 344 nm (Menzel and Backhaus, 1991), while the UVS cone of blue tits is maximally sensitive at 375 nm (Hart, 2001). Similarly, the sensitivities of the other photoreceptor types of honeybees and blue tits peak in different regions of the light spectrum (Menzel and Backhaus, 1991; Hart, 2001). Consequently, object colours will not equally excite the photoreceptors in different visual systems.

Receptor excitation values take into account the photoreceptor transduction process, or how the electromagnetic reflectance of an object translates to neural firing. Coloured objects such as spiders in our case, however, become visual signals only in combination with their natural backgrounds, against which they generate a colour contrast for the perceiver (Spaethe et al., 2001; Heindl and Winkler, 2003). For both bees and birds, the UV contrast between spiders and daisy petals was significantly higher in the natural UV+ spiders than in the UV– spiders. Because the contrast between spiders and daisies was similar for all the other receptors, this translated into a higher colour contrast generated by UV+ spiders on daisy petals. However, the average UV contrast between UV– spiders and daisy petals was also well above the detection threshold of the honeybee and bird receivers. This detectability is due to two characteristics of photoreceptors. First, honeybees and most birds are more sensitive in the ultraviolet than in other spectral ranges (Helversen, 1972; Maier, 1992). Second, each type of photoreceptor is sensitive across a wide range of wavelengths, forming roughly a Gaussian function (Stavenga et al., 1993). To give an example, the sensitivity of the honeybee UV-receptor reaches its maximum at 344 nm. However, the sensitivity of the same receptor type, if normalised to a maximum of 1, is still around 0.14 at 405 nm, which falls into the violet range of the light spectrum (Chittka, 1996). This explains why the elimination of a certain range of wavelengths from the colour of an object affects not only the excitation of one type of photoreceptors involved in colour vision.

Why did UV– spiders repel honeybees, when they still had on average positive UV-contrast with the daisy petals? Fig. 3 shows that the UV excitation of UV– spiders is within the natural range found in daisies. In fact, some UV– spiders excited UV-receptors less than some daisies. An UV– spider on a daisy could alter the radial symmetry of the flower and this chromatic asymmetry may indicate a deteriorating flower to bees, and hence repel them. UV+ spiders, on the other hand, far exceed the daisies in reflecting UV light and exciting the UV receptors (see Fig. 3). This clear signal obviously results in added attraction for the bees. Further research, however, is needed to confirm any interpretation of the differences in the behavioural effects of UV+ and UV– spiders.

In sum, our results provide evidence that the reflection of light in the UV range by UV+ *T. spectabilis* functions to attract honeybees. But it remains uncertain from the spectrometric analysis which components in the perception of the visual signal function to deceive prey. Removing the UV reflectance from spiders translated into a lower UV contrast and a lower overall contrast between spiders and daisies. In conclusion, assigning a change in behaviour to the change in UV-reflection alone may not be straightforward. It is likely that the differences in UV-receptor signals between UV+ and UV– spiders generated a behavioural effect, since the effect of the differences in UV-receptor signals on colour contrast is pronounced.

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